

# AGRICULTURE, FOREST AND AQUATIC SCIENCES IN THEORY AND PRACTICE

Editörler

Assoc. Prof. Dr. Vedat ÇAVUŞ

Asst. Prof. Dr. Senem GÜNEŞ ŞEN



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## Chapter 1

### Sustainability of Wood Material Treated With Fire Retardants

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The popularity of wood as a material, whose historical past is changing day by day and which is known to be used in many goods and structures, continues today thanks to some superior characteristics. Natural and healthy, light and robust, high energy efficiency, easy to transport and assemble, aesthetic and decorative, good thermal insulation, resistant to chemicals, economical and sustainable, etc. can be mentioned as examples of the superior properties of wood material (Örs et al., 1999; Bal and Bektaş, 2018).

Sustainability was developed in the Report of the World Environment Conference held in Stockholm in 1972 depending on the discussions within the framework of the concept of eco-development. It was used for the first time in 1987 with the report prepared by the World Commission on Environment and Development (Çetin, 2010). The concept of sustainability is discussed in many environmental, social, economic and cultural dimensions (Brundland R., 1987).

With the technical and technological development that gained momentum after the Industrial Revolution, there has been an eye-catching change in the construction sector. In addition, with the energy crisis in the 1970s, it became clear that one day resources could be depleted. In the following years, it was realized that measures such as environmental pollution, ecology, climate change, etc. should be taken to leave the world as "livable" to future generations. The effect of sustainability in the following stages was seen in the design and production methods of buildings (Çelebi, 2003).

In addition to protecting human health and comfort, sustainable design should support and protect cultural structure, lifestyle and comfort. We can summarize sustainability as taking into account the needs of future generations, not wasting our resources, evaluating waste, not damaging the environment, leaving a livable world for future generations (Aydın and Alemdağ, 2014).

Today, thanks to technological developments, many materials serve the purpose of production in the best possible way, but as their use increases, the decrease in the possible sources of some substances is an important threat to the future (Graedel et al., 2015). Sustainability is positioned at an important point between factors such as production and design studies and environmental problems (Ekşi and Çırak, 2017). The ecological system, process and productivity that exist today are the main elements of sustainability (Yavuz, 2010) and as a key concept, it is in relationship with many disciplines (Güner et al., 2017). It is necessary to evaluate sustainable development in terms of economy, use more in the production process of renewable resources and be responsible for the environmental effects of production activities on the environment (Yavuz, 2010). By preventing environmental problems, the total life span of the products will increase and reduce their costs, thus ensuring sustainability (Ekşi and Çırak,

2017). In this context, it can be said that sustainability is a goal that can be achieved by allowing renewable natural resources to themselves (Yavuz, 2010).

The choice of materials and raw materials are the most important criteria. The crisis of the existence of natural resources, which are already limited, can be prevented by the reuse of materials. This may bring more financial burden, but the important thing is that we need these reserves in the future, and also that this material that is not used will become waste, which will lead to environmental pollution (Pearson, 1998). The negative effects of the artificial environments created on the ecosystem over time force architects to take into account how the environment will be designed and how the buildings will be produced, and lead them to reconsider the designs that cause the least damage to the natural cycle (Gezer, 2013).

Wood and stone, which are very old building materials in human history, have been used as carrier elements, flooring, roof elements and exterior cladding in buildings for thousands of years and continue to be used. These materials have always continued to exist in the field of architecture and design despite the changing materials, construction systems and technologies from the past to the present (Aydın et al., 2014).

The re-evaluation of wood materials preserved from the wreckage of historical wooden buildings in the sustainable construction and furniture sector is important in reducing carbon footprint, saving energy, protecting natural resources and achieving environmental goals such as climate change (URL 1, 2024). However, the fact that the wood material is not resistant to adverse weather conditions and biological effects, especially its low fire resistance, still makes people nervous. Fortunately, today, the life of the treated wood material is increasing with different modifications, impregnations and protective layers newly developed. In archaeological studies, it has been determined that fire retardants, various varnishes and paints (Eickner, 1966, Ellis and Rowell, 1989) have been applied to wood materials and have been preserved until today.

In some research, the Greeks used seawater, the Egyptians used aluminium solutions, and the Romans used clay, lime, slime (Bozkurt, 1997), alum and vinegar (Eickner, 1966) as fire retardants. The impregnation of wood materials with chemicals is important in order to increase their resistance to combustion in many places of use (Ellis and Rowell, 1989).

Wood material is a sustainable building material with low carbon emissions produced with natural, edible, low energy (Binggeli, 2008). It is stated that the amount of carbon released from building materials in wood production is 15 kg/m<sup>3</sup>, 120 kg/m<sup>3</sup> in concrete production, 5320 kg/m<sup>3</sup> in steel production and 22000 kg/m<sup>3</sup> in aluminium production. The sustainability of wood material can

be ensured through practices such as correct afforestation studies, conscious tree cutting, utilization of wastes in composite material production and use of alternative wood sources (Çelebi, 2003; Binggeli, 2008).

Combustion is one of the most important disadvantages of wood materials (Istek et al., 2017). In the first phase of the wood combustion process, the wood is exposed to heat and water vapour is released (White and Dietenberger, 2010). In the second stage, cellulose, hemicellulose and lignin, which are the components of wood, are broken down by heat and their thermal degradation begins (Babrauskas, 2003) and flammable gases are released. In the third stage, these gases catch fire and combustion continues. Finally, at the end of the combustion process, charred residues and ash are formed. During these phases, the rate of wood combustion and the rate of flame propagation vary depending on the type of wood, its density, and its moisture content (White and Dietenberger, 2010).

Various methods are used to increase the combustion resistance of wood materials. One of these methods is to apply fire-retardant chemicals to the wood. Fire retardant chemicals ensure fire safety by increasing the resistance of wood to heat and slowing down the combustion process. These chemicals can be applied to the surface or internal structure of the wood and change the combustion properties of the wood through various chemical reactions (LeVan and Winandy, 1990).

There are changes in some physical and mechanical properties of the tree material treated with fire retardant chemicals (Demir and Aydın, 2016). In general, fire retardants should reduce the flammability of wood material, prevent the spread of flame between surfaces, and reduce the rate of degradation and charring. When the heat source is removed, the wood should stop the combustion of the material and prevent ignition (Küçükosmanoğlu, 1993).

Fire retardant chemicals both slow down the heat breakdown process of wood components and increase fire safety by reducing the formation of flammable gases. In this process, phosphates, borates and halogenated compounds from fire retardants (LeVan and Winandy, 1990), and finishing materials (Budakçı et al., 2016) are widely applied to wood material by different methods. These substances increase the carbonization of wood during combustion and slow down the spread of flames by absorbing heat. However, the effects of these chemicals can vary depending on the type of wood, method of application, and conditions of use. Fire retardants such as phosphates, borates and halogenated compounds increase the carbonization of wood during combustion and reduce the formation of flammable gases. Phosphates provide resistance to heat by forming a protective layer on the surface of the wood. Mixtures such as borates (Baysal et al., 2006)

and Firetex (Kesik et al., 2016) slow down the thermal degradation of wood and reduce the formation of flammable gases (LeVan and Winandy, 1990; Kartal and Green, 2003) and reduce the risk of fire. Halogenated compounds, on the other hand, inhibit combustion reactions by forming free radicals and stop the combustion process (Babrauskas, 2003).

If the wood material is to be used in outdoor conditions, fire retardant chemicals may lose their effect due to the effect of precipitation. The effect of fire retardants may vary depending on the method of application (Örs et al., 1999, Kesik et al. 2024). It has been reported that the amount of absorption of fire retardant chemicals will increase depending on the immersion time in the immersion method (Demir and Aydın, 2021) and that it will give more positive results in the pressure method than the immersion method (Uysal, 1997). They determined that the weight loss (31.37%) was less than 1/3 of the control samples and the CO gas emission (80.17 ppm) was less than half of the control samples in Scotch pine wood where long-term immersion method was applied with fire retardant solutions (Kesik et al., 2015).

The application of fire-retardant protective layers can also increase the fire resistance of wood material (Östman and Tsantaridis, 2016). Today, in addition to timber, contra, composite and particle boards impregnated with fire-retardant chemicals, new ones are added to the market every day by adding new ones to those whose surfaces are coated with fire-retardant paints (Östman, et al., 2017). As industrialization and technological developments increase; The search for environmentally friendly, non-toxic, high temperature and flame resistant, functional materials with wide applicability is also increasing (Aydın et al., 2016).

Processing wood material, which is one of the limited natural resources, with environmentally friendly protectors will contribute to the maximum level of economic life and thus to its sustainability. Increasing the fire resistance of wood material is of great importance, especially in terms of fire safety. Therefore, researching and developing the effects of fire retardant chemicals on wood materials, improving fire safety standards (Östman et al., 2017), and making wooden structures safer play a critical role in terms of sustainability. In this study, the single flame-induced combustion resistance of Scotch pine wood material protected by water-based fire retardants will be determined.

The experimental method used to determine the resistance of single flame-induced combustion resistance, the combustion values and findings of wood materials treated with impregnation, fire retardant solution and water-based paints are given below, respectively.

In this study, Scotch pine (*Pinus sylvestris* L.) wood, which is widely used in the woodworking and construction industry, was preferred as an experimental sample. In this study, solution (Hemel FR), impregnation (Hemel Prime), green water-based paint (Y-D 45) and black water-based paint (S-D 45) were used as fire retardants. Fire retardant solutions were procured from enterprises in Ankara, and water-based paints were supplied from Kimetsan. Test specimens treated with impregnated Hemel Prime, fire retardants Hemel FR, Kimetsan Black-D45 and Kimetsan Green-D45 were exposed to single flame-induced combustion and the combustion zone was measured.

Statistical analyses were performed using the IBM SPSS 27 program using the data obtained from the experimental samples. Single analysis of variance (ANOVA) was performed to examine the data obtained, the Duncan test was performed to compare the variables considered important in the analysis of variance with each other and homogeneity groups were determined.

The properties of fire retardant solutions are taken from the company websites (URL2, 2024) and the paint properties are taken from the manufacturer (D45 MSDS, 2015) and are given in Table 1.

Table 1. Features of fire retardant solutions, impregnations and paints

Fire retardants	Hemel FR	Hemel Prime	Kimetsan Y-D45	Kimetsan S-D45
Density (g/cm <sup>3</sup> )	1,09 ± 0,02	1 ± 0,02	1,17	1,23
Amount of solids	11,8	14,3	23,7	24,5
VOC (g/L)	10	0	<50	<50
Shelf Life (5-35°C)	2 years	2 years	2 years	2 years
Covering Capacity (m <sup>2</sup> /L)	Single layer 4-5	4-5	8-10	8-10
Drying Time (20°C, %40-65 Relative humidity)	24 hours	24 hours	24 hours	24 hours
Colour	Colourless	Brown	Light green	Black

The test specimens were prepared in 8x32x1 cm dimensions and kept in the air conditioning cabinet with a temperature of 20±2°C and a relative humidity of 65±3% according to TS 2471 until they reached a constant weight. Fire retardant Hemel FR+Kimetsan D45, Hemel Prime+Kimetsan D45 and Kimetsan D45 brushes were applied to the surfaces of the test samples in three coats.

In the single flame combustion test, the burner tip angle from which the flame comes out is fixed to 45o, and the test is placed directly opposite the sample surfaces. After fixing the flame length of 20 mm and the distance between the flame and the test samples of 2 mm, the test samples were left to be exposed to flame for 60 seconds (Figure 1) (TS EN ISO 11925-2).



Figure 1. Combustion assembly

After the combustion period was completed, the combustion continued on the surfaces of a few of the experimental specimens from which the flame source was drawn, while the combustion did not continue in the others. At the end of the test, the burn zones on the samples were visually examined and the dimensions of the burn marks were measured and recorded separately. The area where the flame affected in the first degree (burning zone) was burned and blackened, and pit formation and cracks occurred in the combustion zone of some samples.

Experimental samples exposed to flame for 60 seconds; treated with Kimetsan Green-D45 (Figure 2a), treated with Hemel FR+Kimetsan Green-D45 (Figure 2b), treated with Hemel Prime+Kimetsan Green-D45 (Figure 2c), treated with Hemel Prime+Hemel FR+Kimetsan Green-D45 (Figure 2d); is given.

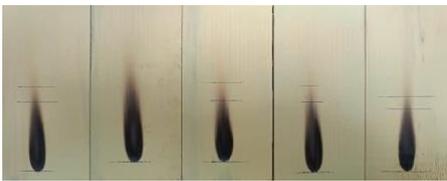


Figure 2a. Kimetsan Green-D45



Figure 2b. Hemel FR+Kimetsan Green-D45

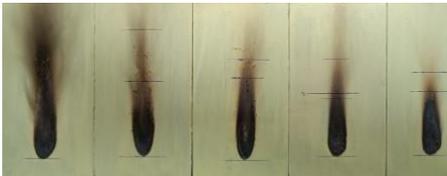


Figure 2c. Hemel Prime+Kimetsan  
Green-D45



Figure 2d. Hemel FR+Hemel  
Prime+Kimetsan Green-D45

Experimental samples exposed to flame for 60 seconds; treated with Kimetsan Black-D45 (Figure 3a), treated with Hemel FR+Kimetsan Black-D45 (Figure 3b), treated with Hemel Prime+Kimetsan Black-D45 (Figure 3c), treated with Hemel Prime+Hemel FR+Kimetsan Black-D45 (Figure 3d); is given.



Figure 3a. Kimetsan Black-D45



Figure 3b. Hemel FR+Kimetsan Green-D45

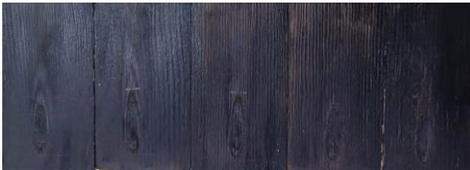


Figure 3c. Hemel Prime+Kimetsan Green-D45



Figure 3d. Hemel FR+Hemel Prime+Kimetsan Green-D45

The analysis of the variance of the dimensions of the burn marks on the surfaces of the experimental specimens left to be exposed to flame for 60 seconds is given in Table 2.

Table 2. Analysis of variance of combustion trace dimensions in experimental samples

ANOVA					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	5393,375	7	770,482	15,310	0,000*
Within Groups	1610,400	32	50,325		
Total	7003,775	39			

\*: Meaningful (according to  $\alpha < 0.05$ )

According to Table 2, statistically significant differences were found in fire retardants ( $p \leq 0.05$ ). The results of the comparison of Duncan test burn trace values at the level of fire retardant treatment type are given in Table 3.

Table 3. Duncan test at the level of fire retardant treatment type

Fire retardants	$\bar{x}$	SD	HG
Kimetsan Green-D45	55,60	3,78	BC
Kimetsan Black-D45	68,20	5,40	D
Hemel FR+Kimetsan Green-D45	39,80	5,63	A*
Hemel FR+Kimetsan Black-D45	53,80	6,42	B

Hemel Prime+Kimetsan Green-D45	64,00	13,21	BD
Hemel Prime+Kimetsan Black-D45	78,60	8,02	E
Hemel FR+Hemel Prime+Kimetsan Green-D45	66,80	4,60	D
Hemel Prime+Hemel FR+Kimetsan Siyah-D45	73,80	5,12	DE

$\bar{X}$  : Arithmetic average, HG: Homogeneous groups, SD: Std. Deviation

According to Table 3, in the experimental samples exposed to the combustion effect caused by a single flame for 60 seconds; It was the lowest in those treated with Hemel FR+Kimetsan Green-D45 (39.8mm) and the highest in those treated with Hemel Prime+Kimetsan Black-D45 (78.6mm); Combustion trace values were determined. The low burn scar length in the burning areas indicates that the fire retardants used are successful.

In this study, the fire resistance properties of Scotch pine wood materials exposed to the combustion effect caused by a single flame vary with the effect of impregnating agent (Hemel Prime), different fire-retardant solutions (Hemel FR) and paints (Kimetsan Y-D45, Kimetsan S-D45). In the study, it was thought that the combustion resistance properties would increase by applying the impregnating agent and fire-retardant solution to the sample surfaces consecutively and then coating them with fire retardant water-based paints. However, the opposite situation was determined in solution applications made after impregnation. This may be because the impregnating agent applied before does not accept much of the solution, as it fills the surface of the tree. Applying fire retardants with a brush can be effective in this outcome. In the literature (Kesik et al., 2024), they reported that when different fire retardants were applied to wood material consecutively, they increased the combustion resistance more than the control samples. This view supports the results of the study.

Fire retardant paint applications were made with brushes, and paint covering was provided especially in third coat applications on Hemel FR. It is thought that applications with spray guns will be easier. In the study, it was determined that the application of Hemel FR+Kimetsan Yeşil-D45 increased the fire resistance and gave the most successful result. It is thought that the application of fire retardants such as Hemel Prime and Hemel FR with different application methods will penetrate deep into the wood material and increase fire resistance. The literature (Yalınkılıç, 1992; Uysal et al., 2000) supports our view by stating that impregnation with immersion and pressure method will give positive results. Users may be advised to choose one of the immersion or pressure methods in fire retardant applications. In future studies, CO measurements can be made in wood materials with fire retardant and its impact on the environment can be discussed.

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## **Chapter 2**

### **Organic Fertilizer Use in Crop Production**

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## **INTRODUCTION**

It is known that it is necessary to increase agricultural production due to the increasing world population. The interest in researches for the solution of the problems experienced in plant production stages is increasing day by day. The number of studies on plant nutrition and fertilization, which is one of the problems encountered in crop production, is increasing day by day.

Many practices used in crop production can reduce the amount of organic matter in soils, leading to a decrease in soil fertility (Aytenuw and Bore 2020; Gerke 2022; Chaudhary et al. 2023). In addition, changes in crop production techniques, especially the use of water on non-irrigated land, can increase crop yields (Kaduyu and Musinguzi 2021; Li et al. 2024). At the same time as irrigation is introduced, the crops produced can also change. Producers produce crops with higher yields and economic gains in irrigated farming models (Zhang et al. 2021). It can be predicted that the organic matter in the soil may decrease over time due to increased tillage in addition to water use and production of high-yielding crops (Krauss et al. 2022; Delandmeter et al. 2024). Organic fertilization is necessary to maintain soil fertility and to create higher soil fertility. In addition to contributing to sustainable soil fertility, organic materials positively affect the quality of crop production models (Tavali et al. 2014; Maltaş et al. 2017). It has been reported in many studies that microbial diversity and activity in soil increases as a result of organic fertilization (Uz ve Tavali 2014; Tavali et al. 2019).

Binding of atmospheric gases to the soil through plants and storing them in the soil is one of the most strategic research topics today (Dziejarski et al. 2023). In addition, preventing the increase in atmospheric carbon dioxide in the terrestrial ecosystem and storing carbon dioxide in the soil is a sustainable approach as it is cheaper and low-cost (Alsarhan et al. 2023; Basheerve et al. 2024). In this context, it is necessary to make use of existing resources to find solutions to the environmental pollution problem caused by organic fertilizers, which are produced at a significant level, and different wastes released in agricultural production. Excess chemical nitrogen fertilization consumed in agricultural production causes nitrate accumulation in plants, which threatens human health. However, it was reported that plant nitrate contents did not change significantly when organic fertilizer applied plants were compared with non-fertilized plants (Wu et al. 2024).

As a result of increasing awareness of human health and environmental protection, interest in studies on this subject is increasing (Moshood et al. 2022; Lim 2024). These studies aim to reduce the use of pesticides, chemical

fertilizers and herbicides. Instead of traditional production methods, alternative production methods that do not use chemicals as inputs or use them less are being investigated. In addition, the plants grown are also improved through breeding methods (Chojnacka et al. 2020). Disease-resistant varieties with high fruit yield are being bred (Khan et al. 2020; Khan and Korban 2022; Murthyve et al. 2022; Bairagi et al. 2024). One of the most important factors for all these efforts to achieve their goals is to protect and even increase soil fertility (Kaplan et al. 2020; Maltaş 2023).

### **The Effects of Organic Matter in Soil**

Soil organic matter includes all components that are partially decomposed in the soil: plant and animal wastes, soil fauna+soil flora (microbial biomass) and humus, which is considered as a stable form. Very little of the organic matter applied to the soil is transformed into humus (Pikuła et al. 2022; Zhao et al. 2023). A significant portion breaks down and loses its organic matter properties (Hayes and Swift 2020). Organic fertilizers contain different amounts of macro plant nutrients (Table 1) and micro plant nutrients that plants need.

Table 1. Macro plant nutrient contents of some organic fertilizers (kg/ton)

Nutrients	Cattle manure	Liquid Manure of cattle	Sheep manure	Chicken manure	Pig manure	Liquid Manure of pig
N	4-5	4-6	8.0	15.0	4.0	4-6.5
P <sub>2</sub> O <sub>5</sub>	2-3	1.5-2	2-3	15-20	2.0	3-5
K <sub>2</sub> O	5-6	5-7	8.0	10.0	6-7	3-3.5
CaO	5-8	4.0	10-12	15.0	-	3.0
MgO	1-2.5	1.0	2.5	2.0	-	1.0

Due to the mineralization of organic matter in the soil, plant nutrients, especially nitrogen, are released (Shaji et al. 2021). The rate of decomposition of organic matter is generally preferred to be slow (Ferreira et al. 2020). However, the rate of decomposition of organic matter is significantly affected by soil texture, soil temperature and the amount of water present in the soil (Li et al. 2020; Angst et al. 2021).

Due to the decomposition of organic matter, plant nutrients are regularly released. In such cases, the amount of nutrients in the soil increases even though the plant does not need them and they can accumulate in the soil because the plant does not take them up (Shaji et al. 2021; Niederve et al. 2024). Such situations are not a problem in soils with low organic matter content. The

general problem can be encountered in cases where organic matter is added by producers. For this reason, producers often avoid or pay attention to the use of organic matter with high nutrient content (Maltaş et al. 2024). Often, to avoid problems with the use of high-nutrient organic fertilizers, organic fertilizers with a slightly lower nutrient content are preferred. In order to optimize the nutrients from organic fertilizers, organic fertilizers should be allowed to incubate. When organic fertilizers are applied just before sowing seeds or transplanting seedlings, the possibility of plant damage increases. For this reason, organic fertilizers should be applied to the soil before sowing seeds or transplanting seedlings. In practical cultivation, it is very difficult to meet all the nutrient needs of plants only with organic fertilizers. For this reason, organic fertilizers and chemical fertilizers are used together in agronomic cultivation except for special cultivation conditions such as organic farming (Tavali et al. 2019).

Organic matter also has an effect on soil pH value. There is a close relationship between pH, one of the important chemical properties of soils, and soil fertility. Soil pH has a direct and indirect effect on the availability of nutrients in the soil, the activity of fungi, bacteria and actinomycetes that provide productivity and efficiency to the soil, and the formation of soil structure (Laurent et al. 2020). Deficiencies of elements such as potassium, calcium, magnesium, phosphorus and molybdenum can be observed in acid soils, as well as elevation of elements such as aluminum, manganese and iron to toxic levels (Bhattacharya and Bhattacharya 2021; Bhatla and Lal 2023). On the other hand, in alkaline soils with high pH values, phosphorus, especially phosphorus, binds with calcium as insoluble tricalcium phosphates and becomes useless, which are the pH-dependent properties of these soils (riaz et al. 2020). There are many studies indicating that nutrient availability may be low in soils with high pH and lime values. It has been stated that lowering the soil pH value in high pH soils may be beneficial for plant nutrition (Maltaş and Kaplan 2018). Failure to apply organic fertilizers also generally lowers the soil pH value and provides better utilization of plant nutrients in the soil (Bhunja et al. 2021). It also ensures that the plant nutrients applied with fertilization are more easily taken up by the plants. Thus, it prevents extra fertilization. It prevents the formation of salinity in the soil. Another effect of organic matter is the formation of chelates. Due to chelate formation, it increases the usefulness of micro elements (iron, manganese, zinc and copper) (Dong et al. 2023; Yang et al. 2023). In addition, it is known to increase the usefulness of elements that form insoluble compounds such as phosphorus in soils with high lime content (Jindo et al. 2023; Wang et al. 2024).

Organic fertilizers generally contribute to the colloidal properties of the soil. Thus, they increase the cation exchange capacity of the soil (Eltohamy et al. 2023). Due to the increase in the cation exchange capacity of the soil, they prevent the washing of plant nutrients in the soil. They also ensure that the plant nutrients given to the soil by chemical fertilization are retained in the soil. In terms of these properties, the addition of organic matter, especially in sandy soils, increases the cation retention capacity of these soils and the washing of nutrients from the soil by precipitation is largely prevented (Singh et al. 2020).

Organic fertilizers also have effects on soil structure. It has an effect especially on aggregate formation and strength. It is known that there is a significant relationship between aggregates larger than 0.05 mm in the soil structure and the amount of organic matter in the soil. When the aggregate size increases, the relationship between large aggregates and organic matter becomes stronger. Both cases indicate that organic matter plays an important role in aggregate formation (Gautam et al. 2022). However, it can be said that the effects of organic matter on macro aggregate formation are more than the effects on micro aggregate formation. In terms of better soil physical properties in crop production, macro aggregate stability is very important for soil fertility. The effect of organic matter on aggregate formation is also related to soil texture (Tisdall 2020). Clay minerals are also effective on aggregate formation (Xue et al. 2022; Wang et al. 2023). However, clay minerals, which are part of the soil texture, decrease in soils with high sand content. Therefore, utilizing the effect of organic matter on structure formation in soils with high sand content textures is more important for soil fertility. With aggregate formation, the infiltration movement of water in the soil is also facilitated. The effect of organic fertilizers in forming water-resistant aggregates is also quite high (Zhapyayev et al. 2023). As a matter of fact, water is used in many crop production. In the use of water, the aggregates existing in the soil can disperse (Okolo et al. 2020). However, aggregates formed by organic matter can remain more stable against water. In this case, it can be said that the presence of water-resistant aggregates formed by organic matter is more beneficial both in terms of plant cultivation and especially in terms of preventing soil loss by water erosion. Another benefit of organic fertilizers in the formation of good structure in heavy clay soils is the prevention of the formation of the cream layer (Radulov and Berbecea 2024). The hard structure formed on the surface of the soil in heavy clay soils is called as the cream layer. Especially in plants grown by sowing seeds, the presence of the cream layer creates a great deal of negativity. After the germination stage, the seeds may not reach the soil surface by breaking the hard layer of clotting cream (Sethi and Kaur 2021). Thus, plants

that cannot reach the soil surface often die. In this case, plants grown from seeds may have problems. Since the number of germinated plants in the field decreases, it becomes difficult to manage all the cultural processes to be carried out afterwards. As a matter of fact, since the number of plants decreases, there is a great loss of yield in the grown product.

Another problem encountered in clay soils in crop production is the amount of air in the soil. Although clay soils are generally sufficient in terms of total porosity; they are insufficient in terms of the amount of air available to the roots (Eden et al. 2020). Because the number of micropores in clay soils is high, the total air capacity is high. However, the amount of macropores is more important in an ideal soil. In this context, organic fertilization is one of the most easily applicable methods for improving the air capacity of clay soils in terms of air capacity useful for plants (Singh et al. 2022). The opposite problem with the lack of air capacity in clay soils is experienced in the useful water capacity in sandy soils. Unlike clay soils, sandy soils have plenty of air. However, an air-water balance in the soil is essential for adequate root development in plant production (Gavrilescu 2021). In sandy soils, water capacity decreases due to the abundance of air. In such cases, the use of organic fertilizers increases the total water holding capacity of the soil and increases the useful water capacity in the soil (Table 2). Because organic matter has the ability to hold much more water than its own amount (Tisdal 2020; Hoffland et al. 2020).

Table 2. Effect of increasing amounts of barnyard manure on water holding capacity of soil

Amount of manure (ton/ha)	Water holding capacity (%)
0	31.5
10	33.5
20	34.3
40	36.6

Although organic matter retains water, the water retained by organic matter is retained with a strength that plant roots can easily take up. Organic fertilizers also contribute to the movement of water in the soil (Rashmi et al. 2020; Shaji et al. 2021). In clay soils, it is a problem that water is carried away by surface runoff without being able to move towards the lower parts of the soil. Conversely, in sandy soils, the problem is that water infiltrates directly into the deeper parts of the soil without spreading on the soil surface. Water going deeper can also cause easy washing away of the nutrients in the soil that are given by fertilization. This situation can both increase the consumption of

chemical fertilizers and cause water pollution as nutrients reach groundwater. With the use of organic fertilizers; in clay soils, organic matter loosens the soil and allows water to seep into the lower layers of the soil (Rahman et al. 2020). In sandy soils, organic matter retains water due to its high water retention capacity and prevents water and nutrients from going deeper into the soil. Thus, it contributes to preventing the pollution of groundwater by chemical fertilizer applications.

Tillage is facilitated due to the use of organic matter (Rahman et al. 2020; Man et al. 2021). When the water content is high in heavy clay soils, the soil sticks to the tillage tools during tillage (Mwiti et al. 2023). Soil structure may deteriorate due to soil adhesion to tillage tools. In addition, the tools and equipment used during tillage are easily damaged and worn out. Energy use also increases according to the degree of difficulty in tillage.

Organic fertilizers have an effect on the water coverage of soils (Abd-Elrahman et al. 2022). When organic fertilization is applied to a soil with a clay texture, it becomes faster tillage even if there is a high level of water. In other words, the time it takes for soils to become annealed is shortened. Even after high levels of rainfall, soils rich in organic matter can be cultivated earlier, as they are more annealed earlier. Organic matter not only shortens the time of annealing but also prolongs the time the soil stays annealed (Jashothan 2021). Because it brings the water it contains in its structure to the soil as it mineralizes. This is especially valuable for tillage and plant cultivation in sandy soils.

Organic fertilizers are darker in color, especially compared to mineral soils (Alzamel et al. 2022). Due to their dark color, they reflect the sun rays less, so the sun rays are largely retained in the soil. Due to the good retention of sun rays, the amount of energy in the rays in the soil increases. This shortens the warming time of the soil and makes the total temperature higher (Barros et al. 2021; Filimonenko et al. 2024). Higher soil temperature due to organic fertilizer applications has many advantages in crop production (Singh et al. 2020). Plants can be positively affected by even small increases in soil temperature during cold periods (Bergstrand 2022). For example, while the majority of roots can be damaged at low temperatures, the contribution of organic matter to soil temperature can reduce root damage. Another possible contribution is related to the availability of plant nutrients (Waqas et al. 2020). Plants take up mostly water-soluble forms of plant nutrients in the soil. Temperature is a factor that generally positively affects solubility. Therefore, increases in soil temperature due to its dark color during cold growing periods contribute positively to nutrient uptake. Furthermore, organic fertilizers are themselves energetically

charged (Šarauskis et al. 2021). So, in addition to their dark color, they can also increase the temperature of their surroundings during decomposition.

Organic fertilizers also have significant effects on soil organisms (Lazcano et al. 2021; Bhuni and others 2021). Soil organisms are recognized as an important component of soil quality. The total number and diversity of microorganisms in the soil is an important indicator of soil fertility. After the application of organic matter to the soil, different groups of organisms are involved in each stage of the process until humus formation (Xu et al. 2024). Organic matter functions as a source of energy and structure for microorganisms (Matheron and Caumette 2015). Therefore, C/N ratios of organic fertilizers applied to soil are considered as an important criterion (Cui et al. 2022). In case of using organic fertilizers that are rich in carbon and poor in nitrogen, temporary negative situations may occur. There are conditions in which microorganisms that find sufficient energy sources can increase greatly. While increasing the number of microorganisms in this situation, they can use the nitrogen they need to receive initially. At low C/N ratio, initially high nitrogen may occur in plants. Negative conditions such as resistance to cold, heat, common pests and nutrient dilution may occur due to high nitrogen nutrition. With this change, low C/N ratio and high C/N ratio are undesirable, although the organic fertilizers recommended to be used in plant production vary depending on the plant type and development differences. It is more beneficial for the ideal C/N to be in the range of 12-20 (Geisseler et al. 2021). It is more beneficial to use organic fertilizers with high and low C/N groups by composting them rather than using them directly. If these fertilizers are to be distributed directly, the subsequent parts must be distributed correctly.

They reported that pathogenic fungi and antagonistic organisms increased in soils where organic fertilizers were applied, but pathogens could not cause significant diseases in plants, and antagonist organisms kept diseases under control (Tao et al. 2020). In a study conducted to investigate the effects of organic and synthetic fertilization on microorganisms in fields where organic and conventional agriculture was applied, a significant increase in *Trichoderma* species was detected in plots where organic fertilization was applied, while a decrease in *Pythium* and *Phytophthora* species was found in the same plots compared to plots where conventional agriculture was applied, and there was no difference in the amount of *Fusarium* (Leyva-Morales et al. 2024). It has been reported in many studies that microbial diversity and activity in the soil increased as a result of organic fertilization (Sabir et al. 2021; Barber and Richards 2022). In addition, organic fertilization limits the effectiveness of soil-

borne pathogens of the genera *Pythium*, *Phytophthora* and *Fusarium* (Naghman et al. 2023).

### **Conclusion**

Organic fertilizer sources cover a wide range and play an important role in maintaining and improving soil fertility in agricultural production. Both plant and animal-based organic fertilizers contribute to sustainable agricultural practices by increasing soil fertility. The use of organic fertilizers forms the basis of environmentally sensitive and long-term productive agricultural methods. The main goal in plant production is to obtain more yield from the unit area. In addition to increasing yield, it is also aimed to maintain or even increase the quality of the product grown. Adequate, balanced and economical fertilization is of great importance to achieve these goals. Organic fertilizers applied to soils poor in organic matter mostly positively affect the physical, chemical and biological properties of the growing environment. In addition, considering that organic fertilizers contribute to yield and quality in plant production due to the fact that they provide plant nutrients to the growing environment, the use of these fertilizers is absolutely necessary. This requirement necessitates that every organic fertilizer source be evaluated as an input in agriculture. However, when choosing organic fertilizer, the characteristics of the fertilizer we will use should be well known. If there are problems, they should be resolved before use, or for problems that could not be resolved at the beginning, it is necessary to determine in advance the precautions to be taken against the problems that are expected to be encountered after using organic fertilizers.

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### **3. Bölüm**

## **Macro Element Deficiencies in Plants**

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## **Introduction**

Carbon, hydrogen, oxygen, nitrogen, potassium, calcium, phosphorus, magnesium and sulfur are known as macronutrients in plant nutrition (Zewide and Melash 2021).

These elements are considered to be non-mineral plant nutrients, since plants obtain hydrogen, carbon and oxygen mainly from air and water. Although they constitute a very large proportion of the plant mass, they are generally not considered in fertilization because they are in short supply.

Soil is very complex in terms of its physical, chemical and biological properties (Sung et al. 2017; Delgado and Gómez 2024). Therefore, it is often not possible to determine the exact amount and type of plant nutrients in the soil. In this context, chemical and organic fertilization is recommended (Gram et al. 2020; Han et al. 2021). In addition, even if fertilization is applied after determining the amounts of nutrients, it is known that the applied nutrients cannot be taken up by the plants due to many different factors. Therefore, deficiencies of these elements may occur in plants. The symptoms and effects of plant nutrient deficiencies vary according to the type of nutrient and severity of deficiency (Shrivastav et al. 2020; Asis and Niscioli 2024).

## **Nitrogen Deficiency in Plants**

Nitrogen, the most deficient plant nutrient, is taken up by plants in the forms of  $\text{NO}_3^-$  and  $\text{NH}_4^+$  (Zhu et al. 2021). The nitrogen content of plants may vary according to plant variety and even genotypes (Ben Mariem et al. 2020; Lemaire and Ciampitti 2020). The role of nitrogen in physiological and biochemical reactions occurring in plants is very important. Because nitrogen is found in the structure of many organic compounds such as proteins, nucleic acids and cell walls. It is also the basis of the chlorophyll molecule. In nitrogen deficiency, plants with reduced growth and smaller leaf area index benefit less from solar energy and therefore photosynthesis occurs less (Tian et al. 2022; Feng et al. 2023).

In nitrogen deficiency, the growth rate of plants decreases, i.e. plant growth slows down (Abbas et al. 2021). The plant remains small, leaves shrink and old leaves usually fall prematurely. Root development decreases and the amount of capillary roots that take the most water and nutrients decreases, especially since branching in the roots weakens. However, the root/stem ratio usually increases in nitrogen deficiency (Mathew and Shimelis 2022; Van Antwerpen et al. 2022). Nitrogen deficiency leads to degradation of chloroplasts and a decline in chloroplast formation. Therefore, chlorosis is observed in leaves in nitrogen deficiency. Chlorosis means yellowing of the leaf by losing its green color.

Chlorosis in leaves in nitrogen deficiency occurs as a homogeneous yellowing of the whole leaf. Yellowing is first seen in old leaves. In the advanced stages of deficiency and if it is very severe, necrosis can be seen in the leaves (Ahmad et al. 2020; de Bang et al. 2021). With this feature, nitrogen deficiency differs from potassium and magnesium deficiencies. Yellow and brown colored leaves appear early in plant development in potassium and magnesium deficiencies (Abbas et al. 2021; Yadav et al. 2023).

In nitrogen deficiency, plants mature early and the vegetative growth period is shortened. This premature senescence is probably due to the effect of nitrogen on cytokinin synthesis and transport. In nitrogen deficiency, the hormone cytokinin is reduced and this causes premature senescence and short vegetative growth period (Deepika and Singh 2021; Huang et al. 2023).

### **Phosphorus Deficiency in Plants**

Phosphorus is taken up by plants in the forms  $\text{H}_2\text{PO}_4^{+4}$  and  $\text{HPO}_4^{-2}$  and its amount in soil solution varies between 0.3 and 3 mg/kg (McDonald et al. 2001). Plants need phosphorus more in the early stages of their development (Grant et al. 2001). Phosphate compounds play an active role in all metabolic events occurring in plants (Dissanayaka et al. 2021; Khan et al. 2023). On the other hand, phosphorus is effective in the formation of genes and chromosomes, cell division and development in plants (Bechtaoui et al. 2021). In phosphorus deficiency, plant growth is retarded, seedling height, stem and leaf diameter shrink, and the number of leaves decreases. Therefore, the plant body has a delicate appearance instead of a woody appearance (Saengwilai et al. 2023).

In plants with phosphorus deficiency, growth is retarded and the stem/root ratio is usually reduced. Spike formation in cereals is negatively affected. In fruit trees, growth rate decreases, new shoot and bud formation is greatly reduced in phosphorus deficiency. Seed and fruit quality deteriorates and ripening is delayed (Muneer et al. 2024).

In phosphorus deficiency, leaves are usually darker green than normal. The leaves and stems of many annual plants turn red or reddish purple in P deficiency. The red color is due to increased anthocyanin formation in phosphorus deficiency. Anthocyanin is the red color substance. The leaves of fruit trees turn brown and fall off early (Sinha and Tandon 2020).

In phosphorus deficiency, the phosphorus coverage of plants is generally very low and is 0.1% or less in dry matter (Wissuwa 2023). The phosphorus content of plants that can receive sufficient phosphorus in the early developmental stages is in the range of 0.3 to 0.4% in dry matter. Generally, young plants and young plant organs have higher P contents. For this reason, P

contents of mature grain stalks are quite low, between 0.10-0.15% in dry matter (Malhotra et al. 2018). However, the P content of the seed is around 0.4-0.5%. These values indicate that there is an influx of P from leaves and stems to the seed during grain filling (Julia et al. 2016). Extremely high phosphate concentration in the root zone can negatively affect growth. This effect is mainly due to the fact that excess phosphorus negatively affects the uptake and transport of micronutrients such as Fe, Zn and Cu (Fan et al. 2021; Hui et al. 2022).

### **Potassium Deficiency in Plants**

Potassium is taken up by plants in the form of  $K^+$  and, although abundant in nature, the coverage of potassium ions in soil solution varies between 0.1% and 0.2% (Sardans and Peñuelas 2021). Potassium, which is vital for plants, improves the quality and quantity of plants, that is, their quality, and provides protection from frost, diseases and pests (Torabian et al 2021). The element potassium has a vital role in activating enzymes in plants, protein synthesis and photosynthesis. In potassium deficiency, shrinkage and cell death occur due to insufficient turgor formation in plants, vegetative growth and development are retarded, and root development is negatively affected (Sardans and Peñuelas 2021; Rawat et al. 2022). In the advanced stages of potassium deficiency, chloroplasts and mitochondria are damaged as the necessary energy is not transferred. In addition, water use efficiency in the plant is impaired due to the decline in cutin formation (Kusaka et al. 2021; Imtiaz et al. 2023).

Potassium is present in plants as a cation of organic and inorganic salts. However, it is not included in the structure of any organic compound. However, it is known that potassium element causes significant damages in plants due to its important physiological functions in plants (Mostofa et al. 2022).

Visible symptoms of potassium deficiency in plants may not appear immediately. First there is a decline in growth rate (latent starvation), but then chlorosis and necrosis appear. Potassium deficiency symptoms usually appear first on the older leaves. Because in case of deficiency, potassium is transported from old leaves to young leaves. Potassium is a mobile element in plants. Deficiency symptoms are first seen on leaf edges and tips in many plants. The leaf margins turn yellow first, then the color turns dark brown in these parts. If the deficiency is very severe, these parts die and dry and fall off (Fontana et al. 2020; Abbas et al. 2021). In potassium deficiency symptoms, which are very typical especially in fruit trees, although the edges of the leaf die after the discoloration occurs, the rest of the leaf can maintain its normal green color and appearance for a long time.

In potassium deficiency, heterogeneously distributed necrotic spots may occur on the leaves of some plant species. Some virus diseases and symptoms that occur in unfavorable climatic conditions such as drought can be likened to potassium deficiency (Ortel et al. 2024).

In potassium-deficient plants, turgor pressure decreases and the leaves of plants wilt when water stress occurs. Sensitivity to drought and frost increases. Similarly, plants are more susceptible to disease agents and saline soil conditions (Bhattacharya and Bhattacharya 2021; Perelman et al. 2022). Abnormal developments are observed in plant tissues and cell organelles. The formation of xylem and phloem tissues is retarded. Ligninization decreases in tissues. Potassium is a plant nutrient element that directly or indirectly affects many quality factors in plants. For this reason, potassium deficiency leads to a decrease in quality criteria in plants (Sardans and Peñuelas 2021; Huang et al. 2022; Grzebisz et al. 2022).

### **Calcium Deficiency in Plants**

The plant nutrient calcium is taken up by plants in the form of  $\text{Ca}^{+2}$ . Calcium plays an important role in cell division, elongation and strengthening of membranes (Pathak et al. 2020). It also regulates the functioning of stomata (Naz et al. 2024). In the presence of sufficient amounts of calcium in the rhizosphere, the negative effects of biotic and abiotic stresses can be somewhat eliminated (Jing et al. 2024). Calcium also maintains the balance between nutrients in plant tissues and plays an important role in eliminating the toxic effects of heavy metals (Riyazuddin et al. 2021).

Calcium deficiency in plants slows the growth of meristem tissues. Deficiency symptoms first appear in the growth points and young leaves. Young leaves are deformed and black and brown necroses appear on the leaf margins. As the cell walls dissolve in deficiency-damaged tissues, the deficient cells become soft (Panter et al. 2020; Torres et al. 2024).

Since mineral soils are usually rich in calcium, calcium deficiency symptoms in plants are rare. In contrast, decreases in calcium flux to fruit and storage organs more often cause calcium deficiency symptoms. In all plant tissues calcium ions are transported by bottom-up water movement in the xylem tubules due to transpiration. If the concentration of calcium ions in xylem water is low or transpiration from the fruit is low, the amount of calcium ions reaching the fruit is insufficient and symptoms occur (Alvarado-Camarillo et al. 2024). High nitrogen nutrition in the form of ammonium, soil water deficiency and high EC value reduce the amount of calcium ions in xylem water (Prasad and Shivay 2020; Sharm and others 2024; Zheng and others 2024).

The uptake and transport of calcium from the soil solution is through the root tips. Therefore, conditions that prevent the formation of new roots, such as low and high temperature and low oxygen capacity, can prevent calcium uptake and cause calcium deficiency (Johnson et al. 2022; Zafar et al. 2024).

Calcium in phloem tissues is not transportable. For this reason, it is very difficult for the previously absorbed calcium to be transported in the phloem and reach the fruit during the fruiting period. During the ripening period of the fruit, if sufficient amount of calcium element is not taken from the soil and does not reach the fruit through the xylem; calcium deficiency may occur in the fruits. For this reason, analyzing plant leaves to determine the calcium nutrition status does not provide any benefit. Because the calcium element in the leaves is not transported from the leaves to the fruit (Reitz et al. 2021; Jaime-Guerrero et al. 2024).

### **Magnesium Deficiency in Plants**

Magnesium is taken up by plants in the form of  $Mg^{+2}$ . In addition to its positive effects on roots and stems in plants, magnesium has very important effects on chlorophyll formation, protein synthesis, photosynthetic carbon dioxide assimilation, chlorophyll formation, protein synthesis, and activation of many enzymes (Rodrigues et al. 2021; Jiao et al. 2024). In addition, magnesium has important functions in the realization of many different physiological and biochemical events such as energy formation, utilization and distribution of photosynthesis products (Mariyam et al. 2023).

There may be differences between plant species in terms of magnesium deficiency symptoms. However, there are some common characters. Since the element magnesium is mobile within the plant, it is easily transported from old leaves to newly formed leaves in case of deficiency. For these reasons, Mg deficiency is first seen in old leaves (Ishfaq et al. 2022). The most prominent symptom of magnesium deficiency is yellowing between the veins in old leaves. While the primary and secondary veins retain their green color, the third and thinner veins and interveinal areas become lighter in color and yellowing occurs. Due to the color changes, especially the old leaves have a mottled appearance. At very high magnesium deficiencies, brown discoloration occurs in chlorotic tissues (Treter et al. 2022; Li et al. 2024). In cases where such observations occur, yield and quality loss can be intense.

Especially in sandy soils, magnesium deficiency is common towards the maturity stages of plant growth (Chaudhry et al. 2021). In vegetables whose fruit is eaten, magnesium deficiency that occurs during the harvest period may not cause a major loss in the product. In vegetables whose leaves are eaten, the

market value of the plant may decrease as the leaf quality decreases. Soil compaction, waterlogging and drought effects exacerbate magnesium deficiency (Nawaz et al. 2020). Since magnesium is the central atom of chlorophyll, chlorophyll formation is first retarded in magnesium deficiency (Ali et al. 2023; Li et al. 2024).

### **Sulfur Deficiency in Plants**

Sulfur is taken up by plants in the form of  $\text{SO}_4^{2-}$  (Ranadev et al. 2023; Yasin 2024). Sulfur is present in the structure of amino acids such as cystine, cysteine and methionine in plants (Narayan et al. 2023). Sulfur element is both present in the structure of the chlorophyll molecule and is an essential plant nutrient element for chlorophyll synthesis (Tiwari et al. 2020; Narayan et al. 2023). In addition to its role in reduction and oxidation reactions, it is also involved in the activation of enzymes (Miękus et al. 2020). Sulfur is an element that is effective in the formation of vitamins and the synthesis of some hormones (Li et al. 2020). Sulfur nutrition is important for drought, cold resistance and prevention of soil-borne diseases (Rupp 2020; Najafi et al. 2021; Narayan et al. 2023).

Since the element sulfur is present in the structure of proteins, protein synthesis in plants is reduced in deficiency. Proteins cannot be synthesized due to the lack of sulfur-containing amino acids such as cysteine and methionine, which are the building blocks of proteins (Youssef-Saliba and Vallée 2021). Sulfur deficiency is similar to nitrogen deficiency, but sulfur deficiency is observed in young leaves, while nitrogen deficiency occurs in older leaves (Carciochi et al. 2020; Abbas et al. 2021). Sulfur deficiency causes the color of the leaves to lighten and depending on the severity of the deficiency; the color of the leaf changes from light green to yellow. Plant growth is reduced in sulfur deficiency, but the developmental retardation in vegetative development is greater than the retardation that occurs especially in the root part. Therefore, in sulfur deficiency, the plant stem is usually thinner and therefore easily breakable (Karthika et al. 2023; Chaudhary et al. 2023; Hammerschmiedt et al. 2024).

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## **Chapter 4**

### **Pesticides and Effects On Aquatic Ecosystem**

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## **Abstract**

Pesticides are the chemicals that are used to eliminate insects, microorganisms and pestilents decreasing nutritional value and harming to plant during production, consumption and storage of nutrients. The chemicals that are used in many areas have toxic effect and they can continue their existence in nature for a long time. When reach into water sources, they accumulate in aquatic organism tissue and this can be harmful. Despite their harmful effects, use of the pesticides both in agricultural land and other manufactured products is a indispensable necessity. The pesticides cause to change in physico chemical properties of water, massive death or changing habitat of fish and other aquatic living as a result of moving them with both groundwater and rain or moving of factory waste into river. In an aquatic ecosystem, massive death of planktonic organisms occurs depending on exposure to the pesticide residue of the organisms that part essential nutritional source and have quite high biomass amount. This dramatic decline in the biomass of the planktonic organisms also affects productivity of upper level of the aquatic ecosystem. As an inseparable part of technology around the world, residue risk in use of the pesticide and negative effect of it to environment are subjects must be considered carefully.

**Keywords:** Ecosystem, Pesticide, Water pollution, Toxicity

## **Introduction**

With the developing world and increasing population, the need for food, one of the most important needs for life, is rapidly increasing. Products produced through uncontrolled industrialization, chemicals used to increase the durability of these products, destroyed agricultural areas, and substances used to increase production quantities to meet the increasing food demand are being tested in various ways. For this purpose, plant growth regulators are widely used to increase agricultural productivity. One of the basic conditions for increasing yield and quality in agricultural areas is to combat wild plants, fungi and insects. It is stated that if agricultural control is not carried out against these pests, an average of 35% of the crop loss will occur every year (Bekbölet, 1992; Uluocak & Egemen, 2005). For this reason, the use of pesticides in today's agricultural practices and in many different areas further increases the negative impact of these substances on the ecological environment.

In order to meet the food needs of the increasing world population and to ensure the increase in agricultural products, the use of pesticides has taken a large place in the monoculture practices that became widespread as a result of agricultural policies implemented in the 1960s and 1970s (Akdemir, 2002; Altındağ & Özgökçe, 2006).

The World Health Organization (WHO) and the Food and Agriculture Organization of the United Nations (FAO) define pesticides as "a substance or mixture of substances used to control or prevent undesirable plants and living things." In other words, it is stated as a general name given to all chemical compounds used against various factors that may cause the decline of agricultural products, such as plant diseases, harmful insects and harmful weeds (Şişli, 1994; Ağca, 2006).

Natural pests are called pests, and synthetic organic substances used to combat them are called pesticides (Öncüer, 1991; Gündüz, 1994; Ağca, 2006). Chemical substances or mixtures of substances used to prevent, eliminate, reduce or repel the effects of entities that harm humans, plants or animals in nature are called "pesticides" (EPA, 2009; Karakoç & Nakiboğlu, 2010). This definition includes disinfectants, fungicides, fish feeds and insect repellents that we use in our daily lives, as well as substances used in chemical methods against organisms that harm agricultural products. Pesticides are chemical substances used to destroy insects, microorganisms and other pests that spoil the nutritional value of food and harm plants during production, consumption and storage. In addition, chemical substances produced in the industry and used in many areas can be included in this group (Figure 1).

The use of pesticides is becoming more widespread as their benefits in agricultural production, animal feeding, post-harvest technology, public health and human well-being are understood. Even if pesticides are used in limited areas, they are carried by rain and floods to larger water bodies such as pools, lakes or rivers and change the physico-chemical properties of the water there (Richardson, 1988; Atamanalp et al., 2002). If pesticides reach water bodies, they harm fish and other living creatures or aquatic products in the water. Their mobility in water depends partly on their solubility and formulation. Pesticides that can dissolve in water or are formulated to dissolve in water disperse in a short time in water. In addition, those formulated in powder or granule form remain suspended in water and cause their active ingredients to spread for a long time. The first and most affected by these changes in the aquatic ecosystem are phytoplankton. In addition, other vertebrate and invertebrate organisms and fish in the food chain are also affected. Fish can be poisoned by pesticides either by absorbing them from the aquatic environment through their gills or by consuming contaminated materials as food (Toros & Maden, 1991; Atamanalp et al., 2002).



**Figure 1.** Use of pesticides in agricultural lands and fruit growing.

Especially in our country and in developing countries, the unconscious and excessive use of pesticides protects agricultural products against diseases, pests and weeds, while at the same time threatening the lives of all living things, especially humans, by creating an environmental pollution problem, and causing negative effects in terms of both the producer and the country's economy (Özgüven & Katkat, 1997; Atamanalp et al., 2002).

## **1- Main areas of use of pesticides:**

- In horticulture and agricultural production
- In ornamental areas (parks, gardens, playgrounds)
- In forestry and animal husbandry
- Fumigation and timber protection
- In fish farming
- In food storage
- Public hygiene, insect control
- Insect control in aquatic environments (sea, lake, river and water bodies)
- Industrial insect control
- In construction, homes and gardens (Wallpaper adhesives, paints, plastering etc.)
- In the pharmaceutical industry (Kutlu, 2006).

## **2- Classification of pesticides:**

Although pesticides can be classified in various ways, the most important classification is based on their chemical structure, active ingredients and the living groups they affect.

Pesticides can be classified in many ways according to their chemical structures.

It is possible to examine pesticides under 3 groups according to their chemical structure (Canyurt, 1994; Ağca, 2006).

- Inorganic substances (pesticides containing copper, sulfur, arsenic),
- Organic substances (petroleum oils, tar oils and those containing organic substances extracted from plants),
- Synthetic organic compounds.

There are also four basic groups of pesticides that are widely known:

### **1) *Organochlorine pesticides***

Organochlorine pesticides belong to the group of chlorinated hydrocarbon derivatives and are synthetic pesticides widely used all over the world. Organochlorines are a group containing carbon, hydrogen and chlorine atoms in their structures. DDT, aldrin, dieldrin, heptachlor, endosulfan, lindane, endrin are examples of this group. They have a wide range of applications in agriculture and chemical industry. These chlorinated compounds are banned in most developed countries due to their high toxicity, slow degradation and bioaccumulation. Although chlorinated pesticides were developed with the concept of target organism toxicity, they generally have quite a lot of adverse effects on non-target species. They are effective through contact and inhalation (Jayaraj et al., 2017; Figure 2).

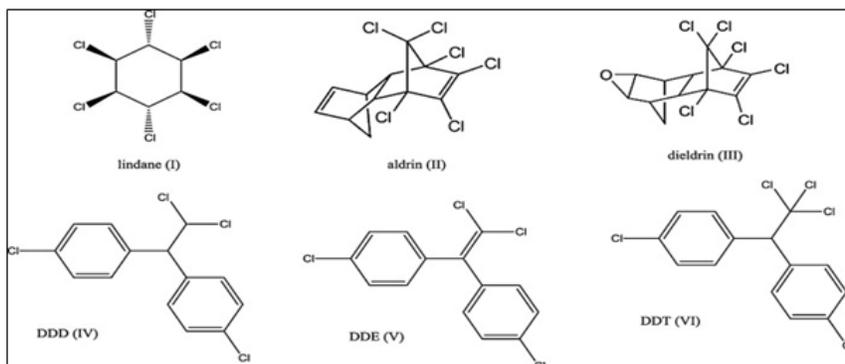


Figure 2. Organochlorine pesticides (Novak & Kovač, 2011)

## 2) Organophosphorus pesticides

Organic phosphorus pesticides are generally in the tryster structure and are phosphoric acid derivatives, and their active ingredients contain phosphorus atoms. Chlorpyrifos, coumaphos, diazinon, dichlorvos, malathion, trichlorfon, parathion, mevinphos are examples of this group. There are more than a hundred active ingredients in this group. Since they are in the tryster structure, the chemical structures of the pesticides included in this group can vary considerably according to the characteristics of the ester groups. They show their effects through contact, ingestion and respiratory tract. (Çetinkaya, 2015; Figure 3).

## 3) Carbamate pesticides

Carbamates are carbamic acid esters. Aldicarb, carbaryl, carbofuran, methiocarb, methomyl, oxamyl, pirimicarb are examples of this group. They are acetylcholinesterase inhibitors. Carbamate insecticides are relatively harmless for vertebrates due to their slow penetration through the blood brain barrier (Jokanovic, 2009). Tarig et al. (2010) stated that carbosulfan compounds can pollute the environment and become a problem when they reach water supplies (Figure 4).

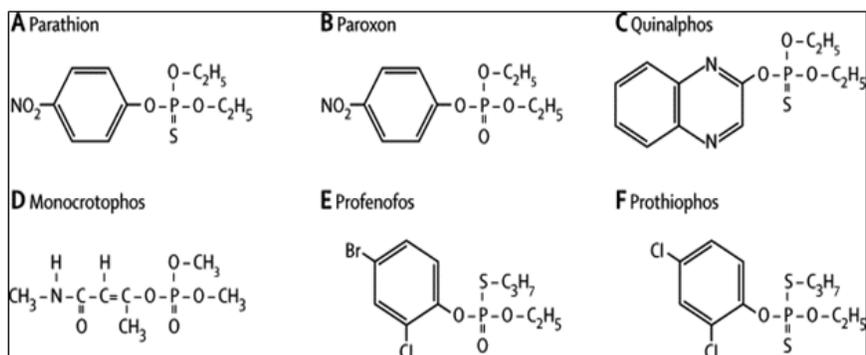
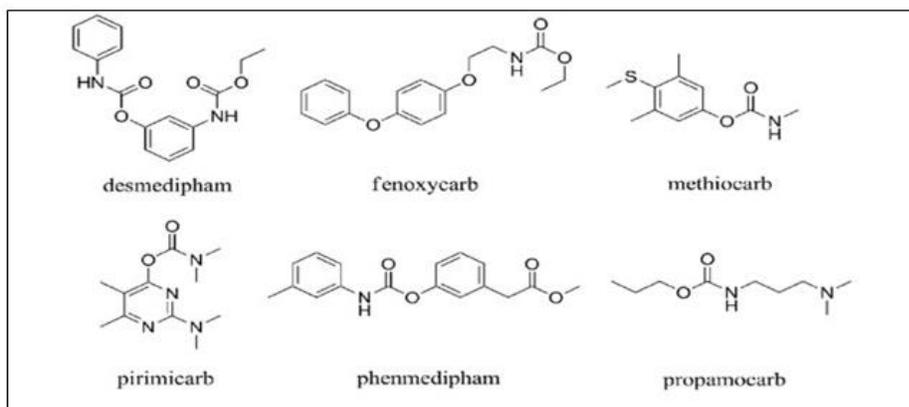


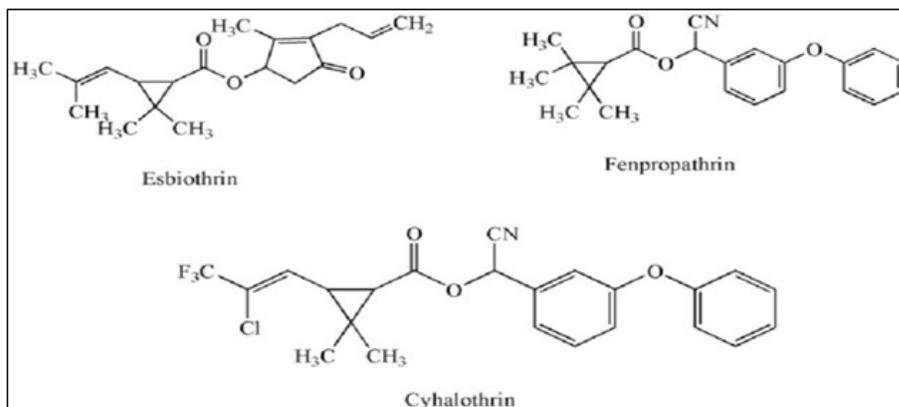
Figure 3. Organophosphorus pesticides (Eddleston et al., 2008)



**Figure 4.** Carbamate pesticides (Vlcek & Pohank, 2012).

#### 4) Pyrethroid group pesticides

Pyrethroids are synthetic analogues of pyrethrins, which are natural compounds obtained from chrysanthemum flowers. They are compounds with significantly reduced sensitivity to light and increased durability compared to pyrethrins. Pyrethrins and pyrethroids are usually used together to create a synergistic effect. Alpha-cypermethrin, cyfluthrin, bifenthrin, lambda-cyhalothrin, deltamethrin, permethrin, fenvalerate are examples of pyrethroids. They show their toxicity by blocking nerve cells. They are effective as contact and stomach poison. Their toxic effect against warm-blooded animals is very low. They are excreted without accumulating in the mammalian body. They can be easily decomposed in nature (Çetinkaya, 2015).



**Figure 5.** Pyrethroid group pesticides (Han et al. 2014).

Pesticides are classified as follows according to their active substances, taking into account the harmful group they are used in (Öztürk, 1990; Karakaya & Boyraz, 1992); (Figure 6).

**A) INSECTICIDES**

- 1- Chlorinated hydrocarbons
- 2- Organophosphorus
- 3- Carbamates
- 4- Synthetic pyrethroid
- 5- Bacteria
- 6- Others

**B) ACARICIDES**

- 1- Halogen and its oxides
- 2- Amine and hydrazine derivatives
- 3- Dinitrophenol
- 4- Sulphurous
- 5- Organic tin
- 6- Others

**C) FUNGUCIDES**

**a- Protective fungicides**

- 1- Copper
- 2- Tin
- 3- Sulfur
- 4- Dithiocarbamate
- 5- Phthalimides
- 6- Nitro compounds
- 7- Others

**b- Systemic fungicides**

- 1- Anilides
- 2- Benzimidazoles
- 3- Morpholines
- 4- Piperazines
- 5- Pyrimides
- 6- Triozoles
- 7- Others

**D) HERBICIDES**

- 1- Phenoxy compound
- 2- Benzimidazole
- 3- Picolinic acids
- 4- Chlorinated aliphatic acids
- 5- Carbomates
- 6- Dinitroamine analine
- 7- Anilides
- 8- Urea compounds
- 9- Triazines
- 10- Uracyls
- 11- Nitrophenol and its derivatives
- 12- Others



**Figure 6.** Various form of application of pesticides in different areas

**Classification of pesticides according to the living groups they affect:**  
(Karadağ, 2007), (Figure 7).

- Insecticides (kills insects)
- Acaricides (kills spiders)
- Nematicides (kills worms)
- Molluscicides (kills molluscs)
- Rodenticides (kills rodents)
- Avicides (kills birds)
- Aphicides (kills aphids)
- Fungicides (kills fungi)
- Bactericides (kills bacteria)
- Herbicides (kills weeds)
- Algicide (kills algae)
- Miticides (kills ticks)
- Larvicides (kills larvae)



Larvicides kills larvae (Anonymous, 2024a)



Molluscides kills molluscs  
(Ansari et al., 2024)



Miticides kills ticks  
(Anonymous, 2024b),



Nematicides kills worms  
(Anonymous, 2024c).



Aphicides kills aphids  
(Anonymous, 2024d).

Figure 7. Some organisms that pesticides affect.

### 3- Pestisitlerin canlı gruplara etkileri

Insecticides kills insects: Pesticides can be classified in various ways based on their chemistry, toxicological effects, or mode of penetration. They are classified according to whether they are effective when swallowed (stomach poisons), inhaled (fumigants), or penetrate the body covering (contact poisons). However, most synthetic pesticides penetrate through all three pathways and are therefore better distinguished by their basic chemistry. In addition to synthetics, some organic compounds occurring naturally found in plants are beneficial insects. Most insecticides are sprayed or dusted onto plants and other surfaces where insects pass or feed, as are some inorganic compounds; some of these are allowed in organic farming practices (Anonymous, 2024e).

Carbamates Molluscicides: A common class of compounds. They are carbamates used as insecticides and pesticides. Molluscicides on the other hand, are compounds that are used to kill or control molluscs, such as slugs and snails. For this reason, compounds belonging to the carbamate class that are used

specifically are called as carbamate molluscicides. Molluscicides are substances that directly kill molluscs or prevent plants from feeding. The way these molluscicides work is by interfering with the nervous system of the target mollusk, eventually to their death. Different formulations, including pellets, powders, and liquids, can be used for different purposes depending on the product and application technique. They might be in the form of baits, sprays, or granules and they can be applied in different ways (Kloos & McCullough, 1982; Kashyap et al.,2019).

Miticides kills ticks: Miticides, also called acaricides, are a chemical substance used to control mites or ticks (specifically species that damage ornamental, food plant and other plants) that are not susceptible to commonly used insecticides. Many miticides kill eggs and larval stages as well as adult animals. Acaricide, any chemical substance used to control mites or ticks (specifically species that damage vegetables or ornamental plants) that are not susceptible to commonly used insecticides. Dicofol, ovex, tetradifon and azobenzene are commonly used miticides. Many miticides kill adults as well as eggs and larval stages. Some are also toxic to honeybees and other beneficial insects. Pesticide (insecticide) any poisonous substance that is used to kill insects. Such substances are primarily used to control pests that infest crops or to eradicate disease-carrying insects in certain areas (Anonim, 2024b).

Larvicides kills larvae: Larvicides target larvae in their breeding areas before they can mature into adult mosquitoes and disperse. Larvicide application to breeding areas helps reduce the adult mosquito population in nearby areas (EPA, 2024).

#### **4- The status of pesticides in the world:**

Throughout history, people have tried to protect the environment and plants used as food sources from vertebrates, invertebrates and various harmful microorganisms. For this purpose, many chemical substances, generally called pesticides, have been used to this day. There are records of the preparation and use of pesticides against lice, fleas and wasps in a papyrus dating back to 1500 BC. The first substances used as pesticides in history were sulfur and arsenic. It is known that sulfur, which was used as a fumigant by the Chinese in 1000 BC, was used as a fungicide in Europe in the 1800s and continues to be used as an important pesticide today. In the 16th century, the Japanese used a mixture of whale oil and vinegar and arsenic-containing compounds by the Chinese, in the late 17th century, liquid obtained from tobacco leaves, and in the 19th century, substances such as copper sulfate and lime were used as pesticides (Dağlıoğlu, 2004; Yücel, 2007; Akçan, 2008).

With the advances in modern synthetic chemistry in the 1930s, many pesticides were developed. A significant portion of organic insecticides were synthesized by a group of chemists led by Schrader in Germany. It was understood that these substances were extremely toxic and some of them were used as chemical warfare weapons under the control of the Nazis in World War II. The synthesis of Tabun and Sarin, which were effective chemical weapons and were described as nerve gases, was kept secret. In 1944, Schrader synthesized a more durable compound, Parathion, and its oxygen analog, Paraoxon. The production and use of organic pesticides increased significantly after World War II. As a result of the increase in pesticide use, residual pollution occurred in water, soil, air and the food chain, and errors in production and use increased acute and chronic poisonings (Vural, 1996a; Klassen, 2001; Dağlıoğlu, 2004; Akçan, 2008).

With the rapid increase in the world population, the need for agricultural products is also increasing (Ni et al, 2004; Karakoç & Nakiboğlu, 2010); however, agricultural lands are constantly decreasing due to non-purposeful uses (summer houses, factories, highways, settlements, etc.) (Kozak, 2009; Karakoç & Nakiboğlu, 2010). The efficient production of agricultural products is also becoming more and more important every passing day. One of the main factors affecting productivity is various plants, animals or microorganisms that prevent the emergence and development of products. Different methods are used to prevent these pests and the most commonly used is the chemical method (Delen et al., 2005; Karakoç & Nakiboğlu, 2010).

One of the most important problems of today's world is the inability to obtain enough food to meet the needs of the rapidly increasing world population. According to the reports of the Food and Agriculture Organization (FAO), people need approximately 15-20 million tons of agricultural food every year. Since the world's surface area is limited, it is not possible to open new areas to agriculture to meet this ever-increasing need. For this reason, various chemicals grouped under the name of pesticides have begun to be used in order to increase the yield obtained from unit area (Dağ et al., 2000; Könen, 2007). The most important areas of use of pesticides are agricultural control. They also play a role in the control of some diseases such as malaria, plague, yellow fever, and typhus, which are spread by vectors. Since these compounds are sold freely in the market, their use is quite widespread. Today, approximately 1500 pesticide active ingredients, most of which are organic compounds, are used against 2000 agricultural pests (Vural, 1996a; Dağlıoğlu, 2004; Akçan, 2008).

In the daily work of forensic medicine, poisoning is frequently encountered among the causes of death. Poisoning can occur due to many different factors

and acute pesticide poisoning, which constitutes a significant part of these, is a significant cause of mortality all over the world. An average of 2.5 million tons of pesticides are used annually worldwide. Although 80% of pesticides are used in developed countries, most of the poisoning cases occur in developing or underdeveloped countries. According to the World Health Organization reports, approximately 3 million pesticide poisonings occur each year and 220,000 of these poisonings result in death (Vural, 1996a; Malley, 1997; Dağlıoğlu, 2004; WHO, 2007; Akçan, 2008).

Pesticides are an integral component of modern agriculture. Today, the production process in all agricultural ecosystems requires at least one or more pesticide applications. For this reason, pesticides are considered to be indispensable substances all over the world (Dağ et al., 2000; Könen, 2007).

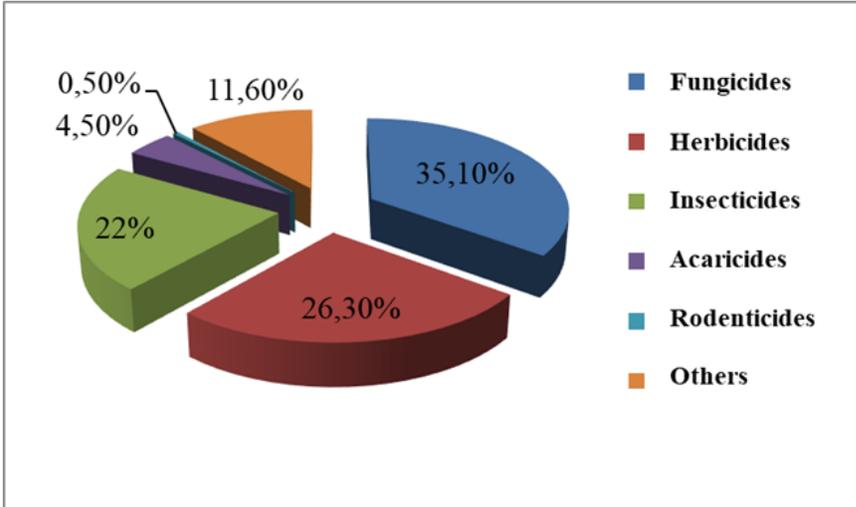
### **5- The situation of pesticides in Turkey:**

In pesticide use in Turkey, organophosphorus compounds are in the first place with a rate of 60.84%, followed by synthetic pyrethroids (17.65%) and carbamates (13.23%). Due to both the excessive use of organophosphorus insecticides and the acute poisoning effects of these compounds, most pesticide poisonings in Turkey are caused by organophosphorus insecticides (Zeren, 1994; Yalvaç et al., 2004)

In Turkey, 40% of annual pesticide consumption is concentrated in 3 provinces, namely Adana, İçel and Antalya. When the İzmir region is added, this value exceeds 65%. The pesticide use rate in Adana is 10.39% and in İçel it is 15.69%, and a total of 26.08% of pesticides are used in these two provinces. These figures, which are seen in the official recommendation, reach 35-40% with the encouragement of drug dealers and non-experts on the subject (Zeren & Erem, 2000; Yalvaç et al, 2004).

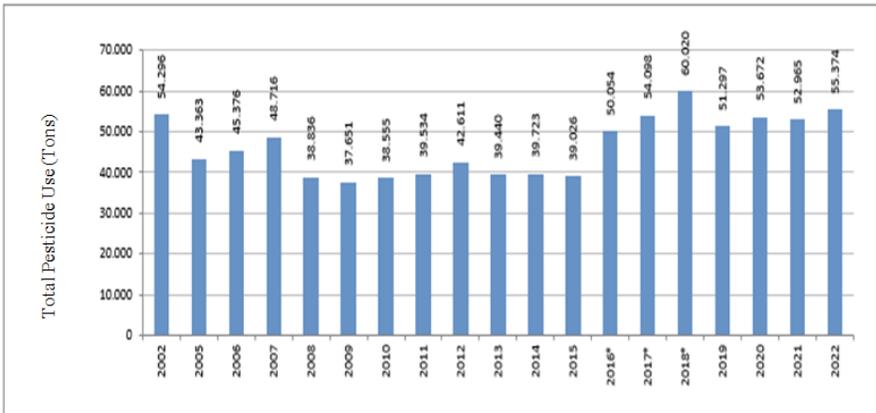
The total amount of pesticide use in Turkey in 2022 increased by 4.5% compared to 2021, reaching 55,374 tons. When the amounts of pesticide use are examined on a group basis, fungicides (fungi killers) constitute the largest group in our country, as in the world. It was determined that 35.1% of the total agricultural pesticide use was fungicides, 26.3% herbicides (weed killers), 22.0% insecticides (insect killers), 4.5% acaricides (miticides), 0.5% rodenticides (rodent killers) and 11.6% others (plant activators, plant growth regulators, insect attractants, fumigants, nematocides, sulfur, mineral oils), (Figure 8). The top 5 provinces with the highest pesticide use were Antalya (4,272 tons) with 7.7% of total use, Manisa (4,213 tons) with 7.6%, Mersin (3,985 tons) with 7.2%, Adana (3,276 tons) with 5.9%, and Malatya (2,280

tons) with 4.1%. Total pesticide amounts used in Turkey by year are shown in Table 1 (T.O.B., 2023).



**Figure 8.** Consumption percentages of total pesticides according to their groups in 2022 (in Turkey).

**Table 1.** Total Amounts of Pesticides Used in Turkey by Year



**6- Dispersion of pesticides in the natural environment:**

As a result of the application of pesticides to soil, plants or other industrial products, they reach water, air and soil as a result of various transports depending on the chemical properties of the substance and cause significant environmental problems in the atmosphere.

- After pesticides are applied, they can be absorbed by the soil and accumulate in the soil over a period of time.

- Without undergoing any chemical change, they can pass into the atmosphere through evaporation and from there into surface waters through rainwater.

- Pesticides found in the soil can pass into natural waters through rainwater and drainage water.

- They are washed down to the lower layers of the soil by infiltration water and are removed from the soil and can accumulate by passing into underground water resources.

- Some of them can be absorbed by plants.

- They can undergo changes in the soil surface and plant surface, in the water environment, under the effect of light energy.

- They are decomposed by microorganisms.

Pesticides carried into the atmosphere by wind reach the sea with rain and atmospheric dust. In rivers, they come attached to particles. The fact that pesticides are found even in Antarctica is very important in terms of showing that agricultural chemicals are transported to non-target areas (Raymond et al., 1992; Yalvaç et al., 2004).

### **7- The effect of chemical pesticide use on the environment:**

While some of the pesticides used evaporate and cause environmental problems in the atmosphere, some of them break down by photochemical means and turn into toxic or non-toxic substances. Some of them are retained in the soil, pollute the soil, and break down as a result of chemical and microbiological activities in the soil. Another part is dragged from the soil surface with snow and rain water, polluting lakes, rivers and groundwater.

The flow chain of pesticides in nature is as follows:

Air – plant – soil – human

Soil – water – animal – phytoplankton – fish – human (Güven, 2005).

It is generally reported that 80-94% of the applied pesticide reaches the soil, depending on the application technique, the phenological status of the plant and plant density (Gürkan & Ünal, 2001; Balcı & Gedikli, 2011).

Some of the pesticides are used by the plant, some of them remain in the soil and disrupt the physical structure of the soil, while others dissolve with irrigation water or snow and rainwater and reach streams, rivers, lakes and eventually the seas. The biological structure of streams and seas polluted with pesticide waste is disrupted. The movement of pesticides towards surface or groundwater can cause the pollution of clean water beds and can cause the

dissolved oxygen level in the water to fall below the lethal threshold value (Freeze & Cherry, 2003; Todd & Mays, 2005). This causes the number of certain plant and animal species to decrease. Mass fish and animal deaths occur. As a result, it causes the ecological balance to be disrupted or negatively affected.

Chemical drugs also have environmental damage caused by their production. The pesticide industry is one of the main sources of pollution in the chemical industry. Its production produces large amounts of highly toxic, concentrated waste gas and water. Due to its high inorganic salt and organic chloride content, wastewater with high chemical oxygen demand is difficult to dispose of (Klein, 1999; Anonymous, 2009; Balcı & Gedikli, 2011).

Another problem with pesticides is that organisms develop resistance to pesticides over time. Despite the high toxicity of chemicals, their use tends to decrease over time. Unwanted insects have gradually developed resistance to chemicals. Because there are so many insects and a small number of them may have genes that make them immune to pesticides by chance. Applying pesticides to a field may kill 99.99% of the insects, but 1 in 10,000 will survive. If one insect survives by showing resistance to the pesticide and comes into contact with other insects, the others will show similar resistance. These insects, which are genetically resistant to the drug, will form new resistant populations. As a result, the effectiveness of pesticide applications will cease (Brennan & Witthgott, 2005; Balcı & Gedikli, 2011).

Pesticides entering the food chain accumulate and magnify and accumulate in the biotic and abiotic environment, creating a significant environmental problem. The most important problem with pesticide use is the difficulty of precisely determining when, where and how it interacts with the environment and the damage it can cause to humans and nature over a long period of time (Klinghard, 1995; Yalvaç et al., 2004). Pesticides enter animal organisms through various means (oral, skin and respiratory tract). When they enter organisms, they cause some cells and tissues to be affected and even death (Karadağ, 2007).

### **8- Effects of pesticides on aquatic ecosystems:**

Pesticides that reach the sea, lake and stream environment through various transport mechanisms are diluted and spread by processes such as current and wave. Due to surface tension, various particles are held on the water surface and during this process, the persistent Organic Phosphorus Pesticide is probably adsorbed with the particles and accumulates in the sediment (Tanabe et al.,

1991; Uluocak & Egemen, 2005). Benthic sediments have a high pesticide binding capacity in aquatic ecosystems. Therefore, since the pesticides bound by benthic sediments become stable in the ecosystem, they have a negative effect on aquatic organisms and increase the accumulation rate. Pesticides that enter the aquatic ecosystem in significant amounts through soil leaching accumulate in living organisms (Kemp, 2004; Radhiah & Rad, 1990; Yalvaç et al., 2004).

The two main mechanisms of pesticides in aquatic systems are biological concentration and biological increase. In the effect of biological concentration; pesticides cause a decrease in the number and diversity of aquatic invertebrates, DNA damage, and various mutations in cells. Biological increase is the accumulation of pesticides at each successive stage of the food chain. Even if a pesticide is found in small amounts in water, it will be retained by aquatic plants and eaten by insects and then fish. Pesticide concentration increases at each stage of the food chain in aquatic systems (Böke, 2012 ).

According to Yalvaç et al. (2004), Edwards (1973) stated that the rate of pesticide accumulation in aquatic ecosystems is higher than in terrestrial ecosystems. Organisms respond to pollution in two ways: acute and chronic. Acute effects occur in the form of serious damage or death shortly after the organism is exposed to high concentrations of the pollutant. Chronic effects become apparent as serious diseases (cancer, etc.) after a while after exposure to low concentrations of the pollutant (Williams & Feltnate, 1992; Atamanalp & Cengiz, 2002).

In an aquatic ecosystem, planktonic organisms, which are the main food source and have a very high biomass amount, experience mass deaths due to exposure to pesticide residues. These sudden collapses in the biomass of planktonic organisms also affect the high level of productivity of the aquatic ecosystem (Mane et al., 1986; Yalvaç et al., 2004). Chlorinated pesticides that do not dissolve in the aquatic environment accumulate in the fatty tissues of organisms, and this accumulation increases in intensity through nutrition, starting from single-celled organisms in the sea and reaching fish and birds that feed on fish.

### **9- Effects of pesticides on fish:**

The effects of pesticides on fish are seen in different ways. Fish can absorb pesticides from the water environment through their gills or consume contaminated materials as food or they can be poisoned. They can be killed directly or they can affect the fish population by stopping egg laying and reproduction. Fish are sensitive to the damage they cause to the tissues and are more affected by seasonal temperature changes and temporary hunger. Young

fish are damaged by this situation because they are sensitive (Toros & Maden, 1991; Atamanalp & Yanik, 2001). As a result of pesticides being carried to streams in various ways, they cause mass deaths of fish and other aquatic creatures as well as changing their living places. (Atamanalp et al., 2002).

The greatest danger in the use of pesticides occurs when they contaminate water resources. Heavy contamination of water with pesticides leads to oxygen deficiency and therefore poisoning. Because fish and other aquatic organisms have the capacity to absorb chemicals from water and accumulate them in their fatty tissues. This situation becomes even more important in fish that pass large amounts of water through their gills during respiration and in organisms that feed by filtering water. DDT and similar compounds prevent oxygen intake from the gills, leading to mass deaths of fish (Agnihotrudu, 1988; Atamanalp & Yanik, 2001; Figure 9).

Toxic chemical substances found in pesticides used in agricultural areas, liquid wastes of residential areas and industrial wastes accumulate in the body tissue of fish in water resources and become harmful or negatively affect human health through the food chain (Kırımhan et al., 1984; Atamanalp & Cengiz, 2002).



**Figure 9.** Fish deaths affected by marine pollution

### **10- Effects of pesticides on humans:**

Pesticides reach the human body indirectly by entering the body of animals in feeds or directly through plants containing pesticide residues in the land environment. The fact that humans are living beings that feed on both animal and plant foods and are the last link in the food chain causes such compounds to be reflected in humans to a large extent (Egemen, 1990; Balcı & Gedikli, 2011). Pesticides can also enter the human body through direct skin contact. Ingested pesticides can spread throughout the body through the bloodstream, accumulate in internal organs, fat or bones, or be excreted through urine or feces in a short

time. However, when pesticides spread throughout the body, various side effects such as skin burns, poisoning and cancer can be seen. Due to all these side effects on both the environment and humans, it is of great importance to determine the amount of pesticide residues with accurate and sensitive methods (Karakoç & Nakiboğlu, 2010).

Organophosphorus compounds can be absorbed through the respiratory and digestive systems, as well as through the skin to a significant extent. The most important feature of organophosphorus insecticides is that they inhibit the cholinesterase (ChE) enzyme, which plays a role in the functioning of the nervous system, causing irreversible acute and chronic poisoning (Vadhva & Hasan, 1986). Therefore, organophosphorus drug poisoning can be revealed by measuring the cholinesterase enzyme level in the blood. Symptoms such as respiratory difficulties, cough, runny nose, chest tightness, burning and watering of the eyes, insomnia, and distraction occur at enzyme levels that are 20-25% below normal (Vural,1996b; Yalvaç et al., 2004). Organophosphorus insecticides can also cause a decrease in antioxidants because they increase the formation of free radicals. Especially in individuals with low cholinesterase levels, there is likely to be an increase in lipid peroxidation and a decrease in total antioxidant capacity, disruption of the erythrocyte membrane structure, and an increase in oxidative stress can be characteristically observed due to the inhibition of acetylcholinesterase (Datta et al., 1994; Hai et al., 1997; Yalvaç et al., 2004).

Increased pesticide consumption due to excessive and unconscious use has led to various problems in terms of human health along with environmental pollution. These problems are listed below:

- Pesticides cause cancer, birth abnormalities, nervous system damage and long-term side effects,
- Pesticides and their breakdown products contain toxic substances,
- Some of their breakdown products are more toxic and permanent than the main pesticide.
- Depending on the pesticide applied and application conditions, it causes environmental pollution.
- Those that can evaporate excessively pollute the air we breathe.
- Excessive use creates resistance to the drug in organisms, and pesticide application fails.
- It kills the natural enemies of targeted and untargeted pests and beneficial organisms, creating new epidemics (Delen et al., 2005; Yeşil & Öğür, 2011).

The most important problem with pesticide use is the difficulty of determining exactly when, where and how it interacts with the environment and the damage it can cause to humans and nature over a long period of time. For this reason, restrictions have been imposed on the use of synthetic pesticides, which are difficult to break down in nature and take a long time, within legal frameworks (Klinghard, 1995; Yalvaç et al., 2004).

Various studies have shown that pesticides, which have a market size of 130.7 billion dollars worldwide, leave residues in our food and cause serious health damage, as well as permanent damage to the environment. According to the results of the research, it is stated that approximately 385 million pesticide poisoning cases occur in the world every year and 11,000 people lose their lives directly due to these poisonings. It is stated that those who are heavily exposed to pesticides experience heart, lung or kidney failure. It is also emphasized that pesticides cause serious increases in Parkinson's, leukemia, lung and breast cancer, type 2 diabetes, asthma, allergies, obesity and hormone disorders in the world (Stiftung, 2023).

### **11- Safe and effective application conditions of pesticides:**

As an integral part of the agricultural system all over the world, the use of pesticides is an issue that should be carefully considered due to the risk of residue in agricultural products and the negative impact it has on the environment. Different precautions should be taken against the negative effects of drugs used in agriculture in terms of human and environmental health (Durmuşoğlu et al., 2010). The most appropriate conditions for the use of pesticides in terms of the environment and residue and the points to be considered can be summarized as follows (Tiryaki, et al., 2010):

- In chemical control, pesticides with low toxicity to human health and non-target organisms should be applied.
- Pesticides with the lowest possible risk to the environment and organisms living in the environment should be selected.
- Pesticides with low resistance risk in harmful organisms should be selected.
- The drug that causes the least harm to natural enemies should be selected. Beekeepers in the vicinity should be warned, and hives should be closed for a while.
- The most effective spraying time should be selected according to the biology of the targeted pest.

- Application should be made by selecting the most effective method and taking maximum protection measures. A pesticide with a strong initial effect and shorter persistence should be selected.

- Attention should be paid to the period between the last spraying and harvest.

- The chemical and water mixture should be made at the application site. Maintenance and calibration of the application tool should be performed.

- Pesticide application should be made in the recommended dose and number.

- Necessary precautions should be taken before application and applicators should be trained.

- Agricultural pesticides with damaged packaging should not be purchased and they should be kept in safe places away from children, food and drink.

- Application should be made in suitable weather conditions, windless weather and in the cool hours of the day.

- Nothing should be eaten or drunk during the application, eyes should not be rubbed, mouth should not be touched, clothes should be changed after the application and hands and face should be washed with plenty of soapy water.

- Applications should not be made by children and other workers should be kept away from the application area.

- Farm animals should be kept away during the application and animals should not be allowed into the area where the application is made before a certain period of time has passed.

- Used tools and auxiliary containers should be washed thoroughly after the job is finished and empty drug containers should be disposed of appropriately

The most important application area of pesticides is their use as “agricultural warfare drugs” and in our country, 1483 formulations containing approximately 346 pesticide active ingredients are used for agricultural purposes. While occupational poisoning and death may occur due to failure to take protective measures or incorrect applications during pesticide use, non-occupational poisoning is also seen in cases of use at home or for purposes other than intended. Although most of the poisoning cases that occur are accidental in origin, their use for suicide and homicide purposes is also not negligible due to their easy availability (Akçan, 2008).

## **Conclusion**

Pesticides are chemical substances that have high activity against harmful organisms. Therefore, they are used to destroy and prevent agricultural and household pests. Pesticides are substances that are effective in treating plant

diseases, purifying the product from weeds and harmful insects, and fighting against forest pests, vectors such as houseflies, mosquitoes, bedbugs, fleas, lice, and bacteria and fungi. In addition, they are widely used because they provide rapid results when used consciously and in a controlled manner and can also protect the product from toxin-secreting organisms.

The behavior of pesticides in soil and natural waters varies according to their chemical structures. In order to determine the accumulation, decomposition and movement of pesticides in soil, it is necessary to reveal some of their characteristic features and reaction interactions. Pesticides mixed into water and soil environment directly come into contact and interact with microorganisms. The physical and chemical qualities of water, soil and light energy also participate in this relationship. Some studies have shown that different types of microorganisms affect pesticides, disrupt their chemical structures and cause the formation of different compounds (Demireken, 2001). The most important feature sought in pesticides is that they should be highly toxic to harmful animals and insects, but slightly toxic or non-toxic to mammals and humans. However, the vast majority of pesticides are harmful and toxic to both living things and humans. Some pesticides are poisons that can remain intact in the plant, soil and water environment where they are applied for years and can accumulate in the bodies of all living things (Güney, 1992; Ağça, 2006).

The areas of use of pesticides are increasing day by day due to developing technology. Despite their harmful effects, the use of pesticides is an inevitable necessity both in agricultural areas and other industrial products. Necessary precautions should be taken to keep the side effects of pesticides at a minimum level. Therefore, during the use of pesticides, attention should be paid to using pesticides that can decompose quickly and have less permanent properties, instead of pesticides that have a high permanent effect and a high ability to accumulate in foods. In addition, it is important to adjust the amount of use of these substances well in order to reduce toxic effects.

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## Chapter 5

### Adaptation Challenges: Wheat Production Under Changing Climatic Conditions In Pakistan

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## ABSTRACT

Climate change threatens wheat, one of Pakistan's staple crops and a food source for most of its people. This chapter describes how altered weather influences wheat production and highlights key barriers to adaptation. It provides an in-depth analysis of climate trends, their impact on wheat production and phenology, as well as the associated social and economic consequences. This chapter discusses potential adaptation strategies to enhance resilience in wheat production systems, with an emphasis on the importance of farmer participation, policy support, and innovative research in this effort. Climate change is described as a long-term change in global climate including rising temperatures, precipitation, and wind patterns. In agriculture, this could have a direct impact on food production levels. The farming community across the globe witnesses across the board slight negative and incremental impact on available land for farming. This paper specifically focuses on the agricultural investment climate of Pakistan which has been changing precipitation patterns with an increase in the frequency of extreme events alongside increased global temperatures. The study which combines crop life cycle and regional climate models details the location-specific climatic risks and vulnerabilities of wheat crop and how these could severely drop the tonnages in a dryland environment. This research aims to detect climatic extremes around the world and create a visual representation of climatic extremes throughout time. Average international temperatures have risen by 0.6°C since the onset of the commercial Revolution, as reported by the Intergovernmental Panel on Weather Alternate (IPCC). They are projected to heat any other 1.4°C to 5.8°C by using the end of the 21<sup>st</sup> century. various areas have separate climate results, with dryland regions especially threatened consistent with weather estimates, which might result in unique yield consequences in the agriculture spectrum. There is still a major problem with water shortage, although increased atmospheric CO<sub>2</sub> concentrations to some upward effects on some negative consequences of warming temperatures. Ensuring adequate irrigation may ease some of the pressures from the environment. Given Pakistan's changing environment, this study highlights the crucial need for adaptable approaches to preserve wheat production and provide food security.

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## INTRODUCTION

It is the main staple food of Pakistan and plays a vital role in the contribution of agricultural GDP of Pakistan. But a major challenge to its production has emerged: climate change. Traditional farming practices are being disturbed by rising temperatures, erratic rainfall patterns, and an increase in the frequency of extreme weather events. To guarantee food security and the livelihood of millions of farmers, these issues must be resolved. Pakistan's environment is conducive to the cultivation of a wide range of crops. Wheat, a staple crop that is grown on more than two-thirds of farms throughout the winter (Rabi) and makes up almost one-third of the nation's total farmed acreage. As a result, wheat production levels not only affect the food security of millions of farmers and landless rural residents, but it also serves as a significant source of income for them (Mauseth, 2014).

Wheat may be processed into several special items. When raw wheat is beaten and divided into bits, the outer husk, or grains, serves several functions. Wheat is a fundamental ingredient in a huge variety of meals, together with bread, porridge, crackers, biscuits, pancakes, pastries, pizza, cakes, cookies, cakes, rolls, doughnuts, and morning cereal. It's also been discovered in beer, vodka, boza (a fermented drink), and gravy (Food Allergic Reaction Canada, 2019). Gluten is beneficial in the production of wheat merchandise because it imparts viscoelastic useful houses to the dough, permitting the fabrication of a ramification of processed meals along with bread, noodles, and pasta that promote wheat consumption.

Weather is the temporary condition of the atmosphere, whereas climate is the long-term state of the atmosphere. Fossil fuels have been sought to meet energy needs while maintaining growth rates. However, the combustion of fossil fuels discharges gases into the atmosphere, which gradually affects the climate over time. However, rising temperatures, erratic rainfall patterns, and extreme weather events make the country's wheat production more vulnerable to climate change.

The purpose of this chapter is to examine the possibilities and challenges of adapting wheat production to these changing environmental conditions. According to research, Pakistan has already tapped into the Green Revolution's technical potential, and the wheat crop is under pressure from climate change; production growth rates have plummeted from over 7.2% per year in the early 1970s to under 2% in recent years. Sivakumar and Stefanski (2011) indicated that a temperature increase of 10 °C will lower wheat output by 5.7% in Pakistan, whereas Leads (2009) predicts a more than 40% loss in wheat yield by 2035.

## Worldwide wheat production and consumption

In 2022, worldwide wheat output would top 808.4 million tonnes, with China, India, and Russia accounting for 43.22%. Wheat was planted on 220.7 million hectares of land globally in 2021, making it the most significantly grown food crop. the worldwide wheat change outpaces all other vegetation combined. Wheat demand is growing across the world due to its unique and sticky proteins, which can be required for processed foods manufacturing (Shewry et al., 2015).

## Wheat production in Pakistan

In Pakistan, wheat was cultivated on 9.6 million hectares during 2023-24, marking a 6.6% increase compared to the 9.0 million hectares sown the previous year. Wheat production also rose significantly, from 28.2 million tons to 31.4 million tons, reflecting an 11.6% increase (Figure 2). The government set the minimum support price at Rs 3,900 per 40 kg for the 2023-24 season. Despite this growth, a drought in December 2024 poses potential risks to yields. Nevertheless, favorable conditions during the sowing season—such as optimal weather, adequate soil moisture, availability of inputs, and an expanded sown area across provinces—contributed to promising production even under challenging harvest conditions. Wheat remains a vital crop for Pakistan, contributing 9.0% to agriculture and 2.2% to GDP.

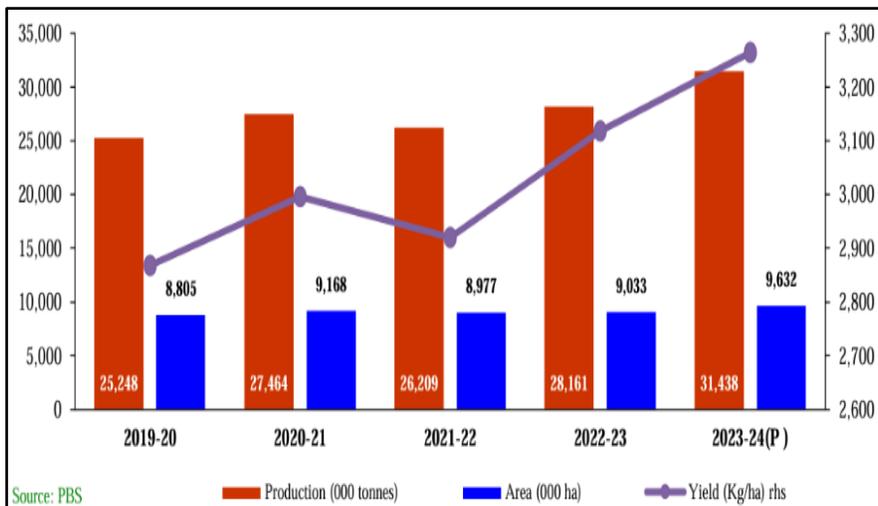


Figure 2. Wheat Production During 2023-24 in Pakistan

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## VULNERABILITY OF WHEAT TO CLIMATE CHANGE

### Definition and Causes

Climate change is defined as longterm changes in climate patterns due to human or anthropogenic activities (IPCC, 2007). Greenhouse gases (GHG), consisting of water vapor, carbon dioxide (CO<sub>2</sub>), methane (CH<sub>4</sub>) and nitrous oxide (N<sub>2</sub>O), are important factors in global warming. These gases are released as a result of various human activities:

- **CO<sub>2</sub>**: Emitted during the combustion of waste, coal, wood, and fossil fuels.
- **CH<sub>4</sub>**: Generated during the extraction and transportation of coal, gas, and oil.
- **N<sub>2</sub>O**: Produced during industrial and agricultural processes.

considering that pre-industrial times, human activities like deforestation and the sizable use of fossil fuels have raised CO<sub>2</sub> tiers from 280 ppm to 380 ppm (Stern, 2006). presently, GHG concentrations are increasing at a price of 23 ppm in keeping with decade, the best upward thrust in 6.5 million years. The electricity sector is the most important contributor (63%), accompanied with the aid of agriculture (13%), land use and forestry (18%), enterprise (3%), and waste control (3%) (Rosegrant et al., 2008).

### Rising Temperatures and Impacts on Agriculture

The average worldwide temperature has risen by 0.6°C because the industrial Revolution, mainly due to increasing GHG concentrations (Brohan et al., 2006). considerably, the 20th century become the warmest on document, with the Nineties being the hottest decade of the millennium. Projections advocate a temperature rise of one.four°C to 5.8°C at some point of the 21st century (IPCC, 2001).

These temperature increases have profound effects on agriculture:

- **Dry Spells and Heat Waves**: Prolonged dry periods and intense heat waves can severely reduce agricultural productivity, particularly during critical growth stages, by inducing heat or moisture stress (Rosenzweig et al., 1994; IPCC, 2001).
- **Regional Vulnerabilities**: Arid and semi-arid regions, such as those in South Asia, are especially vulnerable due to existing water shortages and high baseline temperatures (CGIAR, 2004–2005).

### Wheat: A Case Study

Wheat is highly sensitive to temperature changes, particularly during the grain-filling stage. High nighttime temperatures reduce starch accumulation, negatively affecting yield and quality. The shortening of the crop life cycle due to rising temperatures is another critical impact:

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### 1. **Shortening of Crop Life Cycle:**

Hussain and Mudasser (2007) reported that for every 1°C rise in temperature, the crop life cycle was shortened by approximately:

- 4 days in arid zones
- 5 days in semi-arid zones
- 6 days in sub-humid zones
- 9 days in humid zones

Total reductions in the crop life cycle compared to the baseline were 12, 14, 18, and 27 days, respectively.

### 2. **Growing Degree Days (GDDs):**

Increased temperatures accelerate GDDs, which influences phenological development. While this shortening can decrease yield potential in some regions, it can also enable the cultivation of successive crops in regions where wheat takes longer than optimal to mature.

### 3. **Regional Yield Gains:**

In high-altitude mountainous areas, a temperature rise of up to 5°C has been associated with yield gains of up to 20%, despite a reduced crop life cycle. In humid zones, warmer temperatures mitigate cold stress, supporting higher yields (Hussain and Mudasser, 2007; O'Brien, 2000).

## **Future Projections and Adaptation**

The IPCC predicts that while global warming trends will persist, regional variations in temperature and precipitation will result in diverse socio-economic impacts. Sectors such as water resources, agriculture, health, forestry, and biodiversity will experience both positive and negative effects. Tailored adaptation strategies are essential to mitigate these impacts and ensure food security.

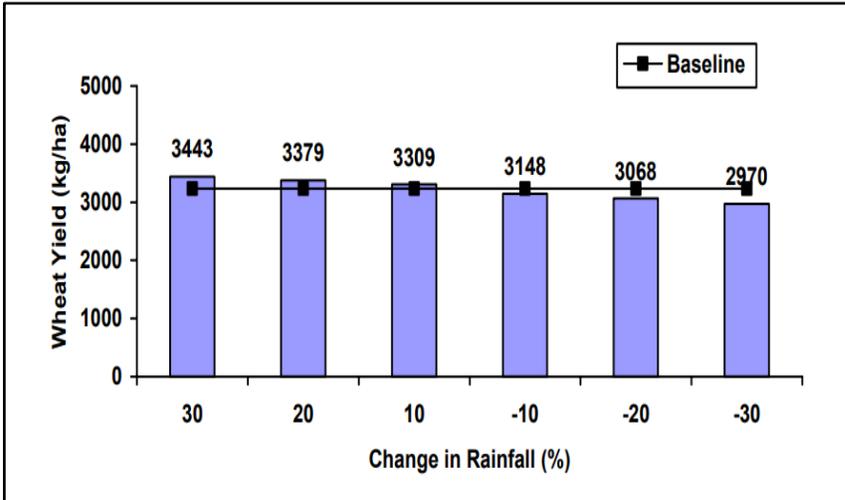


Figure 2. Impact of assumed changes in rainfall amounts on wheat yield

## Impact of Climate Change on Wheat Germination and Growth

### Impact on Germination

#### *Temperature and Germination Rates*

Wheat germination is highly sensitive to temperature, and rising global temperatures significantly impact this critical growth stage. Optimal germination occurs between 10°C and 20°C, while temperatures exceeding 30°C impair germination by reducing seed viability and slowing metabolic processes. Research by Reynolds et al. (2009) revealed that wheat exposed to temperatures above 30°C during germination exhibits reduced germination rates and poor seedling establishment. In Pakistan, regions such as Sindh and Punjab have reported reduced germination during heat waves, exacerbated by increasing average temperatures (Janjua et al., 2024).

#### *Heat Stress and Germination*

Wheat germination is interrupted with the aid of recurrent and excessive warmth waves, which lowers the capacity yield and outcomes in terrible seedling formation. consistent with Lobell (2011), mainly in areas like Pakistan, warmth pressure during the germination period can lower crop yields by as much as 30%. Heatwaves for the duration of the 2015–16 wheat sowing season reduced germination fees and yields in Punjab, according to the Pakistan Meteorological department (PMD).

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### ***Water Stress and Germination***

For wheat to germinate successfully, water availability is important. better evaporation quotes and altered precipitation patterns introduced on with the aid of climate alternate use up soil moisture, which prevents germination. in line with Timsina et al. (2017), a ten% lower rainfall in the course of South Asia's wheat growing season ends in a 15-20% drop in germination success. inadequate rainfall and growing evaporation make germination extra difficult in Pakistan, wherein water shortage is already a good-sized hassle.

### ***Delayed Planting and Early Germination***

Adjustments in sowing schedules delivered on by high warmth or postponed rainfall provide less-than-perfect germination situations. Crop establishment is diminished by way of overdue planting, which often exposes seeds to excessive warmness or dryness. in line with Zhao et al. (2017), heat pressure throughout germination is the main reason why behind-schedule planting may drastically reduce wheat yields in South Asia by 2050.

### ***Impact on Emergence and Early Growth***

#### ***Delayed Emergence and Reduced Seedling Survival***

Water pressure and excessive temperatures can preclude or delay the sprouting of wheat seedlings. Lobell (2011) pointed out that better temperatures inside the early levels of development sluggish down emergence rates, particularly when there may be water scarcity. In Punjab, fewer flora according to unit place and worse seedling survival prices have resulted from reduced water availability after planting (Awan et al., 2019).

#### ***Impact on Tillering***

One crucial improvement stage that influences potential output is tillering. although higher temperatures hasten plant improvement, they can also decrease the overall boom and the number of tillers. in particular, in warmness-touchy wheat types, Porter and Semenov (2005) discovered that warmth stress for the duration of the early vegetative levels outcomes in fewer tillers. In Punjab and Sindh, warmness stress has continuously reduced the number of tillers, which has an unfavorable effect on potential yield (Imran et al., 2020).

#### ***Days to Tillage, Anthesis, and Maturity***

Warmer temperatures might also divulge crops to heat pressure throughout vital reproductive stages because they decrease the length before tillering and anthesis (flowering). according to Raza et al. (2021), higher temperatures

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shorten the grain-filling period and yield by speeding up blooming. Likewise, inadequate grain filling brought on with the aid of early adulthood added on with the aid of warmness stress might reduce yield and quantity (Challinor et al., 2009).

### **Impact on Yield and Yield Components**

#### ***Temperature and Yield***

Overall yields are reduced by high temperatures during anthesis and grain filling because they inhibit fertilization and cause smaller grains. According to Zhao et al. (2017), for every  $^{\circ}\text{C}$  that the temperature increased in these stages, the yield decreased by 10%. Heat stress during annealing and grain filling is associated with reduced wheat yields in Pakistan, particularly in Sindh and Punjab (Imran et al., 2020).

#### ***Drought and Water Scarcity***

Yields are greatly reduced by water stress during germination and grain filling. According to Timsina et al. (2017), there is a 15% decrease in yield when water availability is reduced by 10% to 20% during the wheat growing season. Lower yields and shriveled grains are the result of insufficient moisture at critical stages.

#### ***Yield Projections Under Climate Change***

Climate models predict a  $1.5\text{--}2^{\circ}\text{C}$  temperature increase in Pakistan by 2050, exacerbating heat stress and water scarcity. Imran et al. (2020) estimated a 30% decline in wheat yields by 2050 when temperatures rise by  $2^{\circ}\text{C}$  due to heat stress, changing rainfall patterns, and reduced water availability. Additionally, climate scenarios such as RCP 4.5 and RCP 8.5 predict an average decrease in yields of 1% and 2.5% by 2050, respectively.

### **CONCLUSION**

Beyond weather, trade has had a predominantly bad effect on wheat yield, with future scenarios indicating an intensification of this sensitivity. consequences show that even a  $1^{\circ}\text{C}$  upward push in temperature will adversely affect wheat yield. at the same time an increase in atmospheric  $\text{CO}_2$  concentration (from baseline to 550 ppm) could partly offset those poor results, supplying an internet superb effect up to a  $2^{\circ}\text{C}$  temperature upward push, any further growth might result in overall yield discounts.

In Pakistan, climate change impacts wheat production at every stage of development, from emergence to maturity. Factors such as increased temperatures, water stress, and erratic rainfall patterns have resulted in reduced

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plant populations, fewer tillers, and lower grain counts. These conditions accelerate flowering and maturity, further reducing yields. Projections suggest that wheat production could decline by up to 30% by 2050 if current trends persist.

Farmer perception studies conducted by the Climate Change Centre at the University of Agriculture Peshawar highlight that farmers are acutely aware of changing rainfall patterns and rising temperatures. However, these changes remain underappreciated by planners and researchers, putting the livelihoods of small-scale farmers at significant risk. Rainfed agriculture, which constitutes nearly half of Khyber Pakhtunkhwa's (KP) cultivated area, is particularly vulnerable. In recent years, wheat has not been grown in some rainfed regions due to delays in winter rains, while summer heat has exacerbated water demand, leading to severe shortages in both surface and groundwater supplies.

The national wheat crop yield has also suffered, with unfavorable weather conditions during the 2018-19 Rabi season causing a shortfall of 1.5 million tons from the 25.5 million tons target. Rainfed regions in KP, excluding the cold temperate areas of Upper Dir and Shangla, have seen significant reductions in wheat cultivation, with some areas unable to grow wheat at all in the past two to three years. Adaptation strategies, such as late-season wheat varieties, are urgently needed to address these challenges.

Surface water sources, particularly spring water, are drying up, and groundwater levels are declining rapidly, with central and southern KP districts experiencing drops exceeding 200 feet. Livestock, another critical component of agriculture, faces challenges from climate-induced diseases and reduced feed availability, further threatening agricultural livelihoods. Climate analyses by the Meteorology Department in Peshawar indicate a 25% decrease in winter rainfall and a 40% increase in summer rainfall over the last two decades, along with a 10% rise in day and night temperatures. These changes have adversely affected the yield of fruits and crops, particularly in Peshawar and surrounding areas. Deforestation, unplanned urbanization, agricultural land conversion, and air pollution have exacerbated these trends, with temperatures in some areas rising by 0.5°C per decade.

Adaptation measures must prioritize developing climate-resilient germplasms, adjusting crop sowing times and methods, and implementing soil moisture conservation, rainwater harvesting, and water use efficiency. Climate-smart agriculture practices and district-specific climate adaptation plans, particularly for soil and water conservation, are essential. Redefining agro-climatic zones will also help optimize resource allocation and agricultural practices.

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The recent shift in rainfall patterns during wheat harvesting seasons in KP, coupled with fluctuating temperatures, has caused severe yield losses due to disease outbreaks and poor grain quality. These changes underscore the urgent need for holistic approaches to mitigate the impacts of climate change on agriculture in Pakistan.

### **RECOMMENDATIONS**

- Resistant improved wheat varieties should be planted.
- Cultivation of early sowing and early maturing varieties.
- Planting should be finished before November, 30 to reap maximum yield in KP province.
- Treat the seed with recommended fungicides to manipulate seed-borne illnesses.
- Moisture should be conserved throughout monsoon via the usage of deep tillage era.
- Avoiding thick sowing and heavy irrigation.
- Irrigation during tillering and grain development is critical.
- Use nitrogen and phosphorus fertilizers in a 1:1 or 1:1-1/2 ratio.
- When planting wheat after rice or sugarcane on sandy soil that has been watered with tube well water, it is necessary to apply K fertilizer.
- Destroying the weed plant life and diseased tillers.
- Averting heavy doses of nitrogenous fertilizers.
- Judicious use of k-fertilizer helps in minimizing the susceptibility of plant life.
- To empty extra water from the fields to save your crop from being lodged and getting yellowish.
- If rains prolong then there could be chances of (attack of) yellow rust. It is highly recommended to apply fungicide to reduce infestation. This will reduce the chances of wheat grain yield reduction.

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## Chapter 6

### The Use Of Insects As Natural Dyestuffs

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**Keywords:** *Carminic acid, Dactylopius coccus, Dactylopius opuntiae, Kermes vermilio, kermesic acid, Kerria chinensis, Kerria lacca, Porphyrophora hamelii, Porphyrophora polonica*

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## Introduction

The history of colour is as old as human history. Colour is one of the elements of nature that make man's life on earth more aesthetic and fascinating. In the past, people have tried to decorate their livelihoods in a similar way to the natural colours observed in plants, soil, sky and other sources. The use of colour seems to have fulfilled the need of human spirituality since ancient times. Whatever the culture of the human being, the use of colour is universal. Early humans began to use colourants of plant, mineral or animal origin that they found in nature for purposes such as protective coating, ornamentation and marking. It was also common practice to apply these colourants to their bodies as a sign of attractiveness, intimidation against enemies, special occasions, or belonging to a group or tribe. The increase in the types of colours that could be obtained and the increase in manual skills led people to use paints to paint what they saw around them and to create artistic products. The archaeological literature has associated the use of natural dyes with different types of aesthetic, symbolic and/or artistic practices. These include different types of markings, paintings and drawings, deliberately made on the walls of caves as well as on different types of portable objects.

Natural dyes, which is an application as old as human history, are dyes made with the use of living organisms such as various plants, insects, shellfish, fungi and even lichens. Colourants are used as a name for materials such as dyes and pigments. It has been known since ancient times that natural colourants can be used as natural dyes. Dye is generally used as an aqueous solution. It may require a mordant to improve the fastness of the dye to the fibre.

The most common natural dyes include *Tyrian purple*, *cochineal*, *madder* and *indigo*:

In ancient times, natural dyes were considered a luxury due to the difficulty of obtaining the material to produce them. For example, the colour purple was derived from a mollusc and was very complex to obtain. *Tyrian purple*, obtained from sea snails in the ancient city of Tyre (an important Phoenician city) in the eastern Mediterranean, was called royal purple or imperial purple. By 1500 BC, the Phoenicians had a thriving Tyrian (royal purple) dye industry in Tyre and other cities. Tyrian purple was very special to all around the Mediterranean civilisations and its use over centuries. Because the colour it produced was very bright and colourfast this it was the most expensive dye in the entire ancient world. Due to its properties, its use was limited to royalty, members of the high-ranking public officials, royal family, and priests. To produce one gram of dye, 8,500 molluscs were needed. Purple was the colour of high achievement, ostentatious wealth, symbolic sovereignty and the highest positions in the legal

system. From Tyre, the colour was introduced to the ancient Romans and Greeks, and through them to Byzantium and medieval Europe. The trade, which fetched a higher price than gold around 1500 BC, brought great prosperity to Lebanon, where Tyrian purple was obtained (Puntener and Schlesinger, 2000; Zhang and Laursen, 2005).

In South America, ancient Peruvian textiles dating back to 2500 BC bear traces of natural dyes including the use of plant-based dyes such as cochineal. *Cochineal* (*Dactylopius coccus*), an animal-based natural dye, as in Tyrian purple, was introduced to Europe from Mexico by the Spanish. Cochineal, used as a fabric dye and much later as a food and cosmetic dye, produces about 30 g of dye from 17,000 dried insect individuals (Lucas and Harris, 1999; Crowfoot, 1944).

Plant dyes, on the other hand, are generally cheaper and more readily available. The most common are *madder* and *indigo*. Madder is derived from the roots of 35 plant species found in Europe and Asia. Madder was even found in the fabrics of mummies and was the first dye used as camouflage. One of the main groups of yellow natural dyes is obtained from plants containing flavonoids. Flavonoids, recently well known for their antioxidant role in human health, are a type of natural product (Harborne, 1986; 2000). Again, the nails of Egyptian mummies were painted with henna leaves of *Lawsonia inermis* (Golyon, 1996; Redford, 2001; Van et al. 1994).

Some regions, such as Central Asia, Japan and India, historical records and artefacts also indicate the use of natural dyes for textile colouring. The presence of plant-based dyes has been attested in these regions. Archaeological findings from various regions provide information on the early use of natural dyes. Evidence of dyed fabrics has been found in tombs and burial sites, including pieces of linen and wool dyed with plant-based dyes such as indigo, root dye and safflower in ancient Egypt. Similarly, dyed silk fabrics have been discovered in tombs dating from 202 BC to 220 AD, with evidence of the use of plant-based dyes such as indigo in ancient China. In Europe, the use of natural dyes dates back to ancient civilisations such as the Celts, Greeks and Romans. In Europe, Italian dyers, especially those in Venice and Tuscany, were considered the best dyers from the Roman Empire until the sixteenth century (Gittinger, 1982). During this period, plant-based dyes were widely used, with sources such as *Rubia tinctorum* (madder) for reds, *Isatis tinctoria* (indigo) for blues and *Reseda luteola* (dyer's rocket) for yellows.

This study discusses the history of colours and natural dyes and explains the use of insects as natural dyes, especially in the production of red colour, which plays an important role in natural dyeing.

## **The First Use of Colour and Its History**

Colour, which has a wide historical development in the world, has been the favourite of many civilisations and has been used in many fields from architecture to art. Although some previous studies have estimated that the first examples of paint began to be used around 15,000 BC, recent scientific studies have proven that the use of colour pigments goes back much further. For a long time, this type of artistic production was thought to be mainly related to anatomical modern humans (*Homo sapiens*). However, the discovery of the earliest examples in different parts of South Africa (Blombos Cave), dated to between 100,000 and 85,000 years ago, containing several fragmented ochre pebbles and deliberately cut paint, extends the dating of this paint much further back (D'Errico et al. 2012; Henshilwood et al. 2009). In addition, 73,000-year-old red diagonal line traces found on a rock fragment in Cave Blombos (Henshilwood et al. 2018); 77,000 years old wall painting decorated with the colour obtained from seashells (Henshilwood et al. 2004), Sibudu and Border Cave and other African regions or artistic works dating back at least 40 and 45,000 years in Spain (Hoffman et al. 2018a; Pike et al. 2012) and Indonesia (Aubert et al. 2014; Brum et al. 2021) decorated with ochre are remarkable. The Altamira Cave, also in Spain, and the Lascaux Cave in south-western France are estimated to date back to 17,000 years ago, with around 600 paintings depicting scenes from nature. In these paintings, iron oxide was used as the paint material and the paints were applied without any binder.

Considering the time and effort required to process colouring raw materials, it is clear that its use well-planned activity was not accidental since prehistoric times. Examples such as Cave Blombos in South Africa (Henshilwood et al. 2011), Cave Qafzeh in Israel (Hovers et al. 2003), Cave Porc-Epic in Ethiopia (Rosso et al. 2014), Santa Maira in Spain (Tortosa et al. 2020; Domingo et al. 2012) and Cova de l'Or (Domingo et al. 2012) prove the use of natural colourants in prehistoric times (di Lernia et al. 2016).

Archaeological research has provided evidence for the use of natural minerals and dyes with colouring properties before the global spread of *Homo sapiens*. (Watts et al. 2016; Wolf et al. 2018). The presence of pigment fragments, pigment powders and residues and even paint drops on materials such as shells, stone and bone tools, grinding stones, different types of vessels obtained as a result of archaeological researches is evidence that *Homo sapiens* used natural paint in prehistoric times. The tradition of using natural dyes to produce different types of art in different media, such as bone, stone, horn, pottery, etc., has spread throughout the world over time.

Red paint blown with a mouth or reed-like object on a stalagmite dome at Ardales in Málaga, a mould of a hand at Maltravieso, Cáceres, and a painting resembling the lines of a staircase at La Pasiega, Cantabria, are dated to at least 64 to 66,000 years ago (Hoffman et al. 2018a, Zilhão 2020). However, it is interesting to note that modern *Homo sapiens* had not yet reached the Iberian Peninsula. This suggests that these depictions were produced by Eurasian Neanderthals. Interestingly, various researchers have reported that Neanderthal man (*Homo neanderthalensis*), a human species that lived between 250,000 and 40,000 years ago, used colouring agents as well as personal ornaments containing red colouring residues. For example, Zilhão et al. (2010) interpreted a mixture of coal, dolomite, haematite and pyrite, as well as yellow natrojarosite associated with seashells dating back 115,000 years, from Cueva de los Aviones (Murcia, Spain) as potentially used for cosmetic purposes (Hoffman et al. 2018b). Soressi and d'Errico (2007) suggest that manganese dioxide from the French Mousterian sites of Pech de l'Azé I and Pech de l'Azé IV may have been used for body painting (Soressi and d'Errico, 2007). The discovery of drops of a haematite-rich liquid of unknown use in sediments from the Maastricht-Belvédère deposits in the Netherlands indicates that this mineral was used by early Neanderthals until at least 200-250 thousand years ago (Roebroeks et al. 2012). Although it is difficult to know whether the use of different pigments was practical or symbolic, it has been proven that Neanderthals were already using mineral colouring materials for many activities such as leather tanning and tool polishing long before the arrival of *Homo sapiens* in Eurasia (Hovers et al. 2003).

There are many studies showing that the use of pigments used in colour extraction as natural dyestuffs in prehistoric times was preferred by humans not only for artistic purposes but also for their protective, antiseptic, corrosive, repellent, preservative and even medicinal properties (García-Borja et al. 2004; Hovers et al. 2003; Roebroeks et al. 2012; Rifkin et al. 2015; Soressi and d'Errico, 2007; Velo, 1984).

In the early 1980s, Couraud (1988) suggested the possibility of three different uses of colouring raw materials in the European Palaeolithic period, which were eventually transferred to other regions and periods: (i) the production of different forms of artistic expression, (ii) domestic and functional uses, and (iii) funerary practices. Many researchers have reported that colouring agents were also found in funeral rites from the Middle Palaeolithic to the Neolithic and beyond (Bar-Yosef 1997; Cacho et al. 1996; d'Errico and Backwell, 2016; Formicota 1986; Goren et al. 2001; Molleson et al. 1992; Stringer and Gamble 1996). Based on the different uses identified in the

cemetery contexts of Çatalhöyük, a Neolithic and Chalcolithic site in central Anatolia, Schotsmans et al. (2020) suggest that pigments were probably used for identity as well as social differentiation.

Today, paints are widely used even on carpet and rug in much larger quantities and with an increased number of colour shades (Genç, 2015). Protecting, beautifying, colouring, differentiating or warning against a danger can be given as examples of the areas of use of paints. In addition to the aesthetics and beauty that paint adds to the human soul, it is estimated that the first realisation of its protective effect and its use for covering in a protective sense took place in the Egyptian geography. The Egyptians aimed to protect their ships from external influences by painting them with pitch and pelesenk tree resin. The use of paint in a protective sense has only become widespread since the 18th century, and ancient civilisations have benefited from natural materials and recipes for paint for centuries. Such practices continued until the discovery of synthetic dyes in the 19th century.

### **First Use of Natural Dyestuffs and Its History**

The custom of our ancestors to bury their dead in piles of red-coloured soil dates back to the Old Stone Age (30,000 BC). The use of mineral-coloured soils in caves dates back to 15000 BC. These paintings made on cave walls using minerals strongly suggest that our ancestors were as artistic as they were practical.

Although it is thought that there was no need for dyeing when mankind started to produce clothes from linseed and cotton, it is very difficult to give answers to the questions of when the dyeing of textile products actually started and what were the first dyes (However, the existence of fibres at that time is known for certain). Animal fur and wool were always a resource that people could access after hunting (Genç & Koyuncu, 2017; Koyuncu & Genç, 2017). During that period, wild cotton was native to the American Southwest, Mexico, Central and South America, Africa, Iran and India. Mexican Indians were growing cotton for textile purposes as early as 5000 BC (Hochberg, 1983). Excavations at Mohenjo Daro in the Indus Valley of India have unearthed fragments of cotton fabric, dyed by complex processes, dating to about 2000 BC. However, it is still assumed that all clothing at that time was used in their natural colours of pale grey or near white. After centuries of use of these textiles, as early civilisations developed in the Middle East, Egypt and Asia, the need to differentiate between gender and class became even more apparent, and thus the first natural dyes appeared. Researchers have found proof of the first natural red and orange colours in tombs dating back to 2600 BC. Burial remains

from Egypt and Peru show that many ancient civilisations had excellent experimental dye technologies.

The first colours used in textiles were probably little more than stains, except for stable iron rust yellows and reddish oranges, bark tannin earth dyes, light browns, iron tannin greys and blacks. These stable dyes remained in limited use until the early twentieth century. The bright yellows and yellow oranges from turmeric, saffron and annatto (the colour obtained from the seeds of *Bixa orellana*), and the pinks and rose pinks from safflower, were undoubtedly also used quite early. Among the ancient civilisations, India was probably the most advanced in natural dyes.

The use of natural dyes for dyeing textile fibres is known to date back to 4000 BC. Among the various artefacts found in Tutankhamun's tomb (left to accompany King Tut on his journey to the afterlife) was a small dye box. The paint box was found to contain orpiment powders (a bright yellow mineral composed of arsenic trisulphide), red ochre (a pigment containing ferric oxide, typically clay, ranging from light yellow to brown or red) and malachite (a bright green mineral composed of copper hydroxyl carbonate). The Egyptians began to produce colour in earnest from about 4000 BC.

Examples of cotton fabrics have been found in India and Pakistan dating back to 3000 BC, but did not appear in Europe until the 4th century. Wool, a protein-based fibre and a common medieval fabric in both dyed and natural colours, dates back to 2000 BC. Silk, another protein-based fibre, was imported from China to Persia as early as 400-600 BC and became very popular in the Late Middle Ages. During this period, large silk production centres were established in France, Spain and Italy. Whether cotton, wool or silk, the colours used in clothing were proportional to the importance or wealth of the people. The rich wore bright colours, while the lower class wore white or shades of brown. The clothes of slaves were dyed grey, green and brown. Either way, dyed clothes were expensive all over the ancient world and were a matter of privilege.

Archaeologists studying the surviving coloured fabrics and important ancient manuscripts assume that there were three types of natural dyes: **plant dyes**, **mineral dyes** and **insect or animal dyes**. Mineral dyes are derived from minerals found on the earth's surface and in mines (haematite for red, limonite for yellow, lazurite (the old name; azure spar) for blue, etc.), animal dyes from animals such as insects and shellfish, and vegetable dyes from plants (saffron and safflower for yellow, madder for red, indigo for blue and bluish purple).

Natural dyestuffs have been used especially by the indigenous peoples of Greece, Rome, Egypt, South America, Africa, Asia and North America. As

civilisations grew and expanded during the Middle Ages and the Renaissance, trade routes opened up and the use of natural dyes expanded worldwide. Natural dyeing became a specialised craft and new guilds began to regulate and protect the trade during this period. In the late 15th century, the arrival of the New World in Europe led to the discovery of new sources of natural dyes and the widespread use of new natural dyes such as scale insect (cochineal), which was highly appreciated for its vivid colours. The use of natural dyes in that time was closely linked to the development of textile centres such as England and Flanders, Italy, where expert dyers and weavers produced excellent textiles using natural dyes. At this time, the textile trade and the use of natural dyes became an important part of the economy, and dyeing techniques and recipes became closely guarded secrets in guilds.

All dyes used in dyeing, which started 5000-6000 years ago and continued until 1856, were of animal, plant or mineral origin. Natural dyeing continued in this way until the accidental discovery of the first synthetic dyestuff by the English chemist William Henry Perkin in 1856. Perkin accidentally discovered a purple dye while trying to obtain quinine and called it mauve. Mauveine, created by chemical synthesis, led to the emergence of the synthetic dye industry. Mauve, a vibrant and stable dye, was produced commercially and quickly gained popularity so that it could be used in clothing, textiles and other materials. Indeed, by the late 1880s, every country store in North America was selling synthetic dyes for household use instead of natural dyes. This invention paved the way for the development of many other synthetic dyes, leading to the growth of the modern chemical and textile industries. With the industrial revolution in the 18th and 19th centuries and the discovery of synthetic dyestuffs in the early 20th century, the use of dye plants and other dyestuffs decreased and synthetic dyestuffs were adopted as the main source of commercial dyes due to their advantages such as being cheaper and less burdensome to obtain, allowing the production of a wide range of colours and revolutionised the textile industry. During this period, natural dyeing declined dramatically in certain regions of the world and was reduced to small-scale production, local crafts and traditional cultural practices. With the revival of the craft in the 1920s, traditional methods of natural dyeing were reintroduced by spinners, weavers and knitters, mostly without technical training in dye chemistry, for use in their finished products. The scarcity of dye base chemicals and exotic (imported) dyestuffs in the eighteenth and early nineteenth centuries severely limited craft dyeing in many places, especially for wool. In fact, the majority of dyeing manuals of the late 18th and early 19th centuries contained more recipes for cotton than for wool or silk, preventing the application of

improved methods for dyeing wool and silk and resulting in the application of simple and traditional dyeing methods.

In the 1980s, the importance of natural dyeing came to the agenda again with the determination that many of the synthetic dyestuffs are toxic, carcinogenic and cause environmental pollution. Today, the textile industry continues to use synthetic dyes for large production processes that pollute the environment. Synthetic dyes, which are mostly derived from petrochemicals, have negative environmental impacts such as pollution of water and soil. According to a report by the University of Cambridge, the dyeing process for a T-shirt uses an average of 16 to 20 litres of dyed water, with 80% of the dye remaining in the fabric and the remaining 20% of the dyed water being discarded. In Europe alone, 200,000 tonnes of dyed water is released into nature annually. In recent years, the synthetic dye industry has tried to improve health, safety and environmental conditions to reduce the negative impact, but some companies continue to produce carcinogenic dyes that harm not only their employees but also their customers who use their products (Baker-Brown 2017; Rana et al., 2015).

Natural dyestuffs, which are obtained from renewable resources and biodegradable, do not cause environmental pollution, are non-toxic and non-carcinogenic, and have natural antibacterial and antimicrobial properties. Natural dyes often produce subtle and variable colours that are appreciated for their natural beauty and individuality. The revival of natural dyes is also supported by advances in research and technology. Artisans, researchers and scientists are continually developing new recipes and methods in natural dyeing processes, investigating and experimenting with natural dye sources, dye extraction methods and dyeing techniques. This expands the colour palette of natural dyes day by day. In this context, consumers are increasingly demanding products made with natural dyes as a way to support environmentally friendly practices and reconnect with traditional and cultural heritage. For this reason, many artisans, designers and even brands are creating a market for sustainable and authentic products by adding natural dyes to their products.

Natural dyes are also frequently used in clothing, cosmetics industry (Henna - *Lawsonia inermis*, Catechu - *Senegalia catechu*), pharmaceutical industry (Saffron - *Crocus sativus*, Rhubarb - *Rheum rhabarbarum*) and food industry (Annatto - *Bixa orellana*, Turmeric - *Curcuma longa* and Cochineal - *Dactylopius coccus*) (Shahid and Mohammad, 2013; Melo, 2009). People's eco-protection, eco-safety and health concerns have increased the awareness of environmentally benign and non-toxic sustainability in bio-based colourants

(Shahid et al. 2012; Sarkar, 2004; Khan et al. 2005; Yusuf et al. 2016a; Yusuf et al. 2016b).

However, ecological concerns over the use of most synthetic dyes in the last few decades have motivated R&D experts all over the world to explore new eco-friendly substitutes and various aspects of bio-colourant applications to minimise their negative environmental impact. Therefore, both qualitative and quantitative research investigations are being conducted all over the world on colourants derived from cleaner biological sources with minimal ecological negative impact (Kulkarni et al. 2011; Luan et al. 2013; Swamy et al. 2013; Wang et al. 2016; Yusuf et al. 2012). Consequently, eco-friendly, non-toxic, naturally occurring bio-colourants are re-emerging as the next alternative through green chemistry approaches with wide applicability to textile colouring and other biomedical aspects (Shahid-ul-Islam and Shahid, 2013).

Turkey, which has a very rich geography in terms of natural vegetation and living biodiversity, is in an advantageous position compared to many other countries in the world in terms of the richness of dye plants and other living species used in the production of natural dyestuffs (Aydın, 2005, 2006; Aydın 2011a,b; Aydın & Karaca, 2018,2019; Aydın & Şen, 2020; Dinç, et al., 2015; Lillig & Aydın, 2006; Şekeroğlu & Aydın, 2002; Yaşar et al., 2003). The number of dye plants used all over the world is estimated to be between 300 and 400. In our country, it is known that the number of natural dye plant species is around 250. Due to this plant diversity, natural dye plants used in Anatolia are frequently used in many types of weaving such as carpets, rugs and bags (Aydın & Genç, 2021; Genç, 2020; Genç & Öztürk, 2023; Polat et al., 2023; Yıldırım & Genç, 2022; 2023). In addition, three species of six dye insects in the world are distributed in Turkey. Some of the shellfish species used for dyeing are found in the Aegean Sea, Black Sea, Marmara Sea and Mediterranean Sea. For this reason, both the Selçuk and Ottoman Empires used these rich dye plants in almost every field of textiles (Genç, 2014; 2017). For this reason, the Turks had an important place in the world in the field of dyeing and became famous as 'Turkish Red'. Today, these artefacts are exhibited both in our country and in the world's leading museums.

### **Scale Insects Used As Natural Red Dyes; Their Biology and Life Cycles**

Insects play an important role in the production of natural dyes and pigments of cultural, economic and ecological importance. These dyes are often preferred because they are safe compared to synthetic alternatives. Insect-derived dyes are biodegradable and often have a lower environmental impact compared to

synthetic dyes, which can be toxic and polluting. This makes them a more sustainable option for dyeing.

Certain insects produce pigments that can be extracted and used as dyes. In fact, different colours can be obtained from many insect species and these colours are used in many sectors such as textile, cosmetics and food. However, the insect species mostly used to obtain the red colour have a special importance in natural dyeing. Because the red colour has significant importance in dyeing for many reasons such as cultural significance, historical value, visibility and impact, symbolism in various contexts, variety of shades. Overall, the red colour's aesthetic appeal, and historical context contribute to its importance in dyeing.

Well known examples are the reds based on the ‘**laccaic acids/lac-dye**’ from *Kerria lacca* (Lac insects) and *Kerria chinensis*, ‘**kermesic acid/kermes**’ *Kermes vermilio* (Kermes), and ‘**carminic acid/ cochineals**’ from *Dactylopius coccus*, *D. opuntiae* (Cochineal), *Porphyrophora polonica*, *Porphyrophora hamelii*. The female lac insects, Indian lac *K. lacca* and *K. chinensis*, secrete a red resin, stick-lac, from which is obtained both the lac dye and the shellac resin while in both cochineal (*D. coccus*) and kermes (*K. vermilio*) the red dye is obtained from the female of adult and its eggs. Red or scarlet dyes are derived from the species of *Porphyrophora* spp. (e.g. Polish cochineal *P. polonica* and Armenian cochineal *P. hamelii*). Although species of *Porphyrophora* also contain carminic acid, *D. coccus* and *D. opuntiae* which has been cultivated has a much higher content (15–20%) of the dye, compared with only 0.8% and 0.6% for the Armenian and Polish species, respectively (Genç & Aydın, 2024). The number of natural dye insect species used to obtain red colour was known to be six, but this number has risen to seven species with the newly discovered *D. opuntia* (Genç & Aydın, 2024).

These insects have been historically important in dye production, contributing to various cultural and textile applications. Natural dyes are also valued for their color fastness and sustainability compared to synthetic alternatives.

### **Lac-dye (laccaic acids) - *Kerria lacca* and *Kerria chinensis***

*Kerria lacca* is a species native to Asia. It contains laccaic acid A, laccaic acid B, laccaic acid C, laccaic acid D, laccaic acid E dyes.

These species, including *Kerria lacca*, and *K. chinensis* belong to the family Kerridae (Hemiptera), which comprises about 9 genera and 90 species, and secrete lac resin on trees, i.e. shellac, a natural polymer containing pigment and wax. Shellac is a physiologically harmless and biodegradable resin secreted by

the female lac insect. Shellac, a resin purified from the secretion of lac insects, is an important component in various inks, paints, sealants and varnishes (Gandini, 2011; Debeaufort et al., 1998). Lac resin, a natural polymer, is the only commercial resin of animal origin. Shellac is one of the natural materials widely used as a coating, polishing and film-forming agent in the food, pharmaceutical and cosmetic industries. It is also widely used in chemical, electronic, military, food and other industries worldwide (Ben-Dov and Lit, 1998; Chen et al., 2011; Chamberlin, 1923; Varshney, 1976; Chen, 2005; Kondo and Gullan, 2007; Ben-Dov et al., 2006).

Lac insects have a piercing-sucking mouthparts and feed on plant sap. Lac beetles are known to feed on more than 400 hosts, but are most commonly found on a variety of tropical plants such as *Schleichera oleosa* (Sapindaceae) and *Cajanus cajan* (Fabaceae), and most commonly on *Butea monoperma* (Fabaceae). A female can lay between 600 and 1000 eggs. Hatching nymphs attach themselves to the branches of host plants to feed on phloem tissue. They cover the holes in the branches they pierce with wax secretions. The wingless females, which, like other cochineals, fix themselves, can range in various shades of red.

Water-soluble lac dye was used in India for human decoration and also for dyeing wool and silk, while in China it was used to dye leather. Lac dye is similar in colour to other cochineals such as *Dactylopius coccus* and *D. opuntiae* (Hemiptera: Dactylopiidae). The bright red dye (erythrolactin) of the lac bug is used to dye silk and wool. The dye can be treated with various mordants to produce a range of colours from purple to red to brown. Lac dye remained a valuable product until the development of synthetic dyes in the late 19th century.

The dried and grinded insect is used to obtain red and purple colours by mordant dyeing.

### **Kermes (kermesic acid)' *Kermes vermilio***

*Kermes vermilio* is distributed in the Mediterranean HotSpot. It contains kermesic acid, and flavo-kermesic acid.

Like all species of the Kermesidae (Hemiptera) family, *K. vermilio* lives on evergreen oak trees. Adult females are about 5-7 mm long and dark red to brown in colour with a thin white coating of wax powder.

As with all scale insects, the winged male mates with the wingless female, who has over 1000 eggs in her body. The hatching nymphs wait under the dead mother's shell for a certain amount of time before moving on to the young

shoots. They then go to the young shoots and fix themselves there, secreting secretions as they feed.

As with all scale insects, the female of *K. vermilio* is used as a natural dye. A person can only collect 1 kg of insects per day. However, the collected insects lose 2/3 of their weight when dried. Kermesic acid from *K. vermilio* was used as dye and lacquer pigment in ancient Armenia, Egypt, Greece, and the Middle East.

The dried and powdered insect is dyed with the mordant dyeing method and red and its shades are obtained.

**Cochineals (carminic acid) - *Dactylopius coccus*, *D. opuntiae*, *Porphyrophora polonica*, *P. hamelii***

*Dactylopius coccus* (Hemiptera: Dactylopiidae), is distributed in tropical and subtropical regions from North America to South America (Mexico and the southwestern United States). It contains carminic acid, kermesic acid, flavo-kermesic acid, x1, x2, x3 dyes.

*D. opuntiae* is distributed rapidly in Mediterranean basin. Adult females of *D. opuntiae* produce carminic acid, which occurs naturally in the body.

*D. coccus*, and *D. opuntiae* also known as cochineal, is a scale insect of the family Dactylopiidae, order Hemiptera, from which the natural dye carmine is extracted. These mentioned species are the pest of the cactus genus *Opuntia*. This insect feeds on the sap of the cactus. Cochineal insects are soft-bodied, flat, oval insects. Females are wingless and approximately 5 mm long, cluster on cacti (Martinez and Guzmán, 1998; Miller, 2022). Their mouthparts are piercing-sucking and they feed on the sap of the cactus. The female remains fixed and immobile on the cactus. After mating, the fertilised female increases in body size and produces nymphs (Eisner, 2003). Hatching nymphs (1st nymphal stage) secrete a cottony wax (*D. coccus*) and powder wax (*D. coccus*) white substance on their bodies to protect them from dehydration and excessive sunlight (Genç & Aydın, 2024). This substance makes the insect appear white. However, the bodies of adults and nymphs produce a red pigment that makes the insect appear dark purple on the inside. Young nymphs can be transported by the wind from one host to another using their long wax filaments, find a suitable site and attach themselves (Olson, 2005). The fewer male nymphs continue to feed on the cactus until they reach sexual maturity. After prepupa and pupa period, they then spread their wings and enter the adult stage. However, they do not feed during this period and live only long enough to fertilise the eggs. Therefore, male winged individuals are not often seen

(Armstrong, 2022; Nobel, 2002). The carminic acid and hence its chemical structure is a predator deterrent, which is present in high concentration in the scale insect and prevents predation by other insects.

Carmine dye was used to colour fabrics in America and became an important export during the colonial period in the 16th century. After the invention of synthetic pigments and dyes in the late 19th century, the use of natural dyes gradually declined. Fears about the safety of artificial food additives revived the popularity of red dyes, and because of increased demand made scale insect farming profitable again. Today, however, they are still mainly used as colourants in cosmetics, beverages and histology.

The dried and powdered insect is dyed with the mordant dyeing method and red and its shades are obtained.

It is known that the Polish Kermes, *Porphyrophora polonica* (Hemiptera: Margarodidae), is distributed in France, Germany, Hungary, Czechoslovakia, Poland, Switzerland, Ukraine, Belarus, Russian Federation, Kazakhstan, Kyrgyzstan and Mongolia. It contains carminic acid and kermesic acid. The nymphs of *P. polonica* are a species of scale insects that live on the roots of various plants, especially the perennial *Scleranthus annuus* L. (Caryophyllaceae) (Turkish: Kınavel), which grows in sandy soils of Central Europe and other parts of Eurasia.

The adult female Polish Kermes, which prefers semi-desert ecosystems and plants growing in sandy soils, is about 4.5 mm long and dark reddish brown. Male individuals are 2-3 mm long and have a long, thin, cylindrical shape. In mid-July, the female Polish Kermes lays a white waxy egg case on the ground. The egg case contains about 600-700 eggs. The larvae hatch in late August or early September. However, they do not leave the site and spend the winter in the egg case as 1st instar larvae. The larvae emerge from the soil in late March or early April, and feed briefly on the lower leaves of the host plant before returning underground to feed on the roots of the plant. Nymphs shed their exoskeleton, including legs and antennae, and molt. They become 2nd instar nymphs and form outer protective sheaths (i.e. cysts) on the roots of the host plant. These small, clustered, dark red or purple bubbles take on the appearance of cysts and are the natural source of the dye. Females are 3-4 millimetres in diameter. Males are half the size of females and fewer in number.

Polish Kermes was widely used throughout Europe during the Middle Ages and the Renaissance. In the 15th and 16th centuries, Poland exported Polish Kermes to countries such as southern Germany and northern Italy, France, England, the Ottoman Empire and Armenia. However, the introduction of the

cheaper *D. coccus* from Mexico led to a sudden decline in the trade in Polish Kermes, and by the 1540s the quantities of red dye exported from Poland had fallen sharply. Until then, Polish Kermes had continued to be used locally by peasants who had collected it, mostly as an export product. In the late 18th century, large markets in Russia and Central Asia revived the trade in Polish Kermes. In the 19th century the market expanded and spread to Central Asia. However, *P. polonica* lost its economic importance after the development of synthetic dyes in the late 19th and early 20th centuries.

The dried and powdered insect is dyed by the mordant dyeing method.

*Porphyrophora hamelii* is a species of scale insect found in the Ararat Plain (Igdır Plain), the Aras River Valley and the Armenian Highlands. *P. hamelii* contains carminic acid, kermesic acid, flavo-kermesic acid, x1, x2, x3. *P. hamelii*, also known as Mount Ararat Kermes, feeds mainly on the roots of *Phragmites australis* and *Aeluropus litoralis*. For this reason, they come to the surface almost exclusively for mating (Khachatryan, 2014; Vahedi and Hodgson, 2007; Foldi, 2005).

In spring, newly emerged nymphs (pre-adult stages) burrow into the soil until they reach the roots of plants such as *P. australis* and/or *A. litoralis*. Pre-adult stages feed on the roots of these plants throughout the spring and summer. While feeding, the nymphs become covered with a protective layer of pearly cysts. Adult females emerge from the soil to mate between 5 and 10 a.m. from mid-september to mid-october (Cardon, 2007; Anonymous, 2014).

Red and mottled shades are obtained by mordant dyeing with dried and powdered kermes from Mount Ararat. The misconception is that living insects are used in insect dyeing process instead of dead ones.

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## Chapter 7

### Manipulation Of Growth And Abiotic Stress Resistance Via Salicylic Acid (SA) in Plants

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## 1. INTRODUCTION

The most significant issue in the twenty-first century is climate change, which affects the poorest and most vulnerable people disproportionately and calls for new levels of international cooperation against drought. So that, significant damaging environmental factor that affects crop yields worldwide is drought stress (Khalvandi et al., 2021). Plants engage a variety of intricate defense systems in response to drought stress, including the antioxidant defense system, some phytohormones like SA (Nazar et al., 2015). SA is clearly a stress-signal molecule that, in response to environmental stressors, increases the expression of biosynthetic enzymes and proteins in plants (Nazar et al., 2015; Wang et al., 2019; Khalvandi et al., 2021) and activates the expression of abiotic stress-responsive genes (Li et al., 2013). Numerous physiological processes in plants, including stomatal closure, photosynthesis and respiration, nutrient uptake, nitrogen metabolism, and growth and development as seed germination, vegetative root and shoot growth, flowering, yield, and senescence, plant defense system, plant-water relations under stress, and antioxidant defense system, which together protects plants from abiotic stressors have all been demonstrated to be impacted by exogenously supplied SA (Khan et al., 2010, 2012a,b,c, 2013b, 2014; Nazar et al., 2011; Miura and Tada, 2014, Khan et al., 2015, Wollenweber, 2022). SA has been shown to improve plant tolerance to major abiotic stresses such as metal (Zhang et al., 2015), salinity (Khan et al., 2014; Nazar et al., 2015), osmotic (Alavi et al., 2014), drought (Fayez and Bazid, 2014), and heat stress (Khan et al., 2013b), in addition to its role in the induction of defense-related genes and stress resistance in biotically stressed plants (Kumar, 2014). Because these processes are frequently initiated by low concentrations of SA and blocked by large amounts, SA controls them in a concentration-dependent manner (Wollenweber, 2022).

When plants are under abiotic stress, exogenous application of SA can effectively regulate their growth responses and production while also protecting them from biotic stress. It is widely known that endogenous SA levels in plants are positively correlated with defense mechanisms against hemibiotrophic and biotrophic diseases (Glazebrook, 2005). The amount of SA varies between species, plants, and organs. It also relies on the environment, the stage at which the plant is developing, and other factors (Wollenweber, 2022). As a result, it is impossible to determine the ideal concentration for applying SA exogenously to a particular crop. The examined literature has employed the subsequent concentrations for some plants are listed below (Wollenweber, 2022):

**Table 1.** Suitable SA application range concentrations for some plants (Koo et. al., 2020; Wollenweber, 2022).

<b>Plant</b>	<b>SA conc.</b>
Canola	10-50 $\mu$ M
Paddy rice	60-100 $\mu$ M
Barley	0.1-1 mM
Wheat	0.5-1 mM
Pea plants	0.5-1 mM
Corn	0.5-1 mM
Soybean	0.5-1 mM
Green beans	1-2 mM
Tomatoes	1-1.5 mM
Pepper	0.5 mM
Ornage	0.25 mM
Rubber tree	5 mM
Pumpkin	0.1 mM
Tea flower	1 mM

Furthermore, the application of exogenous SA causes several plant species to develop both systemic and local acquired resistance to diverse forms of stress. In this context, the importance of local studies under various environmental conditions are increasing.

## **2. THE CHEMICAL STRUCTURE AND FUNCTION OF SALICYLIC ACID**

SA is a chemical molecule that consists of a benzene ring with a hydroxyl group (-OH) and a carboxyl group attached. Chemically classified as a phenolic acid, this compound plays a crucial role in regulating defense responses in plants. Salicylic acid aids plants in combating diseases, responding to environmental stresses, and regulating normal developmental processes. These effects are attributed to SA's biological activity and its influence on plant defense mechanisms.

## **3. SALICYLIC ACID'S INFLUENCE ON PLANT GROWTH AND DEVELOPMENT**

Since plants are confined organisms, plant hormones, also known as phytohormones, play a major role in controlling their growth and ability to respond to external factors. Phytohormones control many physiological systems at very low concentrations by acting as endogenous signals in time (Koo et. al., 2020). Senescence, blooming and seed germination are crucial stages in the growth and development of crops. It has been demonstrated that these

mechanisms are significantly impacted by the synthesis and metabolism of SA (Wollenweber, 2022; Bagautdinova et al., 2022; Liu et al., 2022). In most cases, the amount of SA used, the plant's genotype, and the experimental environment determine how much of an impact SA has on plants growth.

Exogenous management of SA has been shown to be beneficial for plant growth, whether it is done by spraying plants with SA solution, adding SA to a hydroponic solution, or seed priming (soaking seeds before sowing) (Hayat et al. 2010; Wollenweber, 2022). Usually, large concentrations of SA negatively limit the growth and development of plants (depending on the species of plant, but >1 mM SA is considered a high concentration) (Koo et. al., 2020). However, using SA at the right quantities has positive benefits. Based on the specific environmental conditions, SA was found to be significantly enhance the growth in various plant species under both typical and abiotic stress conditions (Gunes et al., 2007; Gutiérrez-Coronado et al., 1998; Kováčik et al., 2009; Manzoor et al., 2015; Sakhabutdinova et al., 2003; Yildirim et al., 2008, 2015).

#### **4. THE FUNCTION OF SALICYLIC ACID IN PLANT PHYSIOLOGY**

Salicylic acid regulates plant defensive responses, including systemic acquired resistance (SAR). SAR not only halts infection at the location of pathogen assault but also activates defense-related genes throughout the plant, enabling it to develop stronger resistance against future pathogens. This process is triggered through a signaling pathway in which salicylic acid is actively involved.

Salicylic acid is a vital component in the fight against plant diseases caused by fungi, bacteria, and viruses. Its application enhances plants' resistance to pathogens, effectively countering soil-borne fungal diseases like *Fusarium*, *Phytophthora*, and *Pythium*, as well as certain bacterial infections. Furthermore, it strengthens plant defenses against viral infections. Salicylic acid's ability to boost these defense mechanisms paves the way for the adoption of organic and biological methods in plant disease management.

Salicylic acid also functions as a hormone that enhances plant resilience to environmental stresses such as temperature fluctuations, drought, salinity, and even heavy metal contamination. It helps plants reduce water loss, regulate stomatal openings (tiny pores involved in gas exchange), and optimize photosynthesis. For instance, under drought conditions, salicylic acid aids plants in minimizing water loss, thereby increasing their tolerance to water stress.

Salicylic acid acts as a growth regulator in plants, influencing various developmental processes. It promotes cell division and accelerates growth, while also enhancing root development, enabling plants to utilize nutrients more efficiently. Positive effects of salicylic acid are observed in growth stages such as seed germination, root development, and leaf expansion, demonstrating its crucial role in supporting healthy plant growth.

## **5. THE USES OF SALICYLIC ACID IN AGRICULTURE**

Salicylic acid enhances plants' defense mechanisms against pathogens, making them more resistant to diseases. It has been shown to be effective, especially in preventing fungal, bacterial, and viral infections. This property offers the potential to reduce the reliance on chemical pesticides. Moreover, it can be utilized as a natural treatment method preferred in organic farming.

Environmental stresses can negatively impact plant growth and productivity. Salicylic acid increases plants' resistance to stress factors such as heat, drought, and salinity. This compound helps plants minimize water loss, regulate stomatal openings, and optimize their metabolic activities.

Salicylic acid also influences plant growth. It can promote cell division and elongation in plants, enhancing root development. This effect is particularly beneficial in areas with poor soil structure or limited nutrients, enabling plants to grow more efficiently.

In fruit trees and vegetables, salicylic acid applications can improve productivity by regulating stress responses in plants, it contributes to the development of healthier fruits and vegetables. Additionally, salicylic acid can accelerate fruit ripening and enhance post-harvest quality.

The effects of salicylic acid on soil microorganisms have also been studied. It can stimulate the activity of certain beneficial microorganisms, which improves soil health. Maintaining a balanced microbial environment in the soil is essential for healthy plant growth, and salicylic acid supports this balance effectively.

Soil health is a critical factor that directly influences plant development. Salicylic acid can enhance the activity of certain soil microorganisms. These microorganisms contribute to soil fertility by promoting processes such as nitrogen fixation and the decomposition of organic matter. Additionally, salicylic acid can help suppress harmful pathogens in the soil, further supporting a healthy environment for plant growth.

## **6. METHODS OF USING SALICYLIC ACID**

### *Foliar Application*

Foliar application is a method that allows salicylic acid to be rapidly absorbed by plants. Leaves quickly take up the compound and distribute it throughout the plant. This method is particularly preferred during the early stages of disease outbreaks to activate a swift defense mechanism.

### *Soil Application*

Salicylic acid can also be administered to plants through the soil. Direct uptake by the roots supports root development and enhances overall plant health. This approach is especially beneficial in areas where soil nutrient levels are low.

### *Application via Irrigation Systems*

Using irrigation systems to apply salicylic acid is an efficient method for covering large areas. Through drip irrigation or other watering systems, plants can be regularly supplied with salicylic acid, ensuring consistent benefits.

### *Salicylic Acid: A Key Component in Agriculture*

Salicylic acid is a crucial compound in agriculture due to its ability to activate plants' natural defense mechanisms, enhance resistance to environmental stresses, and support productivity. Applying this compound at the correct dosage and timing can significantly improve plants' resilience against diseases. Additionally, its benefits, such as increasing tolerance to environmental stresses, promoting soil health, and improving productivity, make salicylic acid a promising tool for future agricultural practices.

Various studies on the use of salicylic acid in agriculture have demonstrated its positive effects on plant health and productivity. Below are examples of key studies highlighting how salicylic acid is utilized in agriculture and the results achieved.

## **7. STUDIES ON THE AGRICULTURAL APPLICATIONS OF SALICYLIC ACID**

Abiotic stressors pose a serious threat to the agricultural system and crop productivity. Whether they occur alone or in combination, these stresses have a variety of negative impacts in plants that disrupt physiological, molecular, and biochemical functions. In the end, this may lead to significant declines in the growth, development, and productivity of plants. In this context, the evaluation of comprehensive studies conducted on the subject is summarized and presented below.

Exogenous SA treatment has been shown to increase tomato plants' resistance to TYLCV in two different tomato cultivars: the resistant "Zhefen-702" and the

susceptible "Jinpeng-1," according to Li et al. (2019). The effects of SA were also assessed on the expression patterns of genes associated with stress, the accumulation of ascorbic acid (AsA), the activity of important enzymes involved in the scavenging of reactive oxygen species (ROS), and the expression of biosynthetic genes. The results showed that SA can successfully control the buildup of AsA, especially in the "Jinpeng-1" cultivar. The expression patterns of most of the genes involved in AsA production were negatively correlated with AsA accumulation in both the resistant and susceptible tomato varieties. With the exception of 14 days (APX in "Jinpeng-1" was also at 4 days) following TYLCV infection (dpi), the activity of ascorbate peroxidase (APX) and peroxidase (POD) in plants treated with SA and infected with TYLCV rose over the trial. At the same time, the "Zhefen-702" plants treated with SA + TYLCV had higher SOD activity, while the "Jinpeng-1" plants had lower SOD activity. The viral burden in the SA + TYLCV counseling varied from 2 to 10 dpi, and SA can significantly increase the expression of genes that scavenge reactive oxygen species.

In the National Research Center's greenhouse, cucumber (*Cucumis sativus* L.) plants were cultivated in pots to ascertain the impact of low temperatures on the growth and development of the plants, with the objective of achieving an early output in April. The study was conducted in Dokki, Giza, Egypt (Orabi et al., 2010). The effects of low temperatures on plants were reduced by the application of paclobutrazol (25 and 50 mg/l) and salicylic acid (2 and 4 mM). The application of SA at a concentration of 4 mM, followed by 2 mM, and paclobutrazol at a concentration of 25 mg/l, to plants grown in low temperatures and subjected to foliarly treatment, resulted in the improvement of protective parameters, including antioxidant enzyme activity and carotenoid content. This was observed to mitigate the negative effects of low temperature stress, as evidenced by the results obtained. The greatest increases in GR and APX activities, survival percentage, and carotenoid content were observed in plants treated with SA at a concentration of 4 mM.

Yılmaz and Kızılgöçü (2022) carried out the investigation to ascertain the effects of varying salicylic acid dosages on drought stress during germination and early development stages in two bread wheat genotypes (DZ17-1 and Empire Plus). The bread wheat genotypes' seeds were subjected to 0, 0.5, and 1 mM SA during the investigation. Five distinct drought stress conditions were used to germinate the seeds. The study's findings indicate that all of the traits looked at suffered as drought stress increased. Only the 0.5 mM SA treatment had a beneficial impact on germination power and rate during drought.

In a study by Nasırcılar et al. (2020), radish types in PEG6000-containing environments were treated with several dosages of salicylic acid (0.25, 0.50, 0.75, and 1.00 mM) to boost drought resistance. Depending on the variety, different salicylic acid dosages had varying impacts on radish seed germination and vegetative development parameters during drought; a stimulating effect on germination was found. The SA levels given to the large red radish variety were found to have an inhibiting effect, whereas the treatments typically had a beneficial effect on plant development.

Salicylic acid (SA) has the potential to improve a plant's resistance to drought stress and has an impact on a variety of physiological and biochemical functions. So that Khalvandi et al., 2021 conduct the study concentrated on how the administration of SA under drought stress circumstances affected the proteins, photosynthesis, and antioxidant system of Sardari wheat ecotypes. Treatments included salicylic acid at 0.5 mM (controls were left untreated), drought stress (30% of the field capacity), and Sardari wheat ecotypes (Baharband, Kalati, Fetrezamin, Gavdareh, Telvar, and Tazehabad). The findings demonstrated that all six ecotypes' lipid peroxidation and membrane electrolyte leakage were clearly elevated in drought-stressed environments. Conversely, drought stress reduced the amount of chlorophyll in the leaves, as well as the rate of photosynthetic activity, stomatal conductance, carboxylation efficiency, and transpiration rate. SDS-PAGE results showed that when plants were subjected to drought stress, the abundance of some protein spots decreased while the number of other protein spots increased. Baharband and Telvar showed maximum protein band counts, photosynthetic efficiency, cell membrane stability, and antioxidant enzymatic activity under stress, while Fetrezamin showed the lowest by boosting the activity of antioxidant enzymes, maintaining membrane permeability, inducing stress proteins, and improving photosynthetic performance, salicylic acid treatments successfully reduced the detrimental effects of drought stress on Sardari ecotypes.

It is unknown how SA, a major phenolic linked to stress response, affects rice's reaction to concurrent salt and drought stress. One important element limiting agricultural productivity has been salt and drought stress (Shan et al., 2024). By identifying physiological and biochemical markers as well as the expression of genes linked to drought and salt tolerance, Shan et al. (2024) investigated the impacts and mechanisms of exogenous SA-triggered rice adaptation to dual drought and salt stress. The results showed that giving SA to rice seedlings under drought and salt stress could significantly increase their antioxidant enzyme activities, reducing MDA and H<sub>2</sub>O<sub>2</sub> levels and maintaining rice seedling growth. Additionally, the expression of genes involved in the response to abiotic stress,

such as OsDREB2A, OsSAPK8, OsSAPK10, and OsMYB2, was up-regulated in salt and drought conditions. The use of SA may also enhance the expression of genes like OsDREB2A and OsSAPK8, suggesting that SA may control the activity of antioxidant enzymes by promoting the expression of genes linked to drought and salt tolerance and enhancing rice's resilience to these conditions.

One important tactic in crop water management is agricultural water rationalization, which is demonstrated by irrigating plants less than they need. However, it is true that reduced water supplies result in lower crop output. Thus, the goal of the current study was to use salicylic acid (SA) to reduce sunflower yield losses related to deficit irrigation (El-Bially et al., 2022). In the El Nubaria region of the El Behaira Governorate, Egypt, El-Bially et al. (2022) investigated the effects of three levels of SA (0.0, 0.5, and 1mM, abbreviated as SA0.0, SA0.5, and SA1.0, respectively) and three irrigation regimes (100, 85, and 70% of crop evapotranspiration, designated WR100%, WR85%, and WR70%, respectively) on the performance of sunflower plants over two seasons in 2019 and 2020. According to the findings, the sunflower plants treated with WR100%×SA1.0 had the highest levels of carotenoids and total chlorophyll and the lowest proline concentration. Compared to the WR70%×SA0.0 treatment, the WR100%×SA1.0 treatment yielded 109.7% more seeds in the first season and 125.9% more in the second. SA0.5 and SA1.0 decreased the losses in seed production from 21.0% to 15.8 and 14.4% and 46.2% to 40.8 and 40.1% under WR85% and WR70%, respectively, compared to the farmer standard practice. WR100%×SA1.0 for the iodine value and WR100%×SA1.0 and WR100%×SA0.5 for the seed oil percentage were the highest values. The WR100%×SA1.0 and WR100%×SA0.5 combinations were the most effective at boosting water use efficiency.

During drought, the net photosynthetic rate, tobacco output, and tobacco quality all decline. Salicylic acid (SA) is involved in the regulation of many physiological processes and has been used extensively to improve plant drought tolerance (Feng et al., 2023). In the hydroponic experiment, the fluecured tobacco variety K326 was utilized. SA solutions (0.3 mM) were sprayed on the leaves every day for three days. 15% PEG-6000 was then used by Feng et al. (2023) to simulate mild drought. The results showed that drought stress reduced photosynthetic rate, stomatal conductance, and transpiration rate while increasing the activities of superoxide dismutase, peroxidase, catalase, malondialdehyde, proline, and protein content. Compared to PEG drought stress, SA enhanced the photosynthetic rate, stomatal conductance, and transpiration rate by 45.74%, 26.82%, and 52.22%, respectively. Additionally, SA spraying increased tobacco's drought resilience by changing the expression levels of genes related to carbon

metabolism, photosynthetic-antenna proteins, and photosynthesis. Therefore, by increasing the antioxidant system and decreasing the effect of drought on photosynthesis, SA spraying increased tobacco's resistance to drought.

Due to its low drought tolerance, cocoa (*Theobroma cacao* L.) is not able to withstand extended dry spells (Matias et al., 2024). Matias et al. (2024) assessed the physiological and morpho-anatomical reactions of two clonal cacao seedlings, BR25 and K9, under drought stress. They also investigated the possible mitigating effects of salicylic acid (SA) at 0.5 and 1.0 mM on the early vegetative stage. Two weeks prior to the start of drought conditions, 0.5 and 1.0 mM SA were applied topically to eleven-month-old BR25 and K9 cacao. The control group consisted of seedlings that were well-watered and drought-tolerant without any prior treatments. After 20 days of water deprivation, rewatering began three months later. The findings showed that both clonal varieties showed enhanced ascorbate peroxidase (APX) and catalase activity and a significant decrease in relative water content (RWC) following 20 days of dryness. After three months of rewatering, changes in stem diameter were noted. After 20 days of dryness, K9 showed a discernible decrease in stomatal aperture and root and shoot dry weight, whereas BR25 showed a decline in total chlorophyll content as well as chlorophylls a and b. Additionally, the study's findings indicate that by enhancing RWC, stomatal length, and width, 0.5-mM SA may be able to lessen the severe effects of drought in BR25 and K9 cacao. However, by increasing the seedlings' root dry weight and APX activity, 1.0-mM SA may be able to mitigate the impacts of the earlier drought.

Exogenous use of elicitors, like salicylic acid, is required because plant endogenous systems are not always adequate to reduce drought stress (Melo et al., 2024). This study evaluated the mitigating effect of salicylic acid (SA) on cowpea genotypes under drought conditions and was conducted by Soares de Melo et al. in 2024. Two genotypes of cowpeas and six treatments of salicylic acid and drought stress were used in the experiment. Drought stress lowers both cowpea genotypes' output by reducing their leaf area, stomatal conductance, and photosynthesis. Regardless of the stress conditions, the BRS Paraguaçu genotype outcompetes the Pingo de Ouro-1-2 genotype in terms of growth and production. Cowpea leaves treated exogenously with 0.5 mM salicylic acid exhibit improved production of both genotypes.

The cowpea genotype is less susceptible to drought stress when 0.5 mM salicylic acid is applied, in contrast, the BRS Paraguaçu genotype is more drought-tolerant. Making the best use of the fewest resources possible is a crucial strategy for reducing plant drought. In order to maximize the amount of water

available to boost photosynthesis and plant growth under water-deficit stress, it is critical to raise water-use efficiency (WUE) (Iqbal et. al., 2022). Iqbal et al. (2022) investigated the photosynthetic nitrogen-use efficiency (PNUE) and WUE resource use efficiency indices in mustard (*Brassica juncea* L.) plants sprayed with 0 or 0.5 mM salicylic acid (SA) and grown in water-limited circumstances with low-N and sufficient-N. Through the enhancement of PNUE, SA application raised WUE, osmotic potential, water potential, and the absorption of soil N into photosynthetic activity. Additionally, under water-deficit stress, it enhanced conductance of stomata and intercellular CO<sub>2</sub> concentration, which maximized plant photosynthesis. This rise was larger when there was enough N present, and 0.5 mM SA significantly improved the redox ratio and N metabolism, which reduced oxidative stress. SA treatment decreased the synthesis of ethylene and abscisic acid (ABA) in plants treated with N. It may be deduced that SA improved N usage by raising NUE when it is available, raising GSH levels to preserve the redox balance, and blocking ABA-mediated stomatal closure to improve photosynthetic and resource use. Additionally, SA improved glucose consumption, preventing photosynthetic suppression caused by glucose under stress. Applying SA could therefore provide a possible management option for boosting photosynthetic NUE, WUE, and photosynthesis under drought.

One of the most appealing possibilities for a salinity moderator is salicylic acid (SA) (Youssef et. al., 2023). In order to evaluate the effects of foliar application of SA on growth, productivity, and certain physiological and biochemical parameters, French beans (*Phaseolus vulgaris* L.) were continually exposed to three NaCl levels in a hydroponic experiment (Youssef et al., 2023). Phenolic compound concentrations, total antioxidant capacity, proline and malondialdehyde contents, peroxidase activity, Na<sup>+</sup> and Cl<sup>-</sup> ions, and abscisic acid were all considerably higher than those of the non-salt-stressed controls. Additionally, the NaCl treatment dramatically reduced the yield of green pods as well as the vegetative development indices, membrane stability, relative water content, chlorophyll content, and growth boosters. The varied degrees to which foliar application of SA at 0.75 mM was able to alleviate the deleterious effects of NaCl stress allowed for the achievement of nearly 90% of the yield of control plants. In conclusion, our research demonstrated that foliar spraying SA reduced the adverse effects of NaCl stress in French beans by regulating specific physiological and biochemical processes. This could be the starting point for an effective and reasonably priced salt stress management strategy.

## **8. CONCLUSION AND FUTURE PERSPECTIVES**

The agricultural system is known to be seriously threatened by abiotic stress. Plants use a variety of physiological and molecular strategies to counteract the negative effects of abiotic stress. Research has indicated that SA is a robust and promising strategy for mitigating the negative effects of abiotic stress on plants.

Numerous techniques for applying SA have been demonstrated to strengthen different plant species' resistance to abiotic stressors, including seed priming (soaking seeds in SA before planting), adding SA to hydroponic solutions, and irrigating or spraying plants with SA solutions. The literature reviewed here supported the idea that the primary goal of research on SA in abiotically stressed plants is to reveal different physiological and biochemical mechanisms. Practically speaking, though, not all of these application techniques are suitable for field use, particularly for cereals that are cultivated on expansive plots of land and can be very costly. It is only appropriate to employ hydroponic cultures in greenhouses.

In this content, many studies underscore the versatile applications of salicylic acid in agriculture and its positive effects on plant health. Salicylic acid not only strengthens plant defense mechanisms but also enhances resistance to environmental stresses, regulates growth, and boosts productivity. These characteristics make it a valuable tool, particularly in sustainable and organic farming practices. More detailed information about SA-mediated defense networks and plant immunity, as well as more information about how SA interacts with other defense signaling pathways in abiotic stressed plants, can generally be found by combining genetics, molecular biology, biochemistry, genomics, bioinformatics techniques, and computational biology.

Future research could focus on developing strategies for more effective use of salicylic acid in disease control, stress management, and overall yield enhancement.

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## **Chapter 8**

### **Zebra Mussel Problem In Inland Waters**

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## INTRODUCTION

Zebra mussels appeared in Great Britain following the import of moist timber from the Baltic region in 1824 (Minchin et al., 2002). Timber was exported to many European ports and the Netherlands was soon invaded. Since then, zebra mussels have spread to Sweden, Finland, Switzerland, Northern Italy, France and Ireland (Yıldırım, 2023). The first records of the zebra mussel in our country, which is one of the invasive species that causes the greatest ecological and economic damage in inland waters in the world, date back to 1987. In 1936, it was recorded that *Dreissena* species were found in the fresh waters of Bursa and its surroundings. A series of studies have been carried out by the General Directorate of DSI since 2001 after the zebra mussel caused problems in the Atatürk Dam and Hydroelectric Power Plant since 1997 and in the Birecik Dam in 2000 (Aksu and Yıldız, 2017). Some of the creatures that cause problems are species in the Dreissenidae family and especially the zebra mussel (*Dreissena polymorpha* (Pallas, 1771)). The zebra mussel was described by Russian scientist and explorer Pyotr Simon Pallas from a species community in the Ural River and its tributaries in the Caspian Sea basin. It is understood that research on zebra mussels began in the last 150 years, when the mussel spread from its natural distribution areas to fresh waters in Europe, and studies intensified after it entered North America in 1988 (Sprung and Borcharding, 1991).

In the study conducted by DSI (2005) on zebra mussels, which cause major mechanical problems due to blocking transmission lines, especially in dams, the presence of zebra mussels in more than 1500 irrigation systems throughout our country was reported (Akkuş et al., 2019). In addition to pollution problems, zebra mussels also cause problems in aquatic ecosystems. The main reason for the problems is that the density and mass of mussels can reach very high levels (for example, densities of 30 000 - 100 000 pieces/m<sup>2</sup>; living mass is 10 times the mass of all invertebrate bottom creatures) (ZMIS, 2001). The economic losses caused by zebra mussels in North America are estimated to be approximately 5 billion dollars/year. For this reason, mussels continue to be one of the pests on which intensive research has been conducted (ZMIS, 2001; URL-1, 2024).

Zebra mussel studies in our country have developed with a focus on mechanical problems occurring in dams. In our country, there is almost no data on the effects of zebra mussels on other creatures living in aquatic ecosystems (Yıldırım et al., 1996). Some studies on zebra mussels in our country are summarized below. Altınayar et al. (2001) reported that zebra mussels cause

significant economic problems in HEPPs. It is stated that great damage has occurred in dams and hydroelectric power plants in the Euphrates Basin due to zebra mussels blocking transmission lines and excessive proliferation (Bobat et al., 2004). Disruptions occur in the operations of enterprises due to zebra mussels clinging to the mechanical parts of HEPPs and dams (Aksu et al., 2007). In Sapanca Lake, it constitutes the dominant food item of the rock fish in the lake, which is considered harmful (Gaygusuz et al., 2007). Zebra mussels cling to the carapace region of pincer crayfish living in Çıldır and Eğirdir Lakes; They reported that crayfish have negative effects on mating, avoiding predators, movement and nutrition (Berber et al., 2018). Rapidly multiplying zebra mussels in HEPPs and water transmission systems cause clogging of the systems (Aksu and Yıldız, 2017; Akkuş et al., 2019).

Since *D. polymorpha* has a strong oxidative defense and is long-lived, it is widely used to conduct many toxicological experiments in aquatic ecosystems (Serdar et al., 2021a; Serdar et al., 2021b; Serdar et al., 2023; Cıkcıoğlu Yildirim et al. al., 2024; Serdar et al., 2024).

## **BIOLOGICAL STRUCTURE OF ZEBRA MUSSEL**

The place of the zebra mussel in the classification is given below:

Regnum: Animalia

Phylum: Mollusca

Classis: Bivalvia

Ordo: Cardiida Ferussac,1822

Familia: Dreissenidae (Andrusov,1897)

Genus: Dreissena Van Beneden

Species: *Dreissena polymorpha* (Pallas,1771)

The female mussel can lay 40,000 eggs at a time, and the number of eggs she lays throughout her life can reach 1 million (Bobat et al., 2005). The spawning period may be longer in waters that are warm all year round. The incubation period of the eggs is 3-5 days. The hatched maggots or larvae swim freely in the water for up to 1 month. This period is also called the planktonic period. When the maggots or larvae hatch, they are 80-100 microns in size. The most suitable development temperatures for larvae are 20-22°C. The dispersal of the larvae generally occurs passively through currents. At the end of this period, the larvae look for a place to settle by crawling on the bottom with their feet. Meanwhile, their dimensions reach 180-290 microns. After the larva settles on the bottom, the young mussel or settled period begins. Larvae are attached to the attachment points or the base with the attachment organ, which is located on their bodies

near their feet and consists of strong filaments in the form of fringes (ZMIS, 2001). They can cling to all kinds of natural, artificial and living surfaces with their filamentous structure called byssus. These surfaces include each other's shells. They show monotypic colonization. Since planktonic larvae can enter almost anywhere water reaches, they colonize pipes and cause blockages. This situation creates very costly consequences for power plants and factories. In addition, they increase fuel consumption by adhering to the surfaces of ships and can also cause serious damage by clogging the engine and cooling pipes (Kanmaz, 2015). Although young mussels prefer hard or rocky surfaces to cling to, local mussels and adults of their own species in the waters they live in, they can also settle on vegetation. Towards the adult stage, it becomes difficult for mussels to survive in waters where water speeds are above 2 m/s.

Zebra mussels are creatures that have both water intake tubes and water release tubes and feed by filtering the water. A living creature can filter 1 liter of water a day and feed on the algae in it (URL-1, 2024). In addition, zebra mussels can create new populations by traveling more than 300 km without holding on to a place (Stoeckel et al., 1997).

### **THE EFFECT OF ZEBRA MUSSELS ON ENERGY PRODUCTION AND PREVENTION METHODS**

Rapidly spreading zebra mussels cause serious economic and ecological problems all over the world. Zebra mussels accumulate in water intakes of dam lakes for hydroelectric energy purposes, in the grids of river channel power plants, in the cofferdam covers of small HEPPs, in the sedimentation pool covers in the grids, in the valves in the loading pool, in the cooling water systems, affecting the security of energy production and reducing energy production. There may be a 7% decrease in energy production as a result of the grids being blocked by solid waste (Jones et al., 1997). This closure in front of the grids reduces the flow rate in that area and increases the accumulation of zebra mussels on the grids. If zebra mussels clog the cooling water system, it is possible that more serious damage may occur. Research has shown that the Dreissena species found in Turkey is *D. polymorpha*. However, observations and research on this subject in our country have been limited. Therefore, it is necessary to update the distribution map of zebra mussels and determine the areas where zebra mussels cause problems by examining the dams and HEPPs that have increased rapidly recently. In addition to ensuring operational safety, these studies will make it easier to take precautions in advance, depending on whether there is a mussel problem in the river basins where the dams and

HEPPs in the project phase will be built. The most important effect of this identification study will be the prevention of spread (Aksu and Yıldız, 2017).

It was stated that the occupancy rate of the Atikhisar Dam, which supplies Çanakkale's drinking and utility water, dropped to 28 percent due to drought and millions of mussels clinging to the stones in the dam bed came to light (URL-2, 2024).

Experts stated that if systematic measures are not taken against these mussels, which have spread to drinking and irrigation sources all over Turkey, some cities are at risk of dehydration and that taking precautions after they spread will be difficult and economically very expensive. According to the report prepared by the State Hydraulic Works (DSI) in 2005, it was determined that zebra mussels had spread to a total of 27 natural lakes, HEPPs and dam lakes (URL-3, 2024).

Protection methods that can be applied against zebra mussels in water bodies are summarized below:

1. High pressure water jet cleaning
2. Application of low frequency electromagnetism
3. Heat applications
4. High flow rate
5. Use of repellent building materials
6. Biological control
7. Mechanical cleaning
8. Use of chemical substances
9. Paint systems

Zebra mussels reach every leak through which water passes, and it has been stated that they cause major problems in drinking water facilities, hydroelectric power plants and dam lakes (URL-3, 2024). In particular, the operation programs of dams at risk of zebra mussels should be determined to minimize the negative effects of zebra mussels accumulating in reservoirs, water intakes and valves (Aksu and Yıldız, 2017).

## **THE EFFECT OF ZEBRA MUSSELS ON FISHING AND PROTECTION METHODS**

These mussels, which have spread uncontrollably in the last 10 years and are between 1 and 5 centimeters in length, cling to everything living or non-living that they can hold on to with the spinnerets on them, and live in colonies with their own species. Female zebra mussels, which are among the most invasive mussels in the world, lay 1 million eggs a year and over 5 million eggs in their lifetime. It is seen as a great danger that zebra mussels can completely destroy

fish breeding habitats in lakes. Zebra mussels found in Çıldır Lake are an invasive and marauding species that are very easy to transport and can survive for days in a moist environment outside of water, thus posing a danger that can completely invade the breeding habitats of carp and other fish. Zebra mussels form large colonies on sand and stones in streams. It filters the plankton in the environment. A zebra mussel has the capacity to filter one liter of water per day (URL-4, 2024).

It has been stated that due to their sharp shell structure, they break fishing nets when they breed on fishing nets, damage the reproductive stages of fish by invading fish beds, cause energy loss in hydroelectric power plants and reduce the water retention capacity in dams. Although there are physical and chemical methods of combating Zebra Mussels, chemical methods cannot be used because it is drinking water, and physical methods also cause a lot of financial loss (URL-5, 2024).

Akkus et al. (2019) stated in their study at Sarimehmet Dam in the Van Lake Basin that if the zebra mussels, which have been seen for 2 years and are known as an invasive species, spread to the rivers in the region, they may affect the pearl mullet living in Van Lake. The facility that produces rainbow trout in cages in Sarimehmet Dam buys fry and used materials from nearby provinces and uses them in the dam, carp fry brought from other provinces by official institutions are grafted to the dam, amateur fishermen use the hunting equipment they use in nearby provinces in the dam, and it is possible that zebra mussels are transported to the dam. It has been stated that there are possibilities. Adult zebra mussel individuals filter one liter of water a day and reduce the density of plankton in the environment. This situation causes the relationship between trophic levels in the aquatic ecosystem to deteriorate (Karatayev and Burlakova, 1995). Therefore, it is thought that zebra mussels, which put pressure on the plankton that forms the basis of the food pyramid, will negatively affect the trophic relationships between the creatures in Sarimehmet Dam and Karasu Stream. Thus, it has been stated that the spread of zebra mussels to aquatic ecosystems in Van province, which has important fishing resources of our country, will cause major problems in terms of fisheries management. For this reason, it was emphasized that the possible problems that the zebra mussels seen in Sarimehmet Dam may cause on the aquatic ecosystems and fish stocks in the region should be evaluated and precautions should be taken in advance. In this study, the population structure and some ecological and biological characteristics of zebra mussels, which were seen for the first time in the Sarimehmet Dam within the borders of Van province, were examined, the problems caused by the dam, their effects on the regional

fisheries in the future were discussed, and the necessary precautions to be taken were emphasized (Akkuş et al., 2019).

Since the mid-1990s, upon the increase in zebra mussel problems, physical, chemical and biological protection measures have been tried in order to determine the zebra mussel areas in Turkey and to determine the most appropriate and economical method for the protection of water bodies. In this context, the effects of physical washing measures were investigated by the General Directorate of DSI, and trial studies were carried out on the effectiveness of chlorine injection and painting methods with anti-adhesion dyes. A Bernoulli filter system trial was carried out in 2010 to determine the effectiveness of filtration in closed system irrigation. Recent Zebra Mussel prevention research in Turkey has concentrated on dams on the Euphrates River due to increasing problems. Successive dams built on the Euphrates River have turned the river waters into dam lakes more suitable for zebra mussel development, and zebra mussel accumulation problems have increased (Aksu and Yıldız, 2017).

Bobat et al. (2004) suggested that chemical, physical and biological methods could be used to combat zebra mussels. Kuşku (2022) stated that strong evidence has been obtained by using a crab species of Eurasian origin called *Potamon ibericum* as a biological control method against invasive zebra mussels that threaten fresh water resources in Atikhisar Dam Lake (Çanakkale).

Zebra mussels feed by filtering nutrients from the water, which reduces phytoplankton and zooplankton populations. This can lead to negative impacts on the balance of aquatic ecosystems and have wider ecological consequences. The decline of these organisms disrupts the aquatic balance by reducing the food source for fish and other aquatic creatures. For this reason, studies on these organisms in aquatic ecosystems have been carried out in many areas such as Halbori Springs, Cip Dam Lake, Karabey Stream, Kapıkaya Dam and Çat Dam (Saler et al., 2019; Bulut and Saler, 2020; Çağlar et al., 2022; Bulut, 2023; Bulut et al., 2024).

In conclusion; In the light of this knowledge and experience, it can be said that there is a risk of problems and precautions should be taken in facilities that produce hydroelectric energy, receive irrigation water, use or will benefit from water resources containing zebra mussels as drinking or potable water. Zebra mussels, which pose major problems for aquatic ecosystems, survive in moist environments for a long time; Necessary precautions should be taken since reasons such as transporting fish from aquaculture to another water source and amateur fishermen reusing the fishing gear they use in nearby provinces are considered possible possibilities in the transportation of zebra mussels.

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## Chapter 9

### Turkey Endemic Fish Species and Characteristics Belonging to the Cobitidae Family

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## INTRODUCTION

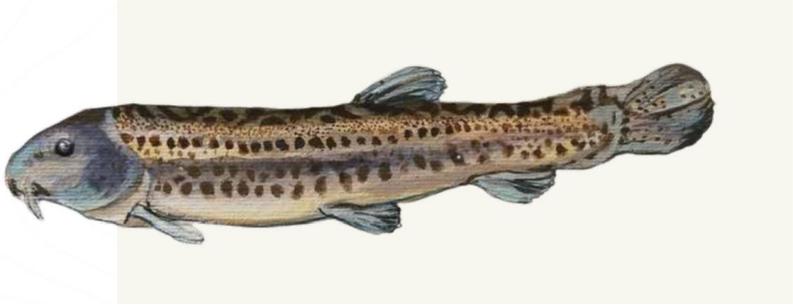
Members of the Cobitidae Family can be found in many different aquatic environments, including stagnant and river ecosystems, due to their high ecological resistance. They are quite resistant to lack of oxygen and can survive even in very polluted waters. When environmental conditions become abnormal and the amount of oxygen in the water drops to a noticeable level, they rise to the water surface and swallow air bubbles, and in such cases, their intestines act as respiratory organs (Geldiay and Balık, 2007). Members of the genus *Cobitis*, one of the genera belonging to the Cobitidae family, have a relatively elongated and laterally flattened body structure. There are no scales on the head, which is narrow and high-shaped. Of the three pairs of whiskers around the mouth, two pairs grow from the tip of the nose and one pair from the corner of the mouth. The eyes are relatively small and there is a suborbital spine under the eye, sometimes buried under the skin (Geldiay and Balık, 2007). It is generally found in clean and cool waters of streams and on sandy and gravelly grounds where the flow rate is slow (Seçer, 2018).

Members of the Cobitidae family are widely distributed throughout Asia, Europe and Africa. There are 207 species belonging to the Cobitidae family reported in the world, and in Turkey, this family includes 22 species (*C. battalgili*, *C. bilseli*, *C. damlae*, *C. dorademiri*, *C. elazigensis*, *C. evreni*, *C. fahirae*, *C. kellei*, *C. levantina*, *C. phrygica*, *C. pontica*, *C. puncticulata*, *C. simplicispina*, *C. sipahilerae*, *C. splendens*, *C. strumicae*, *C. turcica*, *C. vardarensis*, *Misgurnus fossilis*, *Sabanejewia aurata*, *S. balcanica*, *S. caspia*) are represented by species (Çiçek et al., 2015; Froese and Pauly, 2017). Of these species, *C. battalgili*, *C. bilseli*, *C. damlae*, *C. dorademiri*, *C. evreni*, *C. fahirae*, *C. kellei*, *C. phrygica*, *C. puncticulata*, *C. simplicispina*, *C. sipahilerae* and *C. splendens* are for Turkey endemic species (Çiçek et al., 2015; Erk'akan et al., 2017; Seçer, 2018).

### 1. *Cobitis battalgili* Băcescu, 1962:

It is demersal and lives in Central Anatolia (EN). This species is found in three streams in the Beyşehir basin (Çeltek Stream to the north, Eflatun Pınarı Stream to the east, İlırmak Stream to the south) and Manavgat Stream in the Mediterranean Basin. Population Trend: Species habitat size is constantly decreasing. Economic value: None Habitat and ecology: Water flowing in gravel, sand and loam beds, generally prefers dense underwater vegetation. Threats: Although the Manavgat population is not well known, it is affected by a large dam. This species lives in slightly or moderately polluted streams in the Beyşehir basin; The habitat is reasonable and still supports widespread survival

of these species. Conservation actions: There are currently no conservation actions for these species. Protection of the region, management of water resources and awareness of the increase in alien species and its control are the recommended activities. Additionally, the distribution of this species should be better elucidated by some field studies (Freyhof, 2014a) (Figure 1).



**Figure 1.** *Cobitis battalgili* (URL-1).

## **2. *Cobitis bilseli* Battalgil 1942:**

It is demersal and lives in Beyşehir Lake and the streams flowing into Beyşehir Lake. (EN). It is known to occur in Sariöz and Sariçay Streams extending to the northeast of Beyşehir Lake (Freyhof, 2014b). Population Trend: The species is abundant in all regions where it is present and population trends are unknown. Populations are threatened and there is likely a negative population trend. Economic value: This species is not a species for fishing or any other use. Habitat and ecology: It prefers gravel and sand ground in streams with dense vegetation. Threats: The streams and channels where these species live are unpolluted or lightly polluted, the quality of life is fair, and they can still support large eel populations. Due to pike perch, there is a decrease in the diversity of native fish in the lake. Sudak does not enter streams where eels are found. In recent years, alien species *Pseudorasbora parva*, *Knipowitschia caucasica* and *Alburnus escherichii* have increased the population of the lake and the streams where native Beyşehir Lake fish species are found, and their effects on native species are unknown. Water withdrawals are common in streams where the species is found, and water withdrawals for agricultural activities are expected to increase. The lake and some streams are polluted by untreated waste and agricultural waste from nearby villages, but the pollution level is still moderate. Conservation actions: A large part of Beyşehir Lake (88,750 hectares) is a national park, but this protection has no effect on the invasion of alien species into the lake and the streams flowing into the lake. There is an urgent need for a program to assess water flow as well as

biodiversity of all streams in the Beyşehir Lake basin (Freyhof, 2014b) (Figure 2).



**Figure 2.** *Cobitis bilseli* (URL-2)

**3. *Cobitis damlae* Erk'akan & Özdemir 2014:**

It is demersal and lives in the Dalaman stream (EN). Head length is longer than body depth. There are no accessory lobes on the pectoral and pelvic fins bases. The pelvic fin origin is opposite. The tip of the pelvic fin does not reach the anus. The distal anal margin of the fin and the caudal fin are almost flat. The head is elongated and compressed. Head length is longer than head depth. Eye capsules are large but non-functional (Erkakan and Özdemir, 2014) (Figure 3).



**Figure 3.** *Cobitis damlae* (Erkakan and Özdemir, 2014).

**4. *Cobitis dorademiri* Erk'akan, Özdemir ve Özeren, 2017:**

It is demersal and lives in Balıklı stream and Köyceğiz basin (EN). The body is high built and flattened from the sides. The head, which is short and blunt, is 4.7-5.1 (4.8) times the standard size. The eyes are relatively large and are located 5.4-18 5.9 (5.7) times on the head. The lower-positioned mouth is bow-shaped and has 3 pairs of whiskers around it. The pelvic fin begins at the level of the 4th or 5th ray of the dorsal fin and does not extend to the anus. The anal fin does not extend to the caudal fin. The caudal fin is flat. The tail stalk is short

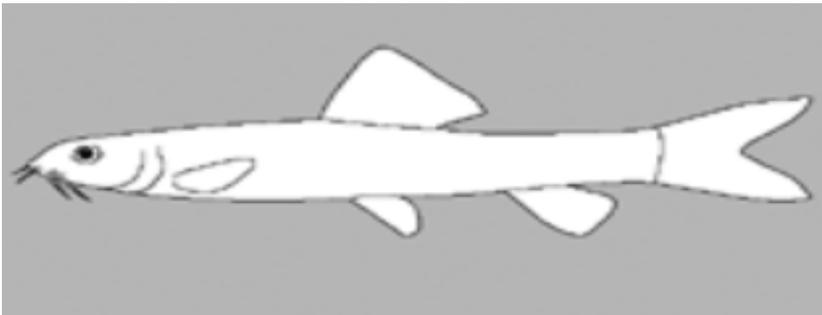
and flat, 8.5-8.8 (8.7) times the standard length. The distance between the eyes is short. Whiskers are relatively long. Maxillary whiskers are longer than the inner rostral and outer rostral whiskers. The lateral line is not complete but extends to the base of the pectoral fin. In males, the body is smaller than in females, and the pectoral and pelvic fins are larger. There are 2 lamina circularis in male individuals. There are 4 gambetta zones. In living individuals, the abdomen and lower part of the head are yellowish white. The body is yellowish brown. There are 5-6 large spots in the predorsal and 5 large spots in the postdorsal. The head is covered with small brown spots. There is no spotting on the pelvic and anal fins. While banding varying between 5-6 rows is seen on the dorsal fin, irregular shaped spotting is seen on the caudal fin (URL-3) (Figure 4).



**Figure 4.** *Cobitis dorademiri* (URL-3)

**5. *Cobitis evreni* Erk'akan, Özeren & Nalbant 2008 :**

It is benthopelagic and lives in the Ceyhan river basin, Kömür stream (EN). There is no photograph in fishbase (Figure 5).



**Figure 5.** *Cobitis evreni* (URL-4)

**6. *Cobitis fahirae* Erk'akan, Atalay-Ekmekçi & Nalbant 1998:**

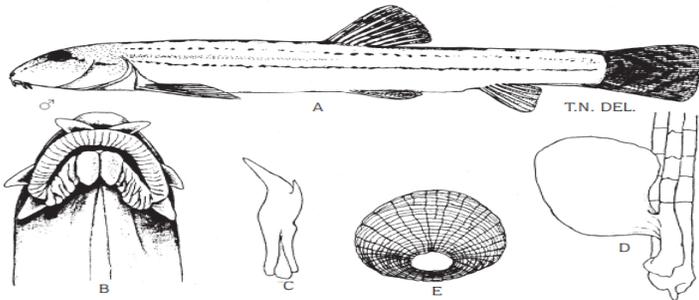
It is demersal and lives in the Küçük Menderes River (LC). It has a high and compressed body with four Gambetta's pigment zones. The black tail spot is round and quite small. The lobes are distinct but not large (URL-5) (Figure 6).



**Figure 6.** *Cobitis fahirae* (URL-5)

**7. *Cobitis kellei* Erk'akan, Atalay-Ekmekçi & Nalbant 1998:**

It is demersal and lives in Göksu (Dicle) stream (CR). Moderately long head with relatively small eyes located on the front half. The mouth is highly arched, with three pairs of very short spines. Both lips are frowning. Lobes are well developed. The suborbital spine (small spine) with laterocaudal processes is reduced. Sexual dimorphism based on well developed at the base of the second pectoralis lamina circuisis ray in males. Color pattern pigment zones are present with four Gambetta, but the third is reduced. The Fourt zone is created with minute rounded dots. There is a small brownish spot at the base of the caudal fin (Erkakan et al., 1999) (Figure 7).

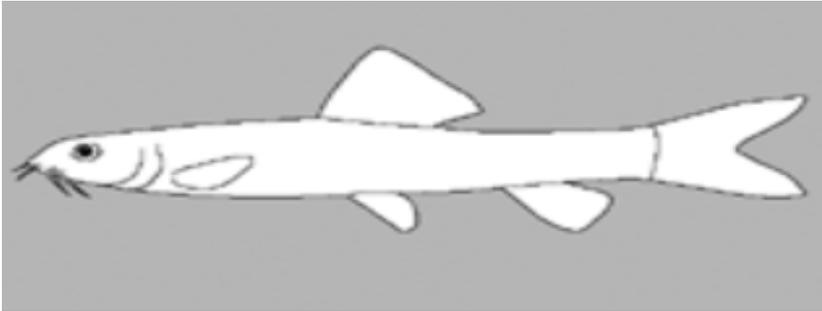


**Figure 7.** *Cobitis kellei* (Erkakan et al., 1999)

**8. *Cobitis kurui* Erk'akan, Atalay-Ekmekçi & Nalbant 1998:**

It is demersal and lives in the Büyük Menderes River and Yuvarlakçay (LC). Black spot smaller in size, brownish and reduced lateral spots (pigment area of the fourth Gambetta) at the caudal base. The mouth is curved and has three pairs

of very short pointed tips. It is available in men with developed circular lamina (URL-6). There is no photo on fishbase (Figure 8).



**Figure 8.** *Cobitis kurui* (URL-6)

**9. *Cobitis phrygica* Battalgazi 1944:**

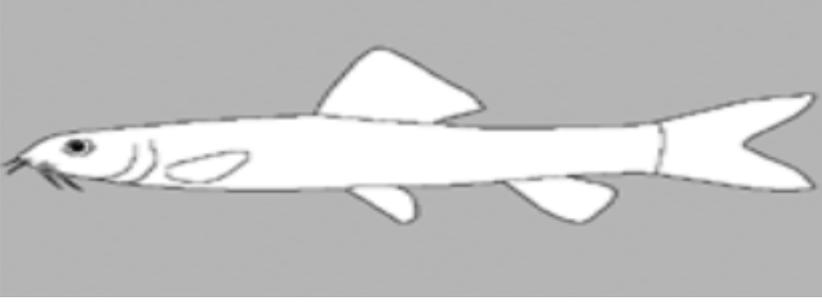
It is demersal and lives in brackish and fresh water. It lives in Acıgöl in the Aegean region (EN). It is a sensitive species that lives in a narrow area in the world (Figure 9).



**Figure 9.** *Cobitis phrygica* (URL-7)

**10. *Cobitis simplicispina* Hankó 1925:**

It is benthopelagic and lives in the Acıgöl (Afyon), Porsuk stream and Sakarya river systems (LC). *Cobitis simplicispina* is not familiar to people in Turkey because they have no economic value. Moreover, their coloring provides good camouflage and they are unattractive due to their benthic niches, so detailed information about their life history is not available (Ekmekçi and Erkakan, 2003). There is no photograph in fishbase (Figure 10).



**Figure 10.** *Cobitis simplicispina* (URL-8)

**11. *Cobitis sipahilerae*** Erk'akan, Özdemir & Özeren 2017:

It is demersal and lives in the Aksu river basin (NE). *Cobitis sipahilerae* differs from other species of *Cobitis* species by the lack of the 5th gambetta zone. Gambetta zone, third zone similar to first zone but smaller, fourth zone triangular or square-like spots, in some specimens spots merged into a stripe; mouth curved, lips slightly frowned, mental lobe moderately marked and covered with papillae and thinner caudal peduncle (Erkakan et al., 2017) (Figure 11).



**Figure 11.** *Cobitis sipahilerae* (Erkakan et al., 2017)

**12. *Cobitis splendens*** Erk'akan, Atalay-Ekmekçi & Nalbant, 1998:

It is demersal and lives in the streams flowing into the Black Sea between Akçakoca and Ereğli (CR). The first place it was found was in small streams flowing into the Black Sea. Its Turkish name is stone-eating fish and its English name is Splendid spined loach. It was detected at İyidere station in Rize province (Bayçelebi et al., 2017) (Figure 12).



**Figure 12.** *Cobitis splendens* (Bayçelebi et al., 2017)

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## **Chapter 10**

### **Effects Of Soil Zinc Application On Phytic Acid/Zinc Ratio And Phytase Activity In Grains Of Different Cereal Species**

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## **Abstract**

Studies to reduce the phytic acid (PA) / zinc (Zn) molar ratio in different grain species to reasonable values are increasingly important. This study focused on the examine the effect of increased Zn levels on the PA and Zn concentrations, PA to Zn molar ratio and phytase activity in the grains bread wheat, durum wheat, triticale, barley, rye and oat varieties grown in Zn deficient soil. For this, four levels of Zn as 0, 30, 60 and 90 kg Zn ha<sup>-1</sup> from ZnSO<sub>4</sub> 7 H<sub>2</sub>O were applied to the soil. Based on the results, it was seen that Zn applications increased grain Zn concentrations and phytase activity in al cereal species. On the contrary, Zn application decreased grain phosphorus (P) and PA concentration and PA to Zn molar rate. Also results showed that, the parameters examined showed significant variations depending on the cereal species. From these results, it can be concluded that Zn fertilization could be a good option to decrease PA to Zn ratio in the grains regardless of the cereal species.

**Key words:** Cereal species, grain zinc, phytase activity, phytic acid, zinc fertilization

## TOPRAKTAN ÇİNKO UYGULAMASININ FARKLI TAHIL TÜRLERİNİN TANE FITİK ASİT/ÇİNKO ORANI VE FITAZ AKTİVİTESİ ÜZERİNE ETKİLERİ

### Özet

Farklı tahıl türlerinde fitik asit (PA) / çinko (Zn) molar oranının makul değerlere düşürülmesine yönelik çalışmalar giderek önem kazanmaktadır. Bu çalışma, Zn eksikliği olan topraklarda yetiştirilen ekmeklik buğday, makarnalık buğday, tritikale, arpa, çavdar ve yulaf çeşitlerinde artan Zn seviyelerinin PA ve Zn konsantrasyonları, PA/Zn molar oranı ve fitaz aktivitesi üzerindeki etkisini incelemeye odaklanmıştır. Bunun için toprağa  $ZnSO_4 \cdot 7 H_2O$ 'dan 0, 30, 60 ve 90 kg Zn ha<sup>-1</sup> olmak üzere dört seviyede Zn uygulanmıştır. Elde edilen sonuçlara göre, Zn uygulamalarının tahıl türlerinde tane Zn konsantrasyonlarını ve fitaz aktivitesini artırdığı görülmüştür. Buna karşın, Zn uygulaması tane fosfor (P) ve PA konsantrasyonunu ve PA/Zn molar oranını azaltmıştır. Ayrıca sonuçlar, incelenen parametrelerin tahıl türlerine bağlı olarak önemli farklılıklar gösterdiğini ortaya koymuştur. Bu sonuçlardan, Zn gübrelemesinin tahıl türlerinden bağımsız olarak tanelerdeki PA/Zn oranını azaltmak için iyi bir seçenek olabileceği sonucuna varılabilir.

**Anahtar kelimeler:** Tahıl türleri, tane çinkosu, fitaz aktivitesi, fitik asit, çinko gübrelemesi

## INTRODUCTION

Phytic acid is natural plant compound and storage form of phosphorus. It is found in most cereal and legume grains, nuts, oilseeds, tubers, pollen, spores, and organic soils. During germination, PA is used for biosynthetic requirements of young growing tissues. Although it's critical role as reserve P for seed germination and in many physiological processes in growth, it is well reported that PA has the ability to bind minerals, proteins etc. So, high concentrations of PA in seeds can prevent mineral bioavailability by complexing each other (Ferguson et al., 1989; Imran and Rehim, 2017). Next to the other minerals, such as Ca, Na, Mg, Fe, Mn and Cl, Zn is the most affected mineral by PA because it forms the most stable and insoluble complex (Oatway et al. 2001). Phytic acid concentration in seeds is closely related to plant P nutrition and P concentration in seeds (Su et al., 2018; Taliman et al., 2019). The concentration of PA in seeds is highly dependent on the rate of P uptake and its translocation from leaves into seeds. Phytic acid concentrations in grain of many cereals and legumes greatly vary with environmental factors, agricultural practices, fertilizer application etc. But genotypic differences are the other main factors influencing grain PA concentrations (Ning et al., 2009; Ahn et al., 2010; Taliman et al., 2019; Fukushima et al., 2020). As indicated in different studies, PA concentrations changed between 0.83-2.2% in corn, 0.39-1.35% in wheat, 0.50-1.89% in triticale, 0.74- 2.10 in beans, 0.54-1.46 in rye and 0.28-1.26 in chickpeas (Lolas et al., 1976; Reddy et al., 1982). The proportion PA in total seed P can be up to 85 % in cereals and legumes. There is a close relation between PA and Zn in terms of bioavailability of Zn in foods. For bioavailability of Zn to humans and animals, molar ratio between PA and Zn is more important than their concentrations in the diets. The molar ratio of PA to Zn is an indicator of Zn bioavailability in the diets. It was indicated that when PA to Zn molar ratio was above 25-30 bioavailability of Zn decreased sharply (Oberleas and Harland, 1981). Generally, Zn bioavailability is thought to decrease when the PA to Zn molar ratio is higher than 15 (Gargari et al., 2007). So, the effective strategy to increase Zn bioavailability may be to reduce PA concentration in grains with decreased P fertilization. But while doing this, the role P in plant shouldn't be forgotten. Decreasing P application can led to many growth retarding problems due to its role in many biochemical processes from seed germination to growth end. For these reasons, P fertilization is indispensable practice in agricultural production. Therefore, the best and the fast way to decrease PA to Zn ratio until favorable value is to increase grain Zn concentration rather than decrease grain P concentration. For this, Zn bio fortification is generally considered to increase

bioavailability of Zn in cereal grains by means of different Zn fertilization techniques (Cakmak et al., 2010; Imran et al., 2016). At the same time, in different studies it was reported that, Zn application led to decrease of P and PA concentrations in grain next to increase in Zn. And this caused significant reduction in PA to Zn molar ratio (Erdal et al. 2002; Das et al., 2019).

Bioavailability of Zn in foods is highly depended on phytase activity. Phytases are enzymes that degrade phytate and permit higher availability of Zn and other mineral nutrients. As found within genotypes of triticale, activity of phytase shows an important genotypic variation (Singh and Sedeh, 1979). In samples of wheat cultivars phytase activity ranged between 206 and 775 mU g<sup>-1</sup> (Barrier-Guillot et al., 1996). There are different results on the effect of Zn fertilization on phytase activity of the cultivars. Although, in some publications it was reported that Zn nutrition has no direct effect on phytase activity (Kaya et al., 2009; Erdal et al., 2014), Moshfeghi et al., (2019) indicated that there is a positive correlation between grain phytase activity and grain Zn concentration.

In this study, it was aimed to investigate the effect of increasing Zn application on grain PA to Zn ratio and phytase activity of different cereal varieties. And also we aimed to compare the effectiveness of Zn fertilization on PA to Zn ratios in grains of different cereal varieties.

## **MATERIALS AND METHODS**

### *Experimental site*

Study was conducted as field experiment. Some characteristics of the experimental area were as follows: texture is clayey (Bouyoucos, 1951), pH is slightly alkaline (7.9, in 1: 2.5 soil to water suspension), organic matter content is low (1.2 %, Walkley and Black, 1934), lime is very high (32% CaCO<sub>3</sub>, Allison and Moodie. 1965), NaHCO<sub>3</sub> extractable P concentration is sufficient (20 mg kg<sup>-1</sup>, Olsen et al. 1954), DTPA extractable Zn concentration is very low (0.1 mg kg<sup>-1</sup>, Lindsay and Norvell, 1978).

### *Plant materials*

As plant materials 6 cereal species were used. The species and their varieties used for the experiments were as follows: bread wheat (*Triticum aestivum*, cvs. Gerek 79), durum wheat (*Triticum durum*, cvs. Kunduru), triticale (*xTriticosecale* Wittmark, cvs.BDMT-19), barley (*Hordeum vulgare*, cvs. Tokak 157/37), rye (*Secale cereale*, cvs. Aslim) and oat (*Avena sativa*, cvs. Checota).

### *Set up of experiment and fertilization*

Plot sizes were 7.2 m<sup>2</sup> (6x1.2) and each plots consisted of 6 rows with 20 cm spaces. Sowing rate was 550 seeds m<sup>-2</sup>. Four levels of Zn (Zn0: 0, Zn1: 30, Zn2: 60, Zn3:90 kg Zn ha<sup>-1</sup>) from ZnSO<sub>4</sub> 7 H<sub>2</sub>O were sprayed to the soil surface then

incorporated in to the soil with disk-plowing before seed sowing. Next to the Zn fertilization, 30 kg P ha<sup>-1</sup> as triple-superphosphate and 25 kg N ha<sup>-1</sup> as di-ammonium phosphate were given as basal fertilization. Also 55 kg N ha<sup>-1</sup> as ammonium sulphate was applied as top dressing on early spring.

### ***Seed chemical analysis***

#### ***P and Zn analysis***

Grains were dried at 70 C<sup>0</sup> until stable weight in drying oven then milled for analysis. For Zn and P analysis, 0.25 g milled grain samples dry-ashed at 550 °C dissolved with 3 ml 3.3% (v/v) HCl. Zinc was measured using an atomic absorption spectrometer and P was measured using spectrophotometer according to the vanadate molybdate colorimetric method (Jones et al. 1991).

#### ***Phytic acid (PA) measurements***

0.5 g of ground seed sample was put in a 100 ml erlenmayer flask and 25 ml of 0.2 N HCl (pH≤=0.3) was added and shaken during 3 h. At the end of the 3 hours, 25 ml pure water was added and the final volume was brought up 50 ml and shaken by hand. And colored with bi-pyridine solution and the color was read at 519 wavelengths with spectrophotometer (Haug and Lantzsich 1983).

#### ***Phytase activity***

One gram ground seed sample was put in 50 ml volumetric glass flask. Then 10 mL 50 mM sodium citrate buffer (pH= 5.3) and 50 mM Na-phytate was added on the samples and left for 1 h. incubation at 37 C<sup>0</sup> in hot water bath. Then the activity of phytase was measured as described by Scheuermann et al. (1988).

#### ***P amount in PA (phytate-P)***

The rate of P in PA is 3.55. This was calculated by dividing molecular weight of PA (660 g) to molecular weight of P in PA (6x31=186).

#### ***The rate of phytate-P in total seed P***

After the units are equalized, the rate of Phytate-P rate in total seed is found

#### ***PA to Zn ratio***

The molar ratio of phytic acid to Zn was calculated by dividing millimoles of phytic acid with millimoles of Zn

#### ***Statistical evaluation***

Data were subjected to ANOVA using MINITAB 16 statistical package program. Means were compared using Tukey test at 95 % confidence level.

## RESULTS AND DISCUSSION

### *Effect of Zn fertilization on Zn, P and PA concentrations*

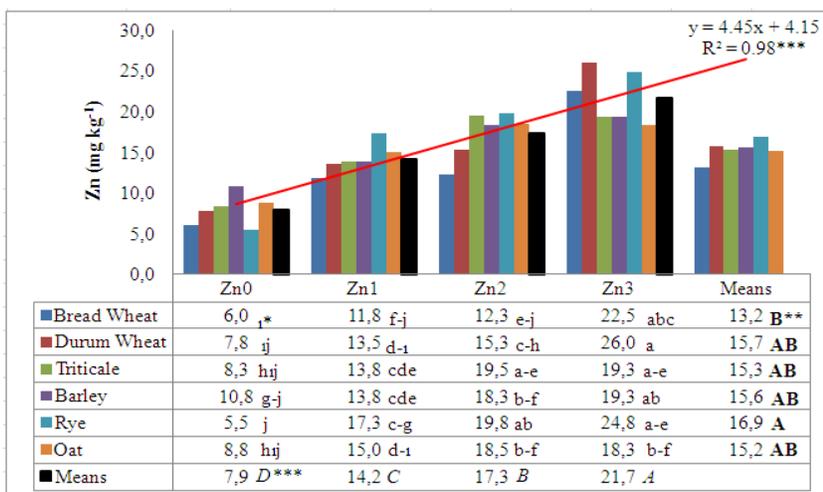
**Table 1.** Analysis of variance for the examined parameters.

Source	f	F values						
		Zn	P	PA	Phytate-P	Rate of Phytate-P in total P	P A/Zn	Phytase
Zn doses (A)		110***	69.75***	42.8***	42***	ns	185***	8.5***
Species (B)		3*	2.7*	5.3***	5.3***	ns	4.3**	128***
AxB	5	3.3***	3.3**	2.2*	2.3*	2.9**	5***	5.4***
Error	2							

\*:P<0.05, \*\*: P<0.01, \*\*\*:P<0.001, ns: non-significant, Df: degree of freedom

PA: Phytic acid, PP: Phytate phosphorus,

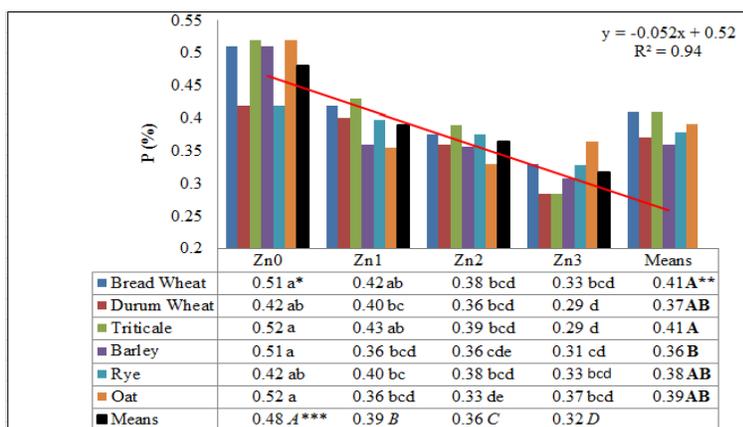
Grain Zn concentration was significantly affected by individual factors and their interaction (Table 1). Zinc concentrations in grains ranged from 6 to 10.8 mg kg<sup>-1</sup> under Zn0 condition. With increased Zn applications these variations were between 11.8 and 17.3 mg kg<sup>-1</sup> under Zn1 (30 kg Zn ha<sup>-1</sup>), 12.3 to 19.8 mg kg<sup>-1</sup> under Zn2 (60 Zn ha<sup>-1</sup>) and 18.3 to 26.0 mg kg<sup>-1</sup> under Zn3 (90 Zn ha<sup>-1</sup>) conditions (Fig.1) Increasing amount of soil Zn applications significantly increased grain Zn concentrations by 80 % under Zn1 dose, 119 in Zn2 dose and 175 % in Zn3 dose when compared to control (Zn0). Significant variations were seen in grain Zn concentrations of different cereal species. But, only the variation between bread wheat and barley grain Zn concentrations was found to be significant. Zinc concentrations in the grains of other species took place in the same statistical group. Increases of grain Zn concentration between Zn0 and Zn3 applications were about 275, 233, 135, 79, 351 and 108% for bread wheat, durum wheat, triticale, barley, rye and oat respectively. Looking at the results, it can be clearly seen that there is a significant positive correlation between Zn dosage and grain Zn concentrations. Looking at Zn concentrations under Zn deficient conditions and its increment rates with Zn applications, we can see the genotypic differences among the genotypes.



\*Shows interaction effects, \*\*Shows the differences among the cereal varieties\*\*\* Shows the differences among Zn dosages. There is not a significant differences shairng the same latters.

**Figure1.** Effect of Zn fertilization on grain Zn concentrations of different cereal species

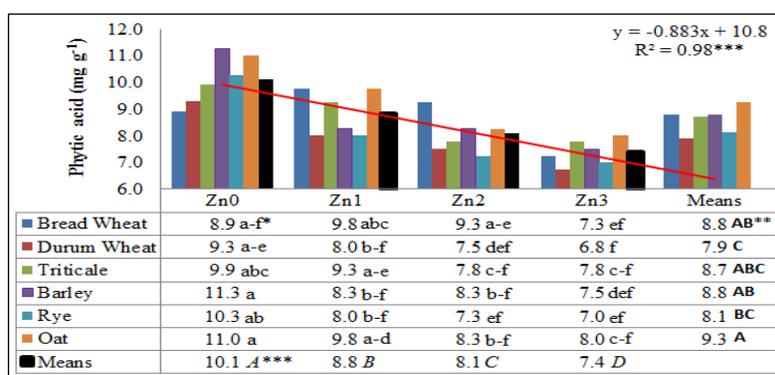
Phosphorus concentrations in grains significantly affected by the individual factors (Zn doses and species) and their interaction (Table 1). Depending on the interaction, the lowest P concentration was found in the grains of durum wheat and triticale as 0.29 % in Zn3 conditions, the highest were found in the grains of triticale and oat as 0.52% from the control (Zn0) treatments (Fig. 2). Mean values showed that P concentrations in grains of different species were in the same statistical group mostly, but P in grains of barley significantly varied from the bread wheat and triticale. Grain P concentrations of all individual species decreased with the increased Zn concentrations. These reduction rates from Zn0 to Zn3 for bread wheat, durum wheat, triticale, barley, rye and oat were 35, 31, 44, 39, 21 and 29 %, respectively. Avarage decrease was calculated as 33 %.



\*Shows interaction effects, \*\*Shows the differences among the cereal varieties\*\*\* Shows the differences among Zn dosages. There is not a significant differences shairng the same letters.

**Figure 2.** Effect of Zn fertilization on grain P concentrations of different cereal species

Phytic acid concentrations in grains varied between 6.8 (Zn3xdurum wheat) to 11.3 mg g<sup>-1</sup> (Zn0xbarley) depending on Zn application x species interaction. Zinc applications played a significant role on these variations for each species and for the mean values. Grain phytic acid concentrations decreased with Zn applications in all cereal grains (Fig. 3). The decreases between control and the highest Zn doses (90 kg Zn ha<sup>-1</sup>) were 18, 27, 21, 34, 32 and 27 for durum bread wheat, durum wheat, triticale, barley, rye and oat % respectively and the mean decrease was 27%.

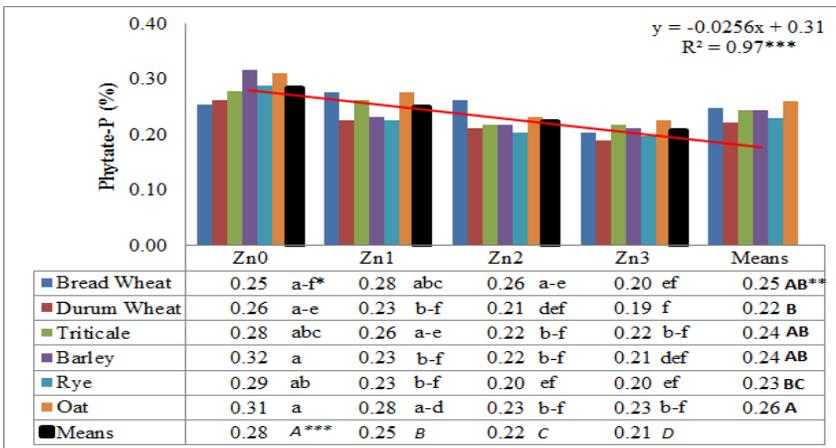


\*Shows interaction effects, \*\*Shows the differences among the cereal varieties\*\*\* Shows the differences among Zn dosages. There is not a significant differences shairng the same letters.

**Figure 3.** Effect of Zn fertilization on grain phytic acid concentrations of different cereal species

*Phytate-P and the rate of phytate-P in total P*

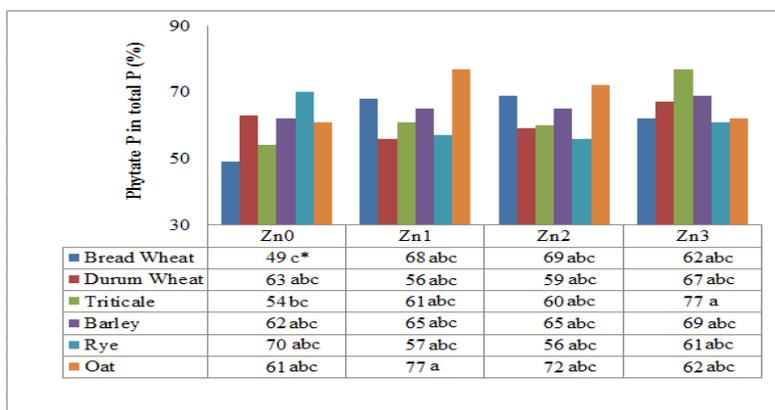
Amount of P as PP significant varied with Zn applications, cereal species and their interactions. The lowest PP (0.19 %) was found in the grains of durum wheat under Zn3 condition, but the highest (0.32 %) was found in the grains of barley under Zn0 condition (Fig.4). Increased Zn dosages led to decreases in phytate-P in all species as in P and PA. Also, mean values showed the same decreasing trend. The average decrease from Zn0 to Zn3 was determined as 25 %. As in P and phytic acid concentrations, phytate-P amounts varied with cereal species. The highest phytate-P concentration was found in the seeds of oat, but the lowest was found in the seeds of durum wheat. Rate of Phytate-P in total P significantly affected by the interaction of Zn doses and cereal species. As given in Fig. 5, the rate of Phytate-P in total seed P varied from 49 % (Zn0 x bread wheat) to 77 % (Zn3 x triticale). As average, 63 % of the total P was in phytic acid form.



\*Shows interaction effects, \*\*Shows the differences among the cereal varieties\*\*\* Shows the differences among Zn dosages.

There is not a significant differences sharing the same letters.

**Figure 4.** Effect of Zn fertilization on phytate-P of different cereal species



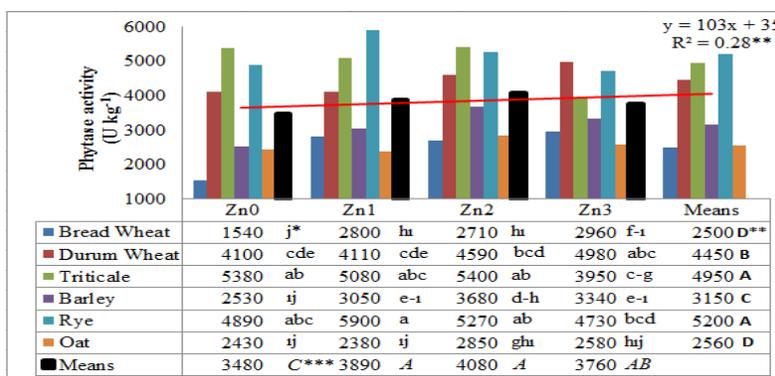
\*Shows interaction effects, \*\*Shows the differences among the cereal varieties\*\*\* Shows the differences among Zn

dosages. There is not a significant differences shairng the same letters.

**Figure 5.** Effect of Zn fertilization on phytate-P of different cereal species

### Phytase activity

Phytase activity significantly ranged from 1540 to 5900 U kg<sup>-1</sup> with an average of 3801 U kg<sup>-1</sup>. Increased application of Zn enhanced the phytase activity regularly until 60 kg Zn ha<sup>-1</sup> (Zn2) from 3480 to 4080 U kg<sup>-1</sup>, but later decreased again to 3760 U kg<sup>-1</sup> (Fig.6). Significant correlation efficiency was found between the Zn levels and phytase activity. Similar results and significant correlations were found in different studies (Liu et al., 2006; Moshfeghi et al., 2019). Phytase activity varied with cereal species. While the highest activities were found in the seeds of rye (5200 U kg<sup>-1</sup>) the lowest activity was measured in bread wheat (2500 U kg<sup>-1</sup>).

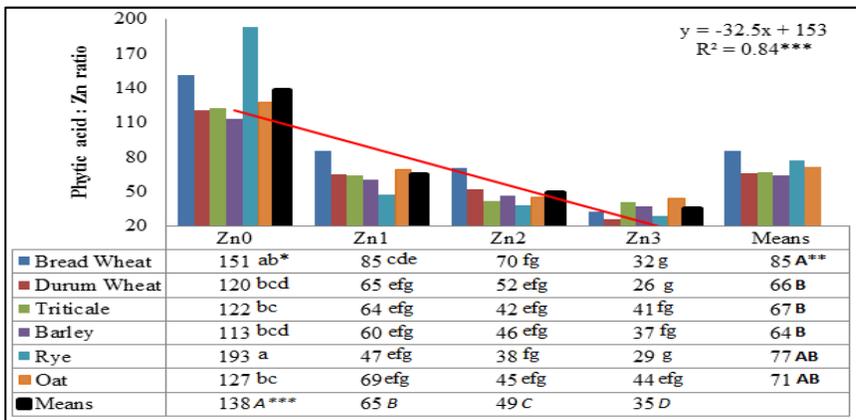


\*Shows interaction effects, \*\*Shows the differences among the cereal varieties\*\*\* Shows the differences among Zn dosages. There is not a significant differences shairng the same letters.

**Figure 6.** Effect of Zn fertilization on phytase activity in different cereal species

### PA/Zn molar ratio

Phytic acid to Zn molar ratio showed great variations ranging from 26 (Zn3 x durum wheat) to 193 (Zn0 x rye) depending on Zn doses x species interaction. Looking at the whole species, it can be seen that PA to Zn ratio decreased sharply with increased Zn dosages. While the PA to Zn molar rates were 151, 120, 122, 113, 193 and 127 in the grains of bread wheat, durum wheat, triticale, barley, rye and oat under Zn0 condition, these rates decreased to 32, 26, 41, 37, 29 and 44, respectively (Fig.7). Significant negative correlation was found between the Zn dosages and PA to Zn molar ratio. Phytic acid to Zn molar ratios decreased from 138 to 65, 49 and 35, respectively with Zn dosages. Thus, 53, 64 and 75 % reductions were recorded compared to control. Phytic acid to Zn molar ratio varied also with the cereal species.



\*Shows interaction effects, \*\*Shows the differences among the cereal varieties\*\*\* Shows the differences among Zn dosages. There is not a significant differences sharing the same letters.

**Figure 7.** Effect of Zn fertilization on phytic acid to Zn ratio in different cereal species

## DISCUSSION

Zinc fertilization has increased the grain Zn concentration, although it varies with the cereal species. So, it can be said that Zn fertilization is very effective to increase grain Zn concentrations as indicated in many studies conducted on different plant species (Cakmak and Kutman, 2018; Dhaliwal et al., 2019, Grewal et al., 2020; Lu,). In previous studies it was documented that genotypic differences in Zn uptake and translocation capacities among the cereals affected grain Zn concentrations (Gomez-Coronado et al., 2016; Dhaliwal et al., 2019).

Zinc applications led to decrease grain P concentration. Similar decreases of P in shoot and grains of different plants depending on Zn fertilization have been

recorded in previous studies (Drissi et al., 2015; Balai et al., 2017; Manzeke et al., 2017). The decrease in P concentrations due to Zn applications may be due to the antagonistic reaction between Zn and P in the uptakes and translocations of P. Their interactions in soils may also be partially responsible for this (Zhu et al., 2001). Application of Zn may decrease solubility and availability of P by making Zn-phosphate complexes (Ghoneim, 2016; Mansuri et al., 2019). Although, PA concentrations in the seeds of cereal species varied with the species (Oatway et al., 2001), PA values were in the ranges indicated by previous studies. Hídvégi and Lásztity (2002) indicated that phytic acid concentrations in the seeds of some cereals varied from 5.2 (durum wheat) to 14.2 mg g<sup>-1</sup> (oat). Results indicated clearly that PA concentration in the grains increased with the grain P concentrations but decreased with the Zn fertilization. When looked at this decreases, it can be seen that there is a close similarity with P decrease tendency. Similar variations of PA concentrations in the grains depending on the Zn applications as P can be result this close relation. It has been reported that the PA concentration in the grains of different plants is increased by P application and there is a close positive correlation between total P and PA (Raboy 2009; Yang et al., 2011). As in P and PA, PP decreased with Zn fertilization. These decrease of can be explained with the natural consequence of P and PA decrease resulted from Zn fertilization (Kaya et al., 2009; Erdal et al., 2014). Phytate- P amount in total P measured in this study are in the accordance with the previous studies (Lolas et al., 1976; Mate and Radomir, 2002; Raboy, 2009; Erdal et al., 2014).

It was found that phytase activity varied with the cereal species. Similar results indicating different phytase activity depending on the genotypic differences in different plant species has been presented (Barrier-Guillot et al., 1996; Lee et al., 2014). The effect of genotypic differences on phytase activity may be explained by the gene dependent synthesis of the phytase enzyme (Dionisio et al., 2011). These showed that phytase activity is largely controlled by intrinsic factors of genotypes. In the literature there are different findings about phytase activity and phytic acid correlations in the seeds. While Lee et al., (2014) found a negative correlation between them; no correlation was indicated by Kaya et al. (2009).

Phytic acid to Zn molar ratios decreased with the increases in Zn levels as a result of Zn fertilizations as indicated previous studies (Wang et al., 2015; Zhao et al., 2018; Zhao et al., 2019)

At the same time, PA to Zn molar ratio showed variations with cereal species. These can be related to differences in the Zn uptake capacity and other inherent genetically factors playing the role on PA to Zn ratio (Welch and Graham, 2004; Tavajjoh, et al., 2011). Looking at the phytic acid to Zn values under Zn deficient

conditions, it can be said that Zn in the grains is not bioavailable or has quite low bioavailability when considered the critical ratios indicated in previous reports (Oberleas and Harland, 1981; Ryan et al., 2008). In cereal based foods having PA to Zn molar ratios greater than 20, bioavailability of Zn is very low (Zhou and Erdman, 1995). It was indicated that when PA to Zn molar ratio was above 25-30, bioavailability of Zn decreased sharply (Oberleas and Harland, 1981).

As conclusion, decreasing grain P concentrations with lower P fertilization or with other attempts can be effective on to decrease grain PA concentrations. But this way can led to many troubles in many physiological processes from germination to harvest due to lack of P. So, the best way to decrease PA to Zn molar ratio can be Zn fertilization. Also regardless of the cereal species, Zn fertilization increased grain Zn concentration and thus PA to Zn molar ratios until favorable values.

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## Chapter 11

### The Potential Use Of Remote Sensing Technologies And Unmanned Aerial Vehicles (UAVs) In Agricultural Fields

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## **Introduction**

Chronic hunger impacts 815 million individuals globally. Asia houses 64% of the global population suffering from hunger. Research indicates that to satisfy the food requirements of a projected global population exceeding 9 billion by 2050, current production must increase by 50% (FAO, 2018; Sylvester, 2018). The agriculture sector must address the impacts of climate change, with diminishing production regions and clean natural resources, to meet these demands. Addressing these difficulties necessitates the swift analysis of data gathered from extensive regions and the immediate transmission of information in agricultural production zones, together with the execution of critical interventions.

Advancements in information and communication technology are increasingly vital in addressing the myriad challenges confronting agriculture. The attainment of the United Nations Sustainable Development Goals also encompasses information and communication technologies (FAO 2018). Agricultural production involves several duties such as tillage, planting, sowing, irrigation, fertilization, spraying, and harvesting. It is vital to monitor these acts. Unlike traditional methods, technical innovations, especially in the aviation sector and diverse sensors and remote sensing technologies, have enabled the implementation of innovative strategies in agriculture.

Plant monitoring studies provide decision-makers with fast access to essential information for agricultural applications. Decision makers in the field are significantly aided in assessing the quantity, timing, and geographical area of agricultural applications by the immediate availability of accurate information. Making appropriate decisions aids in the conservation of natural resources such as soil and water, while also reducing production costs.

In the 1980s, site-specific management, known as precision agriculture and linked to the utilization of contemporary technologies in land management, began to emerge. The basic purpose of precision agriculture is to optimize inputs such as fuel, fertilizer, and pesticides according to local plant and soil conditions (Lukas et al. 2016).

In precision agriculture, the paramount variables to consider are the velocity and precision of situational assessment, together with the implementation of suitable interventions and applications. It is well acknowledged that information-collection methods in conventional agricultural regions are time-consuming, expensive, and location-specific. At now, remote sensing technology is being advanced to facilitate data collection for researchers and cultivators in agricultural sectors. With technological advancements, sensors have diminished in size and cost, enabling their installation on unmanned aerial

vehicles (UAVs) that were formerly deployed on satellite platforms. This technical innovation enables the acquisition of data with enhanced temporal and geographical resolution. According to the collected data, UAVs have become extensively employed in agricultural regions for activities such as fertilization, spraying, and restricted irrigation, as well as for photography.

This study encompasses research on UAV uses in agriculture, fulfilling many roles both nationally and globally. The objective is to enhance the existing literature by meticulously analyzing the historical and modern uses of UAVs in agriculture.

### **UAVs and Sensors**

Recently, there has been significant interest in employing remote sensing technologies to collect data on the physiological condition and biophysical characteristics of plants. Various studies, encompassing plant water stress assessment, vitality evaluation, biomass calculation, and disease surveillance, can profit from the novel UAV and remote sensing technologies. Multispectral, hyperspectral, and thermal images have proven to be more advantageous due to their superior spectral compatibility, despite the extensive use of visible RGB images in various research investigations (Qin et al. 2013, Park and Lu 2015; Pádua et al. 2017). In plant monitoring studies, vegetation indices obtained from diverse ratios of many multispectral and hyperspectral camera bands provide very accurate data transmission (Sener et al. 2018).

Numerous studies have demonstrated the significant potential of hyperspectral sensors in identifying environmental stressors or plant diseases that affect the biophysical (Aasen et al., 2014; Gnyp et al., 2013) and biochemical (Li et al., 2010; Yu et al., 2013) characteristics of agricultural plants and vegetation. The primary challenge of employing hyperspectral cameras on UAVs was their excessive size and weight. Compact, lightweight cameras built for unmanned aerial vehicles (UAVs) have addressed this problem.

In drought conditions, plants cannot absorb sufficient water for transpiration, resulting in elevated temperatures of the leaves and crown. Thermal bands are utilized to monitor plant water stress, whereas RGB and infrared bands are mostly employed to evaluate plant development. Figure 1 illustrates a standard vegetation spectral reflectance curve.

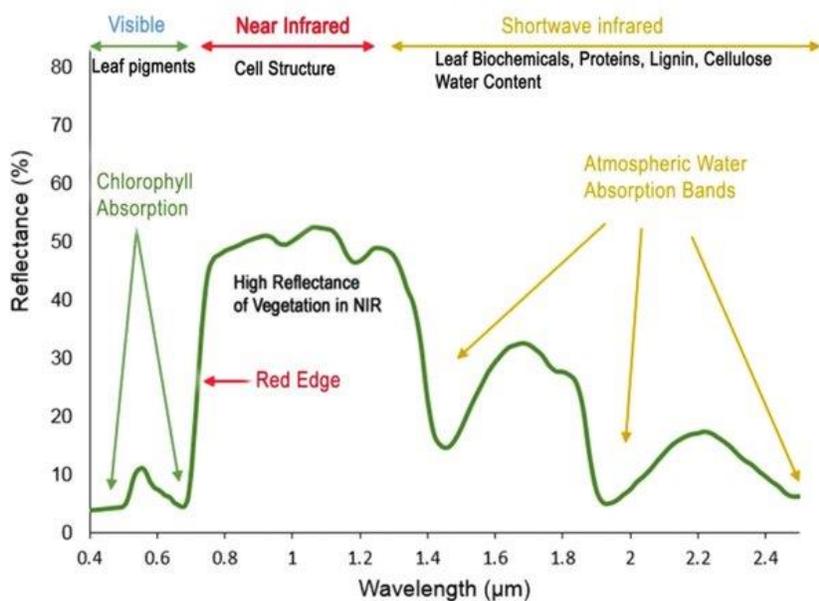


Figure 1. Spectral reflectance of vegetation (Roman and Ursu, 2016)

Four separate types of sensors are deployed in every UAV application for precision agriculture research. Table 1 presents data regarding the utility of these sensors in several application sectors (Maes and Steppe 2019).

Table 1. Analysis of Applications and Appropriateness of Various Sensors

Application		Type of sensor/camera			
		RGB	Multispectral (broad band)	Hyperspectral (Narrow band)	Thermal
Drought stress	Detection in early stages	--	--	S <sup>b</sup>	HS
	Long-term consequences	--	HS	HS	S
Pathogen detection	Detection in early stages	--	--	HS	HS
	Severity of infection	HS	HS	HS	S
Weed detection	Spectral discrimination	--	S	HS	--
	Object based	HS	HS	--	--
Nutrient detection		S	HS	HS	S
Growth vigor	Growth stage	HS	--	--	--
	Canopy height and biomass	HS	HS	--	--
	Lodging	HS	--	--	S
Yield prediction		S	HS	--	--

<sup>a</sup> Abbreviations: HS, Highly suited; S, Suited

<sup>b</sup> Calculation of sun-induced fluorescence from hyperspectral data

## **Agricultural practices**

Advancements in remote sensing technology enable UAVs to provide very accurate data across extensive areas, especially in plant monitoring and assessment studies. UAV-based remote sensing technologies provide significant advantages for enhancing agricultural productivity. Agricultural output can be enhanced by integrating sensor data with real-time information provided across the field parcel. As per FAO (2018), UAVs can provide valuable insights on various topics, such as soil health, plant diseases, fertilizer application, irrigation scheduling, yield estimation, and weather forecasts. This section will explore the applications of UAV-based remote sensing technology in agriculture, emphasizing yield estimation, biomass assessment, soil moisture and water stress analysis, as well as disease and insect management.

### **Crop Yield**

Crop yield is often determined using estimates from remote sensing or direct measurement. Field measuring strategies for providing sufficient inventory data across extensive areas are costly and need a substantial workforce. In this research, estimation strategies utilizing remote sensing markedly enhance efficiency. Remote sensing has significantly enhanced crop monitoring and production assessment.

Upon reviewing the UAV-based yield estimation research, it is evident that they focus on sorghum, barley (Roosjen et al. 2016), corn, rice, and wheat (Schirrmann et al. 2016).

Research employing RGB images of plant height and canopy cover by UAV (Chu et al. 2016), vegetation indices (Gracia-Romero et al. 2017), or multispectral images (Zhou et al. 2017) has achieved a reasonable degree of accuracy in yield estimation. In 2010, Swain et al. assessed rice yield and biomass utilizing a multispectral camera and an unmanned aerial vehicle (UAV).

Maes and Steppe (2019) indicate that yield estimation strategies have focused on plant growth models utilizing empirical regression models derived from UAVs. The GRAMI growth model was successfully employed for rice yield estimation using UAV-based data, as reported by Kim et al. (2017). Moreover, LIDAR technology provides high-resolution elevation data, which can be employed in yield estimates, as noted by Xu et al. (2020).

## **Biomass And Nutrient**

Monitoring plant biomass development yields critical information for agricultural management. Lukas et al. (2016) utilized UAV and LANDSAT satellite imagery to analyze various characteristics during the growth phase of winter wheat plants. Upon conclusion of the study, UAV-derived data exhibited superior precision relative to satellite data, indicating that wheat plants can be utilized for specialized plant management and variable-rate fertilizer applications in specific regions.



**Figure 2.** Biomass measurement studies in paddy with UAV

Utilizing RGB-based imagery to monitor cereal crops can provide comprehensive insights (Schirrmann et al. 2016; Du and Noguchi 2017). Dillen et al. (2016) reported that UAV-based RGB photography yielded satisfactory outcomes for orchards, whereas Watanabe et al. (2017) indicated that these cameras effectively estimated plant heights for cereals.

Zarco Tejada et al. (2012) assert that vegetation indices are commonly employed to assess plant water stress, health condition, metabolic processes, biomass, and yield. In the analysis of plant phenological development, various vegetation indices are utilized, with the NDVI being the most prevalent.

Deng et al. (2018) emphasized the importance of UAV multispectral remote sensing technology in precision agriculture by utilizing multispectral imagery from a multirotor UAV to compare NDVI and reNDVI vegetation indices with field-level SPAD measurements.

Pölönen et al. (2013) utilized a machine learning methodology to assess nitrogen levels and biomass in wheat and barley cultivation areas through the use of a UAV and a hyperspectral camera. The SMART model facilitated atmospheric calibration, whereas radiometric correction was employed for light source calibration.

The quantity of biomass and photosynthesis is generally regarded as an indicator of vitality (Goulet and Morlat 2011). Wine professionals can obtain essential information by generating vigor maps utilizing vegetation indicators like NDVI (Costa-Ferreira et al. 2007). Gnadinger and Schmidhalter (2017) employed remote sensing techniques to ascertain the relationship between plant population and diverse fertilizer applications in relation to plant indices. The study demonstrated the utility of remote sensing in plant development.

Lelong et al.'s 2008 work utilized UAVs to observe wheat plots with high-resolution image processing. The study examined the potential correlation between total nitrogen content and the NDVI, LAI, and GNDVI plant indices. Meng and Cheng (2020) employed UAV flights and an algorithm to improve the resolution of time-series satellite images of cornfields and used these images to assess soil nutrient content. Songyang et al. (2018) assessed the nitrogen content in the leaves of rice plants in China utilizing a UAV and a RapidSCAN CS-45. Gao et al. (2000) assert that the quantity of chlorophyll in aboveground plant biomass is substantial and significantly influences NDVI. In contrast, they asserted that EVI is more effective in connecting with biomass as it directly reflects structural changes in the plant canopy.

Field-based fertilization based on soil and plant N concentration is a crucial component of precision farming techniques. While fertilizing the entire area using the old way costs farm owners a lot of money, it can also pollute soil and water resources. With the use of multispectral and hyperspectral cameras, it is now possible to determine the N content in real time using the spectral reflectance value of plant leaves. According to Maresma et al. (2016) and Hunt et al. (2018), most UAV-conducted research concentrates on using multispectral displays to measure the amount of chlorophyll or nitrogen present. According to Franceschini et al. (2017), vegetation indices and the amount of chlorophyll in potatoes are highly correlated. Lucieer et al. (2014) used a hyperspectral camera-loaded UAV to collect 324 spectral band data between 361 and 961 nm to investigate the validity of the assessment of green biomass in barley and the chlorophyll content in pasture. According to Franceschini et al. (2017), vegetation indices and the amount of chlorophyll in potatoes are highly correlated. Lucieer et al. (2014) used a hyperspectral camera-loaded UAV to collect 324 spectral band data between 361 and 961 nm to investigate the

validity of the assessment of green biomass in barley and the chlorophyll content in pasture.

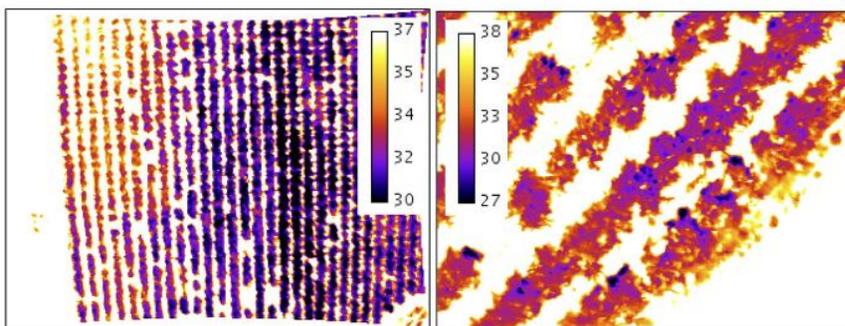
Thermal and infrared sensors, along with light detection and ranging (LIDAR) technologies, have been employed to examine several parameters, including biomass estimation, vegetation canopy height and temperature, and nitrogen and chlorophyll levels (Andujar et al., 2013; Maimaitijiang et al., 2017). Shendryk et al. (2020) utilized LIDAR data and multispectral imagery as inputs for variable nitrogen fertilization in Australian sugarcane fields. Research on this subject remains nascent; specifically, investigations into precision agriculture and variable-rate nitrogen applications are said to be inadequately developed (Maresma et al. 2016).

Sugar beet is mostly employed for sugar production, but it is also utilized for generating renewable bioenergy. Advancements in remote sensing, particularly in LIDAR (Light Detection and Ranging) technology, have facilitated the measurement of the biomass of aboveground vegetation. Xu et al. (2020) employed machine learning to analyze UAV and LIDAR data in China for the estimation of sugar beet output. Among the six methodologies utilized to estimate sugar beet production, the study revealed that Random Forest Regression (RFR) exhibited the highest correlation ( $R^2 = 0.96$ ).

Honkavaara et al. (2012) employed point clouds and hyperspectral reflectance data to develop a novel biomass estimation technique supported by a vector regression machine for UAV-based precision agriculture research. This study aimed to enhance the accuracy of the estimating process through the use of digital elevation modeling, high-resolution imagery, and advanced image analysis. This latest technology was employed to develop a resilient, economical, and locally reproducible remote sensing application.

### **Soil Moisture and Water Stress**

Stomata close and transpiration diminishes because to the temperature increase induced by dryness. UAV-based thermal imagery has been employed in many studies to assess water stress in different plant species for the purpose of monitoring drought effects and irrigation scheduling research.



**Figure 3.** An example of water stress study in orchards using UAV (Zarco-Tejada et al., 2018).

Romero-Trigueros et al. (2017) conducted experiments on citrus water and soil salinity stress using UAVs and multispectral camera pictures. They claimed that the combination of field measurements and remote sensing data produced significant findings.

Gago et al. (2015) assert that the superior spatial and temporal resolution of UAVs significantly enhances water stress management in agriculture. Their review analysis indicates a robust correlation between water stress indicators, such as stomatal conductance (gs) and water potential, and NDVI, TCARI/OSAVI, and PRInorm indicators collected from UAV imagery. They assert that thermal imaging is a remote sensing method commonly utilized as a thermal indication for assessing leaf water stress. Zaman-Allah et al. (2015) endeavored to correlate plant growth parameters with the NDVI plant index they established utilizing data from a multispectral camera. They asserted that the study's remote sensing methodology could detect the different stresses affecting plants.

Diverse growth methodologies, such the leaf area index (LAI) and the normalized difference vegetation index (NDVI), produce valuable insights into the effects of water stress. Remote sensing methodologies can detect water stress in barley genomes (Hoffmann et al. 2016).

Hafizoğlu (2020) utilized drones to amalgamate plant physiology and biochemical research areas and evaluate tolerances to examine the alterations in plants induced by drought. The activity of superoxide dismutase and the relative water content in maize plants exhibited a positive correlation with NDVI data, whereas protein and malondialdehyde levels demonstrated a negative correlation.

Xiangyu et al. (2019) employed images captured by a UAV equipped with a hyperspectral sensor and a machine learning algorithm to assess elevated soil

moisture content. Rapid advancements in remote sensing technologies have led to the emergence of thermal and hyperspectral cameras, alongside radar technology, for evaluating soil moisture content (Sener et al. 2019).

Water use is intimately associated with the temperature of the foliage and canopy as assessed by remote sensing methodologies. During water stress, plants exhibit reduced transpiration, closure of stomata, and an increase in temperature. In this connection, Bellvert et al. (2016) utilized thermal cameras to optimize irrigation scheduling, whereas Zarco-Tejada et al. (2008) investigated water content and the assessment of plant water stress. Thermal imaging is optimal for detecting drought stress (Maes and Steppe 2012).

Zarco-Tejada et al. (2012) investigated water levels in lemon trees with PRI data and regulated deficit irrigation methods. They utilized thermal and hyperspectral sensors to detect water stress by analyzing canopy temperatures, brightness, and reflectance spectra.

Numerous attributes inaccessible in RGB images can be acquired with thermal photography. Direct measurement of plant surface temperatures during stress tests may yield erroneous results. Normalized indicators for surface temperatures, such as the Crop Water Stress Index (CWSI), are utilized to prevent this misinterpretation. A 2019 study by Bhandari et al. examined the efficacy of multispectral and hyperspectral cameras in assessing nitrogen and water stress, employing the Water Band Index (WBI) alongside vegetation indices for citrus.

Galioto (2017) stated that drip and pivot irrigation technologies were essential for field agricultural research, despite UAV-supported thermal camera studies yielding significant results in stress assessments in orchards. Furthermore, a negative association exists between soil temperature and moisture content. Soil temperature increases as soil moisture content decreases. Konstantin et al. (2018) and Fengshuai and Arj (2020) assert a robust association between soil moisture content measurements obtained from heat and radar sensors on UAVs and the actual soil moisture levels determined by the gravimetric method. Irrigated plots inside irrigation zones are recognized with thermal cameras (Sener et al. 2019).

### **Disease And Pest Management**

Diseases and insects can swiftly spread throughout a sizable agricultural area, and their harm can significantly impact the local and national economies. Rapid interventions in wide areas are necessary to eliminate such issues, reduce damages, and ensure food safety. In this regard, UAVs and imaging systems are

seen as helpful resources, particularly when offering comprehensive details regarding the beginning of illnesses and the regions they impact.

Handheld compressed air sprayers and battery-operated shoulder-mounted sprayers are the primary instruments employed in conventional agriculture for the application of pesticides to crops. The World Health Organization has recorded over one million cases of pesticide-related adverse events during manual spraying, suggesting that the use of such sprayers can significantly harm the environment and human health (Toscano et al., 2024). Unmanned Aerial Vehicles (UAVs), capable of various functionalities, have been employed to address pesticide exposure issues for agricultural equipment and personnel, as opposed to conventional spraying methods (Babaoğlu et al. 2021).



**Figure 4.** DJI Agras T50 (Team, n.d.)

The International Association of Unmanned Aerial Systems forecasts that 80% of all UAV applications will pertain to agriculture (Radoglou-Grammatikis et al., 2020). Currently, numerous firms have commercially created a wide variety of spraying UAVs, alongside specially customized spraying UAVs. The market clearly indicates that the DJI Agras models are the most favored rotor systems. Over the years, DJI Agras has developed several models with differing capacity, including the T16, T20, T30, T40, and T50. Each model has been designed with a distinct array of technological attributes and functionalities (Team, n.d.).

Large tanks are currently employed for spraying plants, whereas formerly, small tanks were conveyed by helicopter. The initial step in effectively utilizing such research is to employ picture capturing and analysis to ascertain the pesticide concentration in the treated areas. This will avert economic damage and chemical contamination of the entire region (Wen et al. 2019).

Garcia-Ruiz et al. (2013) utilized UAVs to examine citrus fruit disease and health. They conducted research to identify *Verticillium* wilt, which impedes olive tree growth. The study included thermal and multispectral camera images for early illness detection (Calderón et al. 2013). It was found that eliminating erroneous observations in multispectral imaging allows hyperspectral imaging to detect early-stage disease more effectively. Nonetheless, it is apparent that research employing UAVs to detect fungal diseases via hyperspectral cameras has not been sufficiently conducted (Mahlein, 2016).

Problematic areas were found using NDVI imagery in the study that employed multispectral cameras to detect regions demonstrating stunted plant growth. The flawed regions were subsequently sprayed to address the production problem (Mogili and Deepak, 2018). Uzun et al. (2018) indicate that farmers effectively managed diseases, pests, and weeds with the use of agricultural UAVs, resulting in enhanced success.

Agnihotri (2018) examined the efficacy of thermal and multispectral imagery, supported by machine learning, for pest detection in rice fields. The identification of pathogen-related disorders has been the focus of extensive research. It was said that further study is underway about the application of hyperspectral cameras for disease detection, utilizing wavelengths ranging from 380 to 1020 nm (Thomas et al. 2018; Mahlein et al. 2018; Abdulridha et al. 2020).

Akkamış and Çalışkan (2020) assert that pesticide applications conducted by inadequately trained personnel utilizing agricultural UAVs could provide detrimental impacts. Determining the appropriate pesticide dosage for a certain condition, modifying flight altitude and velocity, utilizing suitable nozzles, and ensuring application by entities with the requisite legal permissions are all essential for the user.

## **Discussion**

UAVs are growing in popularity as inexpensive equipment for monitoring plants and land for in-depth study by management and decision-makers in agricultural sectors. Because RGB, multispectral, thermal, and hyperspectral cameras offer data at varying spatial and spectral resolutions, they are favored for various uses, sizes, and specifications.

Even though remote sensing and UAV technologies appear to be very helpful tools, it is only feasible to comprehend this data by comparing it with data from the ground. One of the most crucial factors in gathering and analyzing data is the researcher's proficiency with the kind of plant and issue under observation. It is crucial to seek assistance from specialists with knowledge of plant, disease, or stress issues to properly examine the data in studies carried out by researchers who lack infrastructure related to the topic of the study and only possess software and hardware expertise.

It has been observed that the reduction in the cost of UAVs and the equipment they require has a favorable impact on the adoption of UAV and remote sensing technologies in agriculture and helps distribute them to end users. It is a truth, nevertheless, that image processing software is not widely utilized outside of the classroom, and memberships are still below what businesses in the modern world would like to have. Since prices are now tied to the nation's economy and the needs of rural areas, it is evident that the only way to promote commercial use is to re-adjust them.

Making these technologies more user-friendly is advantageous for improving the use of UAV and agricultural applications. The contemporary UAV business is still moving quickly in the direction of more practical, user-friendly agricultural vehicles. These studies' primary goal is to assist users in increasing agricultural productivity and cutting expenses.

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## **Chapter 12**

### **The Use of Artificial Intelligence in Watershed Management**

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## **Abstract**

The integration of Artificial Intelligence into watershed management initiates a groundbreaking shift in addressing challenges tied to water and land resources. By leveraging tools like machine learning, geospatial analysis, and real-time monitoring systems, AI facilitates highly accurate hydrological modeling, efficient sediment management, and proactive water quality assessments. These advancements equip decision-makers with the ability to create adaptive strategies for resource optimization and disaster preparedness. However, fully harnessing AI's potential requires overcoming barriers such as data limitations, implementation expenses, and the demand for specialized expertise. This chapter highlights innovative AI applications in watershed governance, focusing on its role in fostering sustainability, mitigating climate-related risks, and promoting inclusive management practices.

**Keywords:** Artificial Intelligence, Watershed Management, Sustainability.

## Havza Yönetiminde Yapay Zeka Kullanımı

### Özet

Yapay Zeka'nın havza yönetimine entegre edilmesi, su ve arazi kaynakları ile ilişkili sorunların çözümünde önemli bir değişimin önünü açmaktadır. Makine öğrenimi, coğrafi analiz ve gerçek zamanlı izleme sistemleri gibi araçlardan yararlanan YZ, son derece hassas hidrolojik modelleme, verimli sediment yönetimi ve proaktif su kalitesi değerlendirmeleri sağlamaktadır. Bu gelişmeler, karar verici noktasındaki yöneticilerin kaynak optimizasyonu ve afet hazırlığı için uyarlanabilir stratejiler oluşturma yeteneğini geliştirmektedir. Ancak, YZ'nin potansiyelini tam anlamıyla kullanmak, veri sınırlamaları, uygulama maliyetleri ve uzmanlık talebi gibi engellerin aşılmasını gerektirmektedir. Bu çalışma, yapay Zeka'nın havza yönetimindeki yenilikçi uygulamalarını ele alarak, sürdürülebilirliğin teşvik edilmesi, iklimle ilgili risklerin azaltılması ve kapsayıcı yönetim uygulamalarının desteklenmesi konularındaki önemine değinmektedir.

**Anahtar Kelimeler:** Yapay Zeka, Havza Yönetimi, Sürdürülebilirlik.

## 1. Introduction

Artificial Intelligence integrated into natural resource management opens a new era in terms of the solution of intractable environmental problems. That is to say, AI technologies provide unparalleled opportunities in watershed management that can be applied to improving the sustainability and resilience of water and land resources. Traditional watershed management approaches—which are largely based on historical records and human judgment—have an increasingly hard time keeping up with the dynamics due to climate change, urbanization, and population growth. With the ability of AI in handling extensive data and extracting patterns, hence, it will be possible to go for proactive, adaptive, evidence-based decision making (Afan et al., 2016; Causevic et al., 2024; Jing et al., 2023; Michael Ayorinde et al., 2024).

Application areas of AI in watershed planning span many domains: from essential ones, such as analyzing meteorological and hydrological data in view of flood risk management and events of flooding, which can be predicted with high accuracy to support early warning and quick response in disaster management (Afan et al., 2016; Kamyab et al., 2023; Zhang et al., 2024). Machine learning algorithms will thus work in managing the water quality through real-time data that sensors and remote-sensing technologies provide, through observing the sources of contamination or foretelling the water-quality trends for timely intervention aimed at safeguarding ecosystems and public health (Kamyab et al., 2023; Michael Ayorinde et al., 2024; Zhang et al., 2024). AI also innovates land-use planning by synthesizing complex data from satellite imagery and GIS for optimal land allocations for agriculture, urban development, and conservation in a balanced ecology and economy (Afan et al., 2016; Buchelt et al., 2024; Imada, 2014).

The role of AI in watershed management encompasses more than just reactive measures; it fosters integrated and predictive planning. AI, in turn, will allow decision-makers to consider various scenarios and long-term impacts by integrating digital twins and simulation models that enable strategies balancing environmental, economic, and social objectives (Causevic et al., 2024; Jing et al., 2023; Zhang et al., 2024). Also, emerging AI will help fill data gaps, especially in remote and under-monitored areas, with IoT-enabled sensors and autonomous data-gathering technologies (Buchelt et al., 2024; Kamyab et al., 2023; Michael Ayorinde et al., 2024). These technologies have been effective in real-time monitoring of water quality, sediment transport, and even the impacts of extreme weather events (Afan et al., 2016; Kamyab et al., 2023).

Challenges like water deficiency, degradation of ecosystems, and impacts due to climate change will find comprehensive solutions with the adoption of

AI-powered systems in every nook and corner of the world. This integration can be illustrated, for instance, by carbon sequestration modeling through the prevention of deforestation in the next generation of monitoring of forest and watershed ecosystems by incorporating AI and EO technologies, among others (Causevic et al., 2024; Jing et al., 2023). These systems also contribute to the realization of SDGs on universal access to clean water, biodiversity conservation, and enhanced climate resilience (Causevic et al., 2024; Kamyab et al., 2023).

The chapter outlines some recent global developments in AI-powered watershed management and demonstrates recent innovation, extant challenges, and future directions. The purpose is to indicate how AI is not just a technique for finding solutions to short-term problems but actually forms the base for creating sustainable, resilient, and intelligent systems that deal with growing pressures on land and water across the globe.

## **2. Artificial Intelligence in Watershed Management**

Artificial Intelligence is changing the face of watershed management studies by efficiently tackling the complex and dynamic nature of these systems. Advanced techniques, such as machine learning, deep learning, and big data analytics, can analyze large datasets for patterns that would be too subtle for traditional methods to detect or would take too much time to process. Some model examples include the Random Forest and Artificial Neural Networks, which demonstrated high-accuracy prediction performances regarding the identification of critical hydrological events, such as floods, sediment movements, or quality variations of water (Hasan et al., 2024; Hitouri et al., 2024; Khankhoje & Choudhury, 2023). These AI-driven approaches are generally effective in solving the challenges posed by shifting climate patterns, fast urbanization, and multiplying stress on water resources, thus creating novelty for water management that is sustainable.

Applications of AI in watershed management are among the strong tools for natural disaster prediction and mitigation. Advanced machine learning algorithms, together with remote sensing data such as Synthetic Aperture Radar, enable precise mapping of flood-prone areas, hence providing timely warnings to reduce the impacts of flooding on communities and ecosystems (Debnath et al., 2024; Hitouri et al., 2024). Similarly, AI-based systems enhance preparedness against drought by forecasting water availability using a combination of remote-sensing and on-site hydrological data (Hasan et al., 2024; Michael Ayorinde et al., 2024).

AI also important in monitoring water quality and managing pollution in watersheds. Advanced systems powered by AI can trace pollution sources using techniques such as spectral fingerprint analysis, enabling the rapid identification of contaminants and their mitigation. These IoT sensors form part of a package coupled to AI systems for the continuous or real-time monitoring of dissolved oxygen, turbidity, or heavy metal concentration, with a focus on aquatic ecosystem conservation and preserving water supplies (Duan et al., 2024; Michael Ayorinde et al., 2024). Further yet, AI contributes to an optimum range of land use at any given watershed by examining satellite images/globally referenced imagery or other global information systems to aid decisions made on agriculture, urban structure, or nature conservational processes. This approach mitigates soil erosion, regulates sediment flow, and enhances the water retention capacity of the catchment area (Khankhoje & Choudhury, 2023; Vekaria & Sinha, 2024).

One of the important aspects of watershed management encompasses solving several interconnected problems related to floods, droughts, pollution, and degradation of land. AI provides integrated solutions for these problems by modeling complex interactions within hydrological systems. For instance, AI-driven sediment discharge prediction models help in the management of reservoir sedimentation and riverbank erosion to ensure stability in hydraulic structures and sustainability in river ecosystems (Hasan et al., 2024; Khankhoje & Choudhury, 2023).

### **3. Applications of AI in Watershed Management**

#### **3.1. Water Resource Management**

AI is changing the way water resources are managed, solving streamflow prediction problems, monitoring water quality, handling flooding, and optimizing the use of available resources. Most conventional hydrologic models are bound to be limited in their ability to capture complex watershed dynamics; therefore, AI-driven approaches have emerged either as an alternative or as a complement. These techniques include ML and DL algorithms, which provide high-precision forecast outputs with valuable insights; thus, helping in bringing more resiliency and adaptation to the changes in environment dynamics in the management of this resource (Chang et al., 2023; Krishnan et al., 2022).

The AI would thus be used under the highly impactful areas concerning stream flow prediction and flood management. Models such as the Long Short-Term Memory network and Adaptive Neuro-Fuzzy Inference Systems have been rendered quite capable of handling the most difficult rainfall-runoff relationships associated with hydrological cycle management. For instance,

applications of ANFIS in the Tigris Basin for the modeling of rainfall-runoff processes were very effective and outperformed conventional methods, thus giving better accuracy to flood forecasting systems (Gerger et al., 2021). Apart from this, machine learning algorithms using Random Forest and Support Vector Machines have been integrated into the use of remote sensing, which has included Synthetic Aperture Radar (SAR) and was used in the delimitation of flood areas. As mentioned above, research conducted within areas such as Morocco or India confirmed that SAR and AI-based models perform well in the high classification of flood susceptibility for the betterment of urban planning and reducing natural disasters (Debnath et al., 2024; Hitouri et al., 2024).

However, AI's role has extended beyond flood management using predictions. AI-based systems allow the integration of diverse data sources such as climate projections, satellite imagery, and IoT sensors into real-time monitoring and early warning mechanisms. With this kind of system, scenario simulations will be made possible, where decision-makers can apply proactive measures to flood control and the distribution of resources accordingly (Hmoud Al-Adhaileh & Waselallah Alsaade, 2021; Krishnan et al., 2022). AI analytics further supports the ability to simulate flood patterns over the longer term—a pre-requisite when designing appropriate infrastructure and flood management policies adaptable to a dynamically changing physical environment (Çubukçu et al., 2022).

Among many, the water quality monitoring domain also received immense benefit because of recent improvements in AI. For analysis of some important parameters such as turbidity, dissolved oxygen, and heavy metals concentrations in water, ANN, as well as the models based on the concept of Federated Learning, have been implemented in respective variants. The ANN and CNN-LSTM models have been applied for water quality prediction in reservoirs and river systems, for instance, and show strong solutions in the identification of contamination sources and optimization of water treatment processes (Hmoud Al-Adhaileh & Waselallah Alsaade, 2021; Jenny et al., 2020). Such systems not only give insight into the real-time status of water quality but also inform policy decisions to reduce pollution and improve resource sustainability.

Effective distribution of the water resource is another pressing challenge that AI serves by providing multi-objective optimization algorithms. This will give a balance by taking socio-economic and environmental information concerning agricultural, industrial, and domestic use while ensuring that resources are managed in a way to prevent further degradation of this basic life resource. Such an AI-based DSS, such as the aiWATER model, has proven useful for

large-scale practical applications in semi-arid and arid areas regarding effective water distribution and optimal planning when the resource base becomes too weak (Çubukçu et al., 2022; Krishnan et al., 2022).

Besides allocation, AI is changing the way water infrastructure maintenance is carried out. Predictive maintenance models identify potential failures in a system by analyzing historical performance data to reduce downtime and maintenance costs while improving the reliability of water supply networks. These applications also go to agriculture, whereby through AI-based irrigation optimization, water wastage has been reduced by a great margin while improving crop yields to support sustainable farming practices (Çubukçu et al., 2022; Krishnan et al., 2022).

Where climatic change is exacerbating this shortage of water, AI makes it possible to provide more adaptive and robust management capabilities that include variable rainfall patterns and long-term droughts under rising water scarcity. Solutions that may work in concert with these disparate uncertainties will be the only answers. Real-time monitoring and predictive analytics are revolutionizing water resource management, providing water managers the necessary tools to meet their extreme-weather challenges (Çubukçu et al., 2022; Debnath et al., 2024). These systems go far beyond flood and drought forecasting. They allow long-run simulations in order to reveal how the climate change may affect their reservoirs and aquifers to meet the water supply on a sustainable basis in future years (Gerger et al., 2021; Hitouri et al., 2024). By integrating variables into one logical framework, the models show the way on how the water resources system could be managed in environments that become increasingly unpredictable in space and time. Soil moisture, groundwater reserves, and recent climate trend variabilities thus become major input parameters (Krishnan et al., 2022).

### **3.2. Land Use Planning**

Innovations in technology are literally changing the face of land-use planning; new tools permit a more profound analysis of spatial data at an unprecedented scale of detail. Integration with advanced computational methods now makes remote sensing capable of easily identifying erosion-prone areas and simulating changes in land use. Further, integrating such remotely sensed data with machine learning models, such as Convolutional Neural Networks and Random Forests, allows one to extract insight into how land cover changes through time. Satellite imagery from multispectral satellites analyzed by these methods has been able to locate areas of rapid urbanization or deforestation (Huang et al., 2022; Ruiz et al., 2023; Ediş & Ulaş, 2017).

The outcomes will serve for applications within the district of Adama in Ethiopia, both in the identification of land-use patterns and debris flow risks. Thus, this approach has allowed better management of local authorities to get more adequate allocations of their resources and reduce environmental hazards (Bojer et al., 2023). These examples illustrate well that inclusion of advanced tools in planning opens possibilities for data-driven decision-making toward better management of the environment and, not least, sustainability.

Soil erosion likely is the most critical watershed management component, wherein the control of effects is supported through AI-enhanced geospatial analyses. Classic models, such as the Revised Universal Soil Loss Equation, have enhanced their output upon the incorporation of remote-sensing data and machine-learning methods, hence raising their credibility two steps ahead. Starting from basic analyses of soil texture and topography, AI assesses a lot of contributing elements derived from high-resolution remote-sensing data to compute detailed maps of erosion susceptibility. These maps offer insight into the identification of areas with high risks and help in implementing targeted soil conservation efforts. For example, steep mountainous regions identified with the highest erosion risks form the basis for developing site-specific erosion control measures (Samarinas et al., 2024).

Similarly, AI is bringing about meaningful changes in the simulation of land-use scenarios and predictions regarding the future. Models like Cellular Automata (CA) and hybrid systems combining CA with System Dynamics (SD) and Backpropagation Neural Networks (BPNN) allow for the modeling of land-use and land-cover (LULC) changes under various climate and socio-economic scenarios (Huang et al., 2022). These simulations enable the assessment of potential trade-offs between urban expansion, agricultural development, and conservation efforts. For example, AI models projected a 37% increase throughout Zhejiang Province in bamboo forests under environmentally focused scenarios, alongside significant declines in coniferous forest areas within the region (Huang et al., 2022).

The performance of AI sounds promising for land-use planning, as applications continue to evolve across a great variety of problems. Multi-objective optimization models evaluate the tradeoffs among competing priorities such as economic growth, environmental sustainability, and social equity. This model enables decision-makers to make adaptive land-use strategies, considering diversified needs of several stakeholders. AI-driven frameworks have already allowed the complete integration of agricultural productivity goals with biodiversity conservation, reducing conflicts between land use types (Ruiz et al., 2023; Zhang et al., 2024). Such models integrate

real-time data streams in order to allow dynamic changes in the plans while the conditions are changing.

AI applications in land-use planning are important in tackling global issues such as urban sprawl, deforestation, and desertification. AI in urban growth modeling tools helps in simulating the expansion of cities by identifying where agricultural or natural lands may be under threat. These are more relevant and valuable in regions where urbanization is happening or will happen quite rapidly and help guide infrastructure planning in a manner that can reduce impacts on the environment. Other forest restorations have also used AI in predicting the outcome of their reforestation projects by basing such projections on land suitability and historic vegetation patterns (Eker et al., 2023; Samarinas et al., 2024).

Taming raw integration of AI-driven simulation models and spaceborne/sensor technologies with diverse, contextually relevant optimization algorithms helps frame land-use planning at an increasingly data-rich level to address emerging complexities wrought either from modern environmental or various socio-economic challenges. This simultaneously offers better information on land use to make decisions that assure land-use planning is appropriately coordinated toward sustainable development, therefore adequately weighing immediate development needs alongside the conservation methods necessary for long-term environmental concerns.

### **3.3. Ecosystem Management and Biodiversity Conservation**

With modern perspectives, artificial intelligence has developed as an important tool within ecosystem management in biodiversity conservation because it offers additional value across advanced solution paths for ecological system monitoring, analysis, and protection. Traditional methodologies of biodiversity conservation normally see scalability issues, problems emanating from data, or higher complexities in environmental challenges that keep growing every other day. Such approaches by AI are implemented with machine learning and remote sensing in place so it leads to timely intervention with a lot of precision (Lyu et al., 2022; Ullah et al., 2024).

Some of the most promising uses of AI in the care for biodiversity include monitoring. Using image and signal processing from drones and satellites, AI algorithms monitor populations of species and changes in their habitat. Example applications range from the classification of forest types to mapping species distribution changes because of deforestation, climate change, or illegal poaching. This enables researchers to monitor biodiversity on larger scales, even in remote areas that would have otherwise been quite costly in time and resources by using the traditional method of field surveys. In addition, AI

systems optimize scarce conservation resources by better selecting the prioritization areas that produce the maximum ecological benefits (Liu et al., 2021; Silvestro et al., 2022).

Besides monitoring species, AI holds significant promise for assessing the services provided by ecosystems, such as carbon sequestration, water filtration, and nutrient cycling. Advanced models simulate the interactions of several ecosystem services to yield information on trade-offs and synergies under alternative land-use or climate scenarios. The resulting simulations support decision-making in developing biodiversity-friendly management strategies that balance conservation against socioeconomic concerns, thus ensuring resilient and productive ecosystems (Saleh et al., 2024; Lyu et al., 2022).

AI also enhances the capacity to foresee and possibly temper the impact of climate change on ecosystems. AI-powered habitat suitability models utilize real-time data from IoT sensors, climate records, and satellite imagery to predict changes in the distribution of species and ecosystems. Such models support designing migration corridors and adaptive management strategies that maintain habitat connectivity and prevent species displacement. Predictive models have mapped out future species migrations, enabling the establishment of protected areas and proactive adaptation to climate change (Ullah et al., 2024).

Also, AI operates upon indigenous ecological knowledge side by side with advanced technologies in cooperative and inclusive forms of conservation. Participatory mapping systems engage local communities within key habitat identification and its monitoring to ensure that the greater thrust of conservation policies, actions, and decisions incorporate the best available science regarding conservation effectiveness and are attuned to both cultural sensitivities and social inclusivities. Community-led programs innovate adaptation uses of AI concerning assessment of the health of their forests, counting the quantity of wildlife, predicting risk for poaching among others; to demonstrate how well it had been serving innovation for bottom-up conservation (Shivaprakash et al., 2022; Silvestro et al., 2022).

#### **4. AI Applications in Watershed Management**

Artificial Intelligence is the new beginning that opens up new horizons in watershed management, both in predictive modeling and real-time monitoring for evidence-based decisions. Advanced resource utilization with the help of AI leads to better disaster management and conservation of ecosystems by use of much-improved technologies like machine learning and neural networks, geospatial analysis, and so on, integrated with the Internet of Things.

AI-driven decision support systems are the backbone for modern water management. These systems allow for the deployment of machine learning models combined with IoT sensors and remote-sensing data to analyze complicated hydrological processes important in event predictions of flooding or drought. For example, advanced techniques of rainfall-runoff modeling have achieved extraordinary accuracy that allows for early warnings against the impact of flooding if it were done timely (Himeur et al., 2023; Shekar et al., 2023). In various case studies, such as the Dicle Basin, the application of AI techniques like ANFIS showed better results in the prediction of the rainfall-runoff relationship than traditional models, demonstrating superior performance in handling nonlinear hydrological complexities (Gerger et al., 2021). These scientific advances have been widely used in managing water distribution and in protecting vulnerable groups.

One of the major contributions of AI has been within flood susceptibility mapping. The integration of Synthetic Aperture Radar data with machine learning models such as Random Forest and Extreme Gradient Boosting has shown promise in enhancing the precision of flood risk assessments. By using such models, very high precision in identifying flood-prone zones was realized within the Metlili watershed of Morocco, supporting disaster preparedness and land-use planning (Hitouri et al., 2024). AI-driven simulations can also enable stakeholders to project the potential effects of different interventions under alternative scenarios, thus establishing a sound scientific basis for adaptive management.

Another successful application of AI is in water-quality monitoring. IoT-enabled devices with ML algorithms perform real-time analysis of turbidity, pH, and heavy metals as certain key parameters of water quality. Artificial Intelligence systems optimize treatment processes by analyzing real-time data in wastewater management to better resource efficiency and environmental regulation compliance (Michael Ayorinde et al., 2024; Zhang et al., 2024). Similar systems like smart water management, integrating AI and IoT, have been used for leak detection and infrastructure monitoring to reduce losses in urban areas (Michael Ayorinde et al., 2024; Pandey et al., 2023).

AI models also significantly enhanced the capability of forecasting sediment flow and erosion risk, important for hydraulic structure maintenance, including dams and reservoirs. Application of dynamic ANN has enabled researchers to model sediment discharge with higher accuracy for sustainable water resource management (Hitouri et al., 2024; Khankhoje & Choudhury, 2023). Improved geospatial analysis, along with models such as the Revised Universal Soil Loss Equation, has offered even more detailed forecasting of the risks of soil erosion

and hence targeting interventions. For example, in Greece, studies are conducted using AI to analyze the soil texture, climate data, and topography for developing high-resolution erosion maps that help in effective regional planning (Chowdhuri et al., 2020; Samarinas et al., 2024).

### **5. Challenges in AI Applications for Watershed Management**

Artificial Intelligence integration into watershed management faces significant challenges on technical, economic, regulatory, and human resources fronts that limit the transformations it promises. Among the technical issues at the forefront is the considerable lack of reliable and good-quality datasets. AI models thrive on comprehensive, accurate, region-specific data, which are precisely lacking in many regions—mostly developing countries—due to a lack of adequate infrastructure related to data collection (Causevic et al., 2024; Çubukçu et al., 2022). Furthermore, the heterogeneity of data itself is contributed by incoherence in format, source, and time scales, making integration and processing very challenging. Inhibited adaptive decision-making due to a lack of real-time data on account of outdated systems and limited computational infrastructure further limits scalability and the widespread adoption of AI solutions (Himeur et al., 2023; Pandey et al., 2023).

It is further exacerbated by economic and regulatory barriers. AI technologies require huge upfront investment in infrastructure, sensors, and computational resources. For instance, the installation of IoT devices and cloud capabilities in most watersheds remains unaffordable. Besides, there are operational costs like maintenance and personnel training (Shivaprakash et al., 2022). Most often, regulatory frameworks are not at par with technological advancement, so guidelines related to data sharing, AI deployment, and ethical concerns are usually not well defined. Overlapping mandates in fragmented governance arrangements often make the elaboration and implementation of coherent AI policies complex, create uncertainty amongst stakeholders, and hamper further progress (Himeur et al., 2023).

Some AI obstacles to integration in watershed management are human resource limitations: namely, a multidisciplinary workforce skilled in both areas involving AI technologies and hydrology—skills that are badly lacking. Professionals in these regions, especially in countries designated as developing, lack competencies in applying AI apparatus effectively. The collaborative nature of AI applications further calls for effective communication and coordination among the technical experts, policymakers, and local communities. Such an approach to addressing such gaps, besides requiring high technical

expertise, calls for leadership plus stakeholder engagement, an element usually lacking (Causevic et al., 2024; Vekaria & Sinha, 2024).

The multifaceted approach will be needed to overcome these challenges. Advanced data collection systems, improvement in computational infrastructure, and the adoption of standardized data formats will significantly enhance the availability and usability of quality data. In addition, it is required that regulatory policies be clearly stated and forward-looking; ethics, transparency, and collaboration across regions should be guiding principles while assuring nondiscriminatory and efficient enforcement of AI technologies. Collaborative research can bridge the technical and institutional gaps to make AI tools more practical and accessible. Integration of AI training in education on environmental sciences will reduce this skill gap, but partnerships among universities, industry, and governments will adequately prepare a new generation of professionals to apply AI to smarter, more sustainable watershed management (Causevic et al., 2024; Himeur et al., 2023; Vekaria & Sinha, 2024).

## **6. The Future of AI in Watershed Management**

Artificial Intelligence is going to bring a new face to watershed management by integrating newer, advanced technologies such as drones, the Internet of Things, and real-time sensor networks. With these tools, AI has huge potential to optimize water resources, enhance ecological resilience, and respond to key challenges such as climate change and growing populations (Afan et al., 2016; Holzinger et al., 2024).

These unmanned aerial vehicles, installed with AI-powered imaging and analysis tools, can map broad watersheds and changes in land use; this quickly gives indications of erosion-prone and flood areas. By integrating drones, equipped with machine learning algorithms, one is able to conduct high-performance estimations of sedimentation models and disruptions in water flows vital to preserve both quality and water supplies (Afan et al., 2016; Holzinger et al., 2024).

IoT-enabled networks enable watershed monitoring in real time via data collection, analysis, and transmission with respect to water flow, water quality, and rainfall. When integrated with AI analytics, such systems enhance decision-making for immediate action on ecological hazards such as flooding or droughts. In fact, this integration has been exemplified to be more efficient in most water management strategies adopted by agriculture and urban uses (Krishnan et al., 2022).

Furthermore, AI-driven predictive modeling based on real-time sensor data opens new horizons for proactive resource management. These models can predict water demands, detect sources of pollution, and simulate the impacts of alternative management scenarios. For instance, predictive models have estimated how activities in an upstream catchment area may affect ecosystems downstream to support sustainable development decisions (Causevic et al., 2024; Eker et al., 2023).

While much has been achieved, some frontiers in AI applications for watershed management remain unexplored. Novel approaches include completely new methods, like digital twins—virtual models of physical environments—that promise to revolutionize planning by modeling long-term impacts of various interventions. Another promising avenue is the adoption of edge computing, processing data locally at IoT nodes, thereby enhancing the reliability and efficiency of AI insights (Holzinger et al., 2024; Liu et al., 2018).

The initial investment for implementing these technologies is already very high, which in the future will be negligible compared to the benefits such resources accrue in economic, ecological, and social perspectives. AI has unparalleled potential to boost efficiency, reduce environmental risks, and bring communities to more active involvement in water management. AI simplifies access to complex hydrological data and empowers policy decision-makers and local stakeholders to drive informed, equitable decisions toward sustainable and inclusive water governance (Afan et al., 2016; Krishnan et al., 2022).

Future efforts should overcome scalability challenges to realize its full potential. This will involve targeted investment in infrastructure, development of ethical regulatory frameworks, and capacity-building programs for training professionals in the effective integration of AI into water resource management. It will require collaboration between governments, academia, and private industries to drive innovation, ensuring both environmental sustainability and social equity (Eker et al., 2023; Holzinger et al., 2024; Liu et al., 2018).

## **7. Conclusion and Recommendations**

Artificial Intelligence is being integrated into watershed management to mark a new epoch in the solution of complex problems related to water resources, ecosystems, and climate change. The current applications involving AI, such as predictive modeling, real-time monitoring, and decision-support systems, have already demonstrated their capacity to optimize water resource allocation, enhance ecological resilience, and improve disaster preparedness. These are notwithstanding the significant technical, economic, and regulatory

hurdles that are limiting pervasive spread of AI yet again underlines the call for continuous innovation and strategic investments.

The further development of AI for catchment management is heavily associated with emerging technologies, notably drones, IoT devices, and real-time sensors. These instruments alone, when combined with high-performing machine learning algorithms, will revolutionize the monitoring and management of hydrological systems. AI-powered predictive analytics can more accurately forecast water demand, land-use changes and their implications, and extreme weather events. In turn, this contributes to efficiency in water use, the protection of biodiversity, and enabling communities to adapt to environmental pressures, all core components of long-term sustainability.

This will only be fully achieved by the development of an increase in technical capacity through focused investments in education and the workforce. Targeted training programs combining machine learning, data science, and hydrology expertise are crucial in narrowing the skills gap in order to fast-track AI technologies' adoption. Of importance at an equal degree is data infrastructure development. While AI applications are as good as the high-quality, region-specific datasets they build on, too many regions lack proper data collection systems. This therefore calls for governments and institutions to give priority to deploying IoT sensors, satellite monitoring systems, and standardized data-sharing platforms to ensure information is reliable and accessible.

The regulatory frameworks are also very critical in shaping the future of AI in watershed management. These policies should address all ethical concerns, be transparent regarding decision-making using algorithms, and provide equitable access to the AI tools. Ensuring coherence in approaches toward catchment governance strategies, a suite of activities across the various jurisdictions must thereafter be effected. Coupled with these policies will be the necessity of engaging communities. First, AI systems need to bring out complex hydrological information into more communicable forms in order for local communities to understand through such decision-making processes, hence utilizing such to create stakeholders' trust.

This means, finally, the full exploitation of the collaboration potential between academia, industry, and government in the promotion of research and innovation in AI applications. Indeed, digital twins, for example, and Edge computing are two of many promising emerging technologies to be further explored that will tackle present limitations with respect to data heterogeneity and inefficiencies of computation. Therefore, innovation would take over if different initiatives of collaborations, pooling resources and knowledge, work in

driving innovation forward with a common cause of gaps in infrastructure or even regulatory issues.

In conclusion, AI offers a transformative path forward for watershed management, enabling more adaptive, resilient, and sustainable solutions to water resource challenges. By investing in technical capacity, improving data infrastructure, establishing clear regulations, and fostering collaboration, stakeholders can unlock the full potential of AI to protect vital water resources and achieve long-term ecological, economic, and social sustainability.

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## Chapter 13

### **Harnessing the Potential of Underutilized and Indigenous Vegetables (UIVs): A Pathway to Sustainable Food Systems and Nutrition Security**

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## 1. Introduction: The Significance of Underutilized and Indigenous Vegetables (UIVs)

The growing global population, projected to peak at 11.2 billion by 2100 (Taagepera & Nemčok, 2024), has intensified the demand for sustainable food systems to meet the dietary needs of humanity. Despite advances in modern agriculture, malnutrition and diet-related diseases persist globally, with 821 million undernourished people, 2 billion individuals suffering from micronutrient deficiencies, and a parallel rise in obesity and diet-related conditions like diabetes and hypertension (Hunter et al., 2019). These challenges call for a shift towards nutrient-rich, sustainable, and diverse diets, where underutilized and indigenous vegetables (UIVs) can play a transformative role.

UIVs represent a vast group of crops, traditionally cultivated in specific regions but underrepresented in modern agricultural systems. With over 7,000 edible plant species globally, only a fraction is extensively used in mainstream agriculture, leaving thousands of nutrient-dense crops overlooked (Hunter et al., 2019). UIVs, often classified as neglected or underutilized species (NUS), are rich in essential nutrients and adapted to diverse agro-climatic conditions. Their inclusion in food systems can address malnutrition, enhance biodiversity, and promote agricultural sustainability. For example, crops like cowpea leaves (*Vigna unguiculata*) and jute mallow (*Chorchorus olitorius*) are rich in vitamins and minerals, offering solutions to micronutrient deficiencies while thriving in challenging environments (Sultanbawa & Sivakumar, 2022; Hunter et al., 2019).

Historically marginalized by specialized modern agriculture, these species hold significant potential for sustainable food systems. Regions such as Africa, Asia, and South America host a wealth of biodiversity, with UIVs offering resilience against climate change, pests, and water scarcity (Rosero et al., 2020). Their role in promoting agro-biodiversity and enhancing diets aligns with global efforts to mitigate food insecurity and environmental challenges.

Projects like Biodiversity for Food and Nutrition (BFN) further highlight the value of UIVs by identifying nutrient-rich crops that contribute to sustainable diets. Examples include sweet potato (*Ipomoea batatas*) for vitamin A, amaranth (*Amaranthus dubius*) for iron and calcium, and spider plant (*Cleome gynandra*) for beta-carotene and calcium (Hunter et al., 2019). These vegetables not only diversify diets but also foster sustainable agricultural practices, emphasizing the critical need to harness their potential.

By integrating UIVs into food systems, we can address pressing challenges like malnutrition, biodiversity loss, and unsustainable farming, paving the way for a resilient and equitable global food system.

## 2. Global Perspective: Potential Vegetable Species by Continent

Neglected and underutilized vegetables (UIVs) hold immense potential in addressing climate resilience, food security, and nutritional diversity. Across various continents, these crops provide not only sustainable agricultural practices but also a pathway to biodiversity conservation and dietary enrichment. Recent research highlights the value of these species in mitigating the impacts of climate change, reviving culinary traditions, and improving human health.

Sánchez-Mata et al. (2012) analyzed the bioactive potential of 15 traditionally consumed UIVs from the Mediterranean region. They found that species such as black bryony (*Tamus communis*), chicory (*Cichorium intybus*), and dandelion (*Taraxacum obovatum*) exhibited unique organic acid profiles, with high levels of citric, malic, and oxalic acids. Vitamin C content varied widely, with species like bladder campion (*Silene vulgaris*) and fennel (*Foeniculum vulgare*) retaining higher levels in raw form, underscoring the nutritional value of these crops.

Mabhaudhi et al. (2017) identified priority UIVs in South Africa, including bottle gourd (*Lagenaria siceraria*), cowpea (*Vigna unguiculata*), and jute mallow (*Corchorus olitorius*). These crops demonstrate resilience to drought and heat stress while providing high nutritional value. The study emphasized promoting these crops through targeted resources and indigenous knowledge integration to enhance value chains and food security.

De Sousa and Solberg (2020) focused on conservation gaps for traditional vegetables in Europe, such as wild leek (*Allium ampeloprasum*) and garden sorrel (*Rumex acetosa*). Reviving these crops could promote biodiversity, improve diets, and address food insecurity. Similarly, Jena et al. (2018) highlighted over 60 underexplored vegetable species in India, such as water spinach (*Ipomoea aquatica*) and winged bean (*Psophocarpus tetragonolobus*), stressing their role in enhancing rural livelihoods and nutritional security.

Bhatt et al. (2009) documented "Tum-thang" (*Crotalaria tetragona* Roxb. ex Andr.), a lesser-known wild edible from India's northeastern hill region. This Fabaceae species, used in traditional cuisine, has potential for conservation and further research. McClatchey (2012) explored wild food plants in Remote Oceania, revealing their historical role in early agricultural societies and current significance in dietary diversity.

- *Africa*: Research on African eggplants (*Solanum macrocarpon*, *Solanum aethiopicum*) by Oladosu et al. (2021) highlights their breeding potential for pest resistance and improved yield.

- *Asia*: Thai wild purslane (*Portulaca oleracea*), studied by Siriamornpun and Suttajit (2010), exhibited high antioxidant and nutritional properties, particularly in its leaves and flowers.

- *Americas*: Indigenous vegetables like amaranth (*Amaranthus sp.*) and nightshade (*Solanum nigrum*) are recognized for their contributions to traditional diets and nutritional value.

- *Oceania*: Singh et al. (2018b) documented 42 indigenous UIVs from the Andaman and Nicobar Islands, emphasizing their richness in essential nutrients such as calcium, iron, and polyphenols.

UIVs across continents provide a wealth of untapped potential for improving global food systems. These crops, with their nutritional, ecological, and cultural significance, highlight the importance of integrating traditional knowledge with modern agricultural practices. Focused conservation, research, and promotion efforts can pave the way for a resilient, nutritious, and sustainable food future.

### 3. SWOT Analysis of Indigenous and Underutilized Vegetables (UIVs)

Indigenous and underutilized vegetables (UIVs) are increasingly recognized for their potential to address global challenges, such as food insecurity, climate change resilience, and biodiversity conservation. These crops are often adapted to marginal environments and possess unique nutritional profiles, making them valuable for promoting health and sustainable agricultural practices. Despite their benefits, UIVs face numerous challenges, including limited market accessibility, insufficient research and extension support, and socio-cultural barriers to adoption. The application of SWOT (Strengths, Weaknesses, Opportunities, and Threats) analysis offers a structured approach to understanding the factors that influence the development and integration of UIVs into modern food systems. Research findings from various regions highlight the critical need for targeted policies, investments, and capacity-building efforts to harness the potential of UIVs while addressing their limitations.

#### *Strengths: Nutritional Density, Adaptability, and Low Input Requirements*

El Bilali et al. (2023) conducted a comprehensive SWOT analysis of neglected and underutilized species (NUS), highlighting their remarkable strengths. These crops exhibit climate resilience, drought tolerance, and adaptability to marginal soils. Their inherent resistance to pests and diseases

further enhances their suitability for sustainable agriculture. Nutritionally, NUS are rich in health-enhancing compounds and possess high consumer acceptability. Similarly, Agulanna (2020) emphasizes the nutrient density of UIVs in Nigeria, such as bitter leaf (*Vernonia amygdalina*), fluted pumpkin (*Telfairia occidentalis*), and green amaranth (*Amaranthus viridis*), which are rich in essential minerals like iron, calcium, and magnesium. These crops contribute to poverty reduction and dietary diversity, particularly in rural areas.

#### *Weaknesses: Limited Awareness and Market Accessibility*

Despite their strengths, UIVs face significant challenges. El Bilali et al. (2023) identify limited access to quality seeds, low production levels, and inadequate processing and storage technologies as major weaknesses. Additionally, the absence of organized value chains and markets hinders their development, leading to low market value and high consumer prices. Onyema et al. (2013) echo these findings in their analysis of underutilized crops in southeastern Nigeria, where the lack of agricultural research institutions and poor access to extension services impede progress. Socio-cultural resistance to UIVs, often perceived as "foods of the poor," further restricts their widespread adoption (Jairu et al., 2023).

#### *Opportunities: Climate Change Resilience and Food Security Potential*

UIVs present immense opportunities to address pressing global challenges. El Bilali et al. (2023) highlight their potential in combating food insecurity and poverty through sustainable, diversified diets. Advances in genetic research and modern breeding technologies open new pathways for improving UIV varieties. Jairu et al. (2023) highlight their role as climate-smart crops, capable of mitigating the impacts of climate change while enhancing nutritional security. In Nigeria, Onyema et al. (2013) note the medicinal and health benefits of crops like *Solanum spp.* and *Amaranthus spp.*, which hold promise for increasing local market production and consumption. Agulanna (2020) also points to the need for targeted policies to promote UIV cultivation, thereby strengthening food systems and improving public health.

#### *Threats: Loss of Biodiversity and Cultural Knowledge*

While UIVs offer numerous advantages, they are threatened by various factors. El Bilali et al. (2023) identify climate change, land degradation, and invasive pests as critical threats to their sustainability. The erosion of cultural knowledge and traditional practices further exacerbates these challenges. Onyema et al. (2013) highlight competing land uses and farmland theft as

barriers to the sustainable cultivation of underutilized crops in Nigeria. Jairu et al. (2023) discuss the economic barriers and socio-cultural resistance that hinder the mainstream adoption of these crops. To mitigate these threats, strengthening public-private partnerships (PPP) and fostering research collaborations are crucial. Onyema et al. (2013) recommend establishing agro-based institutions and promoting modern land-use practices to support sustainable development.

The SWOT analysis highlights the multifaceted role of UIVs in enhancing global food systems. Their nutritional, ecological, and cultural significance highlights the need for concerted efforts to integrate traditional knowledge with modern agricultural practices. By addressing weaknesses and threats while leveraging strengths and opportunities, UIVs can contribute to a resilient, nutritious, and sustainable food future.

#### **4. Cultural and Nutritional Dimensions of UIVs**

Underutilized indigenous vegetables (UIVs) hold immense potential for addressing global challenges related to malnutrition, food security, and agricultural sustainability. As reviewed by Baldermann et al. (2016), neglected and underutilized plant species (NPs) offer diverse nutrients, vitamins, and bioactive compounds crucial for low-income populations, especially in areas where access to animal-based foods is limited. These species also contribute to sustainable agro-food systems by enhancing dietary diversity and agricultural resilience. Studies from diverse regions emphasize their ethnobotanical significance, highlighting their role in traditional cuisines, cultural identity, and nutritional well-being. Coordinated efforts integrating these species into mainstream food systems are imperative to preserve biodiversity, improve supply chains, and ensure food security for future generations (Baldermann et al., 2016).

##### *Traditional Cuisines and Ethnobotanical Significance*

UIVs are deeply embedded in traditional cuisines, serving as vital components of cultural heritage. Kadioglu et al. (2024) documented 128 plant species in the Sivas region of Türkiye, emphasizing the culinary value of wild vegetables, such as honeywort (*Cerintho minor*), barberry (*Berberis vulgaris*), and wild mint (*Mentha longifolia*). These plants enrich local dishes with unique flavors, showcasing their ethnobotanical importance. Furthermore, the repeated use of local names like "Yemlik" (*Tragopogon* spp.) highlights linguistic and cultural ties, reinforcing the role of UIVs in regional identity.

Borelli et al. (2022) further explored the cultural dimensions of wild food plants (WFPs) in Türkiye, highlighting their significance in traditional diets and rural livelihoods. With over 11,000 plant species - 10% of which are edible - Türkiye's biodiversity hotspots are a treasure trove of UIVs that contribute to household diets and local economies. Events like the Alaçatı Herb Festival exemplify the integration of gastronomy with cultural heritage, preserving these traditions while promoting healthier dietary practices.

#### *Nutritional Profiles and Bioactive Compounds*

UIVs are nutrient-dense, offering a sustainable alternative to processed foods. Sultanbawa and Sivakumar (2022) emphasized the health benefits of UIVs like *Solanum* spp., which are rich in calcium, iron, antioxidants, and phytochemicals that combat diet-related diseases. Despite their nutritional potential, limited research on their bioaccessibility and nutrient retention restricts their broader adoption. Expanding knowledge in these areas could unlock their functional food applications.

Similarly, Oseni and Olawoye (2015) highlighted the antioxidant properties of Nigerian UIVs, such as wild eggplant (*Solanum macrocarpon*) and fireweed (*Crassocephalum crepidoides*), which contain polyphenols that mitigate oxidative stress. In Ghana, Nyadanu and Lowor (2015) revealed that IUVs, including amaranth (*Amaranthus cruentus*) and tannia (*Xanthosoma sagittifolium*), are superior in nutritional composition compared to exotic counterparts, making them essential for addressing malnutrition and food insecurity.

#### *Addressing Hidden Hunger and Micronutrient Deficiencies*

UIVs play a critical role in combating hidden hunger and micronutrient deficiencies. Studies from Türkiye and other regions have demonstrated their contributions to dietary diversity. For instance, Cakmakci et al. (2022) analyzed bioactive constituents in *Allium vineale*, a wild edible vegetables species used in herbed cheese production, revealing its high phenolic content and antioxidant capacity. Likewise, Aktar et al. (2020) demonstrated the medicinal value of cultivated artichoke varieties and wild artichoke (*Cynara syriaca*) genotypes, further emphasizing the potential of these plants to improve nutritional health.

#### *Biodiversity Conservation and Genetic Resources*

Conserving the genetic diversity of UIVs is vital for agricultural and nutritional improvements. Altıntaş et al. (2019) studied native *Asparagus* species in the Lake Van Basin, finding significant genetic diversity and

antioxidant properties. These findings highlight the importance of preserving wild species for breeding programs. Similarly, Türkmen et al. (2005) evaluated endemic wild rhubarb (*Rheum ribes L.*) in Eastern Anatolia, selecting high-performing genotypes for further cultivation and conservation. These efforts emphasize the ecological and agricultural significance of UIVs in sustaining biodiversity and enhancing food security.

#### *Integrating UIVs into Sustainable Food Systems*

Integrating UIVs into sustainable food systems requires evidence-based policies, consumer education, and value chain development (Borelli et al., 2022). Efforts to improve supply chains, promote gastronomy, and develop processing technologies can maximize their nutritional and economic potential. Sultanbawa and Sivakumar (2022) advocate for profiling underutilized vegetables to enhance their bioavailability and functional food applications. Such initiatives could address global challenges related to malnutrition, biodiversity loss, and unsustainable food systems.

UIVs represent a nexus of cultural, nutritional, and ecological significance. Their integration into mainstream diets and sustainable agriculture is crucial for addressing hidden hunger, preserving biodiversity, and promoting healthier, more resilient food systems.

#### **5. Sustainability and Resilience: UIVs in Changing Climates**

In an era of increasing climate variability and limited water resources, sustainable agriculture necessitates innovative approaches that enhance resilience and efficiency. Traditional reliance on a narrow range of staple crops has led to agronomic, ecological, and economic vulnerabilities, underscoring the need for diversification. Incorporating underutilized indigenous vegetables (UIVs) and the other neglected and underutilized crop species (NUCS) into agricultural systems presents an opportunity to address these challenges. These crops, often overlooked in modern agricultural practices, are not only adaptive to water-scarce conditions but also contribute significantly to nutritional security and economic stability. This section explores the potential of drought-tolerant species and agro-biodiversity as key strategies to achieve sustainable and resilient food systems in changing climatic conditions.

#### *Water-Scarce Agriculture: Opportunities for Drought-Tolerant Species*

Ebert (2014) highlights the pressing need to diversify agricultural systems by incorporating underutilized traditional vegetables and legume crops. This

approach enhances agro-biodiversity, reducing the risks associated with dependency on a limited number of staple crops. The study introduces amaranth (*Amaranthus spp.*) and mungbean (*Vigna radiata*) as examples of underutilized crops that offer dual benefits: nutritional and economic. These crops are rich sources of essential vitamins, micronutrients, and protein, addressing nutritional security, while their cultivation can bolster household income. Such dual-purpose crops are particularly valuable in regions prone to biotic and abiotic stress, where resilience is paramount. Ebert (2014) advocates for investments in research and breeding programs to unlock the full potential of these species, thus fostering sustainable food systems.

#### *Enhancing Agro-Biodiversity for Food System Stability*

Chivenge et al. (2015) emphasize the untapped potential of IUVs in ensuring food and nutrition security under water-scarce conditions, particularly in Sub-Saharan Africa (SSA). These crops, including African leafy IUVs such as wild watermelon (*Citrullus lanatus var. citroides*), wild mustard (*Brassica juncea* and *Brassica nigra*), and amaranth (*Amaranthus spp.*), are highly resilient due to their adaptability to low-input agricultural systems and inherent drought tolerance. Grain legumes like Bambara groundnut (*Vigna subterranea*) and cowpea (*Vigna unguiculata*), along with root and tuber crops such as sweet potato (*Ipomoea batatas*) and taro (*Colocasia esculenta*), exhibit high water-use efficiency, making them ideal for semi-arid environments. These species not only thrive under harsh conditions but also contribute significantly to nutritional diversity and income generation.

Chivenge et al. further highlight the critical role of scientific research in realizing the commercial potential of these crops. Areas such as breeding for drought tolerance, agronomic optimization, and post-harvest management are identified as key to enhancing the viability of IUVs. By addressing these research gaps, it becomes possible to integrate IUVs into sustainable agricultural practices, thus improving the resilience of food systems in SSA.

#### *Towards a Sustainable Future*

The integration of IUVs into modern agricultural systems represents a transformative strategy for addressing climate-induced challenges in food production. By diversifying crop portfolios and investing in targeted research, these crops can play a pivotal role in building sustainable and resilient food systems globally. Enhanced agro-biodiversity not only mitigates risks associated with climate variability but also promotes ecological stability and economic empowerment in vulnerable regions.

## 6. Economic Potential: From Local Use to Global Markets

The economic potential of underutilized and indigenous vegetables (UIVs) is vast, spanning from local consumption in rural communities to their integration into global markets. UIVs, cultivated in diverse systems like home gardens, play a crucial role in enhancing agro-biodiversity, promoting dietary diversity, and generating income for smallholder farmers. While affluent countries are experiencing a rising preference for organic and nutraceutical products amidst concerns over the contamination of industrially produced food with synthetic fertilizers, pesticides, and additives, poorer regions rely on UIVs out of necessity. These vegetables are often collected from the wild or grown in small-scale agricultural systems, serving as a vital source of nutrition and food security. Bridging this dichotomy requires investments in value addition, post-harvest management, and market linkages to harness the potential of UIVs for sustainable development globally.

### *Value Addition and Post-Harvest Management*

Home gardens are essential for preserving agro-biodiversity and are especially significant for cultivating UIVs. These systems sustain diverse plant genetic resources, including traditional crops and wild species, which are vital for nutrition, food security, and small-scale livelihoods. UIVs grown in home gardens contribute to dietary diversity while preserving agricultural knowledge and cultural heritage. According to Galluzzi et al. (2010), promoting home gardens as multifunctional ecosystems can facilitate the conservation and utilization of UIVs, simultaneously aligning biodiversity preservation with community well-being.

Value addition and effective post-harvest management are critical for unlocking the full economic potential of UIVs. Proper handling, processing, and packaging can enhance the marketability of these vegetables, extending their shelf life and increasing their appeal to consumers in urban and international markets. Investing in infrastructure and training for post-harvest practices can transform UIVs from subsistence crops into profitable commodities.

### *Linking Smallholder Farmers to Markets*

The economic significance of UIVs extends beyond their role in nutrition and food security. Tanimonure et al. (2021) emphasized their contribution to household dietary diversity (HDD) and rural livelihoods in Southwest Nigeria. Factors such as marital status, proximity to farms, farm area dedicated to UIVs, and off-farm income significantly influence the integration of UIVs into household diets. The study demonstrated that incorporating UIVs into cropping

systems and home gardening practices not only supports food diversity but also provides economic resilience through income generation. Additionally, the gross margin from UIV cultivation highlights their potential as a viable agricultural enterprise.

Building market linkages for smallholder farmers growing UIVs can bridge the gap between rural producers and urban consumers. Organized value chains, cooperatives, and support for market access are essential for enhancing the economic viability of UIVs, fostering their integration into mainstream agricultural systems.

#### *Contrasting Perspectives: Wealthy vs. Poor Nations*

A stark contrast exists between the role of UIVs in wealthy and poor nations. In affluent countries, the increasing demand for organic, functional, and nutraceutical foods reflects growing health consciousness and environmental awareness. However, industrial agricultural practices dominate these regions, often leading to concerns about contamination from synthetic inputs. Conversely, in poorer regions, UIVs are indispensable due to limited access to commercial vegetables. Rural communities often depend on wild harvesting and small-scale cultivation to meet their nutritional needs.

Efforts to address this disparity should focus on promoting UIVs in both contexts. For affluent markets, highlighting the health benefits, culinary versatility, and environmental sustainability of UIVs can drive consumer interest. In poorer regions, supporting community-led initiatives, research, and extension services can enhance the productivity and marketability of UIVs, empowering smallholder farmers and improving livelihoods.

#### *Consumer Demand and Market Trends*

Ayanwale et al. (2016) analyzed consumer demand for UIVs using the quadratic almost ideal demand system (QUAIDS) in Nigeria. The study revealed that UIVs such as fluted pumpkin (*Telfairia occidentalis*), Ethiopian eggplant/nakati (*Solanum aethiopicum*), slender amaranth (*Amaranthus viridis*), and English spinach (*Senecio biafrae*) are essential food items with relatively inelastic demand. This indicates their importance in household diets and highlights their role as staple crops. Furthermore, the identification of UIVs as normal goods with inelastic and complementary cross-price elasticities highlights their integration into household consumption alongside other vegetables. The findings from Ayanwale et al. (2016) suggest that UIVs represent a niche market with significant potential. By investing in targeted

marketing, value addition, and consumer education, these crops can gain wider acceptance, contributing to both local and global food systems.

## 7. Research and Policy Frameworks for UIVs

Underutilized Indigenous Vegetables (UIVs) hold immense potential to address global challenges such as food security, climate change adaptation, and nutritional deficiencies. These crops, often neglected in mainstream agricultural systems, are rich in bioactive compounds, adaptable to diverse climatic conditions, and vital for supporting low-income communities. Recent research has emphasized the need to conserve, commercialize, and improve UIVs through innovative breeding, agronomy, and policy interventions (Ibok and Asuquo, 2022; Orkaa and Ayanwale, 2021).

A growing body of literature highlights biotechnological advancements aimed at enhancing the resilience and productivity of UIVs under abiotic stresses. Rai et al. (2021) review the application of genetic engineering tools, including CRISPR/Cas systems and synthetic biology, to improve heavy metal tolerance in neglected legumes such as winged bean (*Psophocarpus tetragonolobus*) and lablab-bean (*Lablab purpureus*). These approaches target the overexpression of stress-induced genes, detoxification pathways, and phytoremediation capabilities, supported by genome-scale models and advanced bioinformatics for precise stress-response predictions. Similarly, Rosero et al. (2020) highlight the value of wild relatives and landraces in breeding programs, leveraging traits for drought tolerance through marker-assisted selection and CRISPR technologies. This integration of genomics and phenomics aims to ensure food security in water-scarce regions.

Efforts to enhance UIV productivity extend beyond genetic tools. Jain (2005) highlighted mutation-assisted breeding programs facilitated by the FAO/IAEA, focusing on salinity and drought tolerance in underutilized crops. These programs utilize tissue culture and DNA marker-assisted selection to diversify food systems and bolster nutrition security. Furthermore, studies like those by Ambika et al. (2022) emphasize the importance of preserving genetic variability during domestication while accelerating crop improvement using “omics” technologies.

Nutritional and functional values of UIVs are equally pivotal. Andarwulan et al. (2012) demonstrated the rich polyphenol, carotenoid, and ascorbic acid content of Indonesian UIVs such as katuk or sweet leaf (*Sauropus androgynous*), suggesting their potential as functional foods. Processing innovations, as discussed by Ibok and Asuquo (2022), can transform these vegetables into value-added products, thereby extending shelf life and

increasing market value. Ekemini-Richard et al. (2022) further emphasized the role of UIVs in climate change adaptation, citing their affordability and adaptability as critical for promoting environmental and economic resilience among farmers.

Policy and socioeconomic factors also play a significant role in promoting UIV adoption and production. Orkaa and Ayanwale (2021) and Ajekiigbe et al. (2018) identified key drivers of adoption, including education, awareness, and access to innovative practices, which significantly enhance productivity and technical efficiency. Strengthening farmer networks and information dissemination systems is crucial for scaling these benefits.

Overall, the integration of advanced breeding techniques, nutritional research, and supportive policies can elevate the role of UIVs in sustainable agricultural systems. By bridging gaps in research and practice, these efforts can contribute to global food security, improved nutrition, and climate resilience.

## 8. Case Studies: Success Stories in Promoting UIVs

Underutilized indigenous vegetables (UIVs) hold immense potential in promoting sustainable agriculture, enhancing nutritional security, and addressing climate change challenges. While many of these species have been overshadowed by the rise of staple crops and more commercially viable vegetables, their resilience, adaptability, and rich nutritional profiles position them as crucial resources in future food systems. This section highlights case studies demonstrating the success of UIVs in enhancing technical efficiency, diversifying diets, and promoting sustainable agricultural practices. By exploring examples such as Indian spinach, seapurslane, and amaranth, this analysis highlights the importance of revitalizing these species through research, breeding programs, and integration into agricultural systems.

### *Indian Spinach*

Singh et al. (2018a) investigated *Basella alba* and *Basella rubra*, commonly known as Indian spinach or Malabar spinach, as underutilized perennial leafy vegetables with high nutritional potential. Belonging to the Basellaceae family, these species are well-suited to diverse soil conditions, from acidic to alkaline and nutrient-poor environments. Despite their rich nutritional composition - including proteins, vitamins A and C, and essential minerals like calcium, iron, magnesium, potassium, and zinc - Indian spinach has been overshadowed by other greens. Historically cultivated in Southeast Asia and China, it is now increasingly neglected. However, its ability to thrive in hot, humid tropical

climates makes it an ideal crop for addressing the challenges posed by global warming.

Singh et al. (2018a) emphasize the species' minimal agronomic requirements and suitability for resource-poor farmers in developing regions. Their findings advocate for expanded field surveys and breeding programs to improve the adaptability, nutritional quality, and environmental resilience of *Basella* species. By encouraging the cultivation of locally available, underutilized species, this approach can enhance agro-biodiversity, sustainability, and nutritional security in tropical and subtropical areas.

### *Seapurslane*

Lokhande et al. (2009) examined *Sesuvium portulacastrum* (L.) L., commonly known as seapurslane, a halophyte with exceptional potential for cultivation in saline and arid regions. Native to subtropical, Mediterranean, and coastal areas, seapurslane serves as both a vegetable and forage crop. Environmentally, it contributes significantly to bioreclamation of saline soils, offering a practical solution for land restoration in arid and semi-arid areas. The study highlighted its geographical distribution in India and potential applications in bioremediation, particularly in coastal and saline environments.

Seapurslane emerges as a viable candidate for seawater agriculture, a method utilizing saltwater for irrigation in coastal and inland salt deserts, which constitute approximately 15% of global land. Lokhande et al. (2009) propose that cultivating halophytes like seapurslane is more cost-effective and efficient than engineering salt tolerance in conventional crops. By integrating halophytes into saline farming systems, this approach can improve soil fertility by absorbing excess salts and provide biomass for human consumption, animal feed, and industrial applications. Moreover, seapurslane's secondary metabolites, such as phytoecdysteroids, show potential for use in industries like sericulture. This case study highlights the potential of halophytes to address food security challenges, restore degraded land, and support sustainable agricultural practices in resource-constrained regions.

### *Amaranth*

Rastogi and Shukla (2013) reviewed amaranth's potential as a future crop with significant nutraceutical value. Amid growing populations and plateauing yields of staple crops, particularly in developing countries, amaranth offers a promising alternative. With its adaptability to harsh growing conditions such as heat and drought, resistance to diseases, and impressive yield potential, this underutilized crop is poised to address global food demands.

Amaranth is nutritionally dense, containing high levels of proteins, vitamins A and C, essential minerals, and amino acids, making it ideal for combating micronutrient deficiencies. Its genetic diversity and phenotypic plasticity present opportunities for developing nutritionally enriched varieties through breeding programs. Beyond its agricultural benefits, amaranth's potential extends to wasteland cultivation, oil extraction, renewable energy production, and pharmaceutical applications. As emphasized by Rastogi and Shukla (2013), integrating amaranth into agricultural systems could play a critical role in mitigating malnutrition and promoting sustainable agriculture in developing regions.

The examples of Indian spinach, seapurslane, and amaranth illustrate the significant potential of underutilized indigenous vegetables in advancing sustainable agriculture, improving nutritional security, and addressing environmental challenges. These case studies highlight the importance of research, breeding programs, and policies to integrate these crops into mainstream agriculture. Revitalizing UIVs offers an opportunity to diversify diets, enhance agro-biodiversity, and ensure resilience in the face of global agricultural challenges.

## **9. Challenges and the Way Forward: Bridging Gaps in Research, Conservation, and Awareness**

Underutilized and Indigenous Vegetables (UIVs) between the other Neglected and Underutilized Crop Species (NUCS) represent a significant untapped potential in global food systems. While staple crops like rice, wheat, and maize dominate global diets, IUVs offer a rich source of essential nutrients and are critical to enhancing biodiversity and food system sustainability. Despite their promise, these species remain underrepresented in both agricultural research and global consumption. Bridging the gaps in research, conservation, and awareness, and adopting multi-stakeholder approaches, are pivotal to scaling up the adoption of UIVS in food systems and promoting agricultural diversification. Efforts to integrate UIVS into school meals, public procurement, and dietary guidelines in countries such as Brazil, Kenya, Sri Lanka, and Türkiye, under the Biodiversity for Food and Nutrition Project, demonstrate their potential to enhance nutrition, support local economies, and contribute to sustainable agriculture. However, significant challenges remain in the way of widespread adoption, which include production, supply chain inefficiencies, and consumption barriers.

Hunter et al. (2019) highlight the key obstacles to integrating NUCS and UIVS into food systems, illustrating the complexities through a schematic diagram. These barriers span various levels, including production, supply chains, and consumption. At the production level, issues such as limited access to quality seeds, inadequate research, and poor market incentives hinder the successful cultivation of these species. The supply chain faces its own challenges, with inefficient processing systems, a lack of infrastructure, and weak market links preventing UIVS from reaching wider markets. On the consumption side, low consumer awareness, cultural biases, and insufficient policy support continue to limit the adoption of these crops. Addressing these barriers through coordinated policy support, investment in research, and raising awareness is crucial for unlocking the full potential of UIVS.

Bhatt et al. (2019) explore the potential of UIVs in addressing critical issues such as food security, nutrition, and income generation. These crops, which thrive in marginal and stress-prone environments, possess unique genetic variability, adaptability, and ethnobotanical significance. While their nutritional and medicinal value is well-known locally, their commercial importance remains largely untapped. Examples of UIVs include leafy amaranth (*Amaranthus* spp.), kangkong (water spinach, *Ipomoea aquatica*), aibika (sunset hibiscus, *Abelmoschus manihot*), and crops from the Cucurbitaceae family like wax gourd (*Benincasa hispida*) and bitter melon (*Momordica charantia*). Bhatt et al. (2019) emphasize the need for targeted research, development of post-harvest technologies, and the creation of strategic action plans to unlock the agricultural and economic potential of UIVs for sustainable development.

Imathiu (2021) discusses the importance of leafy African UIVs for food and nutrition security, particularly in sub-Saharan Africa. These vegetables have been integral to local diets for centuries, offering superior nutritional qualities and climate adaptability compared to exotic crops. Rich in micronutrients and phytochemicals, UIVs play a vital role in combating "hidden hunger" in developing countries. Their resilience to climate challenges, particularly in regions reliant on rain-fed agriculture, positions them as climate-smart crops. The growing recognition of their health benefits has led to increased production and consumption, often resulting in higher prices for farmers. However, challenges such as underdeveloped value chains, limited scientific documentation, and a lack of breeding programs impede the full realization of their potential. To maximize the benefits of UIVs, there is a need for enhanced research, improved seed quality, value addition, and a deeper understanding of consumer preferences. With continued promotion and support, UIVs can significantly contribute to food security and improve livelihoods in rural

communities, supporting both local economies and global efforts to promote sustainable food systems.

## **10. Conclusion: A Call to Action for Sustainable Food Futures**

This chapter has synthesized the growing body of literature on underutilized and indigenous vegetables (UIVs), providing a robust framework for their role in addressing global challenges such as malnutrition, food security, and climate resilience. The value of UIVs extends beyond their ecological and cultural significance, positioning them as vital components in the future of sustainable food systems.

The role of vegetables in promoting healthier diets cannot be overstated. Vegetables are essential to providing essential nutrients and bioactive compounds crucial for human health. However, modern diets, which largely depend on a narrow range of high-yielding, input-dependent vegetable species, fail to provide the necessary diversity to meet the global nutritional needs. This is where UIVs become critical: their rich nutritional profiles, adaptability to marginal environments, and potential to reduce reliance on synthetic inputs make them indispensable for more resilient and diverse food systems. The heavy reliance on artificial fertilizers, pesticides, and intensive irrigation in contemporary agricultural practices has led to the dominance of a few high-yielding varieties, often at the expense of nutrition and sustainability. By integrating UIVs into breeding programs, we can not only revitalize these species but also enhance their resilience and productivity, particularly in the face of abiotic and biotic stresses. UIVs' inherent traits - such as tolerance to drought, poor soils, and extreme weather conditions - make them invaluable assets in addressing food insecurity and climate change.

It is believed that the inclusion of UIVs into breeding programs is critical for advancing agricultural sustainability. These vegetables are highly adapted to complex and often marginal environments, and their integration into modern agricultural systems could help diversify crop portfolios, making them more resilient to the challenges posed by climate change. Moreover, leveraging the potential of biostimulants (Sensoy, 2024), can further enhance the stress tolerance of UIV cultivars, providing a powerful tool in crop development and management.

Rural communities, especially those in developing regions, have traditionally relied on wild edible vegetables as important food sources. However, as urbanization increases, the knowledge of these species is rapidly diminishing. The next generation is less interested in nature and traditional agricultural practices, leading to a disconnect from these vital food resources.

Moreover, wild vegetables, though rich in essential micronutrients, often face challenges such as fast bolting, seed shattering, and bitterness, etc., which hinder their widespread adoption. To make these vegetables more viable for both producers and consumers, extensive plant improvement programs are necessary to enhance their agronomic traits, such as reducing bitterness or bolting. By focusing on improving these wild species, we can create more reliable and accessible food sources for vulnerable populations.

As highlighted by Flyman and Afolayan (2006), wild vegetables are an untapped resource in combating global micronutrient deficiencies, which affect over two billion people worldwide. These deficiencies, particularly of iron, iodine, and vitamin A, have detrimental effects on health, cognitive development, and immunity. Wild vegetables, being rich in these essential nutrients, provide a sustainable solution to the global challenges of malnutrition and food insecurity. Their resilience to extreme growing conditions and ability to thrive without heavy inputs make them particularly valuable for rural and resource-poor areas. However, to fully harness their potential, more research is needed to better understand their nutritional properties and the bioavailability of the nutrients they contain. This research could pave the way for incorporating UIVs into mainstream diets and global food systems.

In conclusion, underutilized and indigenous vegetables offer significant promise for sustainable food systems, nutrition security, and climate resilience. Their potential to contribute to biodiversity, enhance dietary diversity, and support sustainable agricultural practices should not be underestimated. However, the path forward requires collaborative efforts in research, policy, and agricultural innovation to integrate UIVs into modern food systems. By bridging the knowledge gap and addressing the barriers to their adoption, we can unlock the full potential of these crops, ensuring that they play a pivotal role in building a more sustainable, resilient, and nutritious food future for all.

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## Chapter 14

### **Genetic Approaches to Enhancing Nutritional Quality in Vegetables through Nutraceutical Breeding**

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## 1. Introduction

### *Importance of Nutraceuticals in Vegetables*

Nutraceuticals, which are bioactive compounds derived from food sources, are increasingly recognized for their health-promoting properties, particularly in the prevention and management of chronic diseases. These compounds, including vitamins, minerals, and phytochemicals, contribute to improved physical and mental health, offering an alternative or complement to conventional pharmaceuticals (Parulekar et al., 2019; Lal et al., 2020). By enhancing dietary intake of these bioactive elements, nutraceutical-rich vegetables address critical global health concerns, including the rising prevalence of cancer, cardiovascular diseases, and diabetes.

The global burden of micronutrient malnutrition - often referred to as "hidden hunger" - further highlights the role of nutraceuticals in human diets. This form of malnutrition, linked to deficiencies in essential vitamins and minerals, disproportionately affects developing regions. Efforts to address hidden hunger through the consumption of biofortified vegetables are increasingly vital (Fatima et al., 2013). Recent advancements in genetic engineering and molecular breeding now facilitate the development of vegetables rich in essential nutrients, enhancing their capacity to improve public health and address global food security challenges.

### *The Role of Vegetables in Human Nutrition and Health*

Vegetables, consumed in various forms worldwide, are fundamental components of a balanced diet. They are typically low in fat and carbohydrates but high in vitamins, minerals, and dietary fiber. Leading producers, such as China, India, the United States, and Türkiye, supply a diverse array of vegetables (FAOSTAT, 2022), each offering unique health benefits. For instance, Brassica species (cole crops) contain glucosinolates, compounds with notable anticancer properties, while solanaceous crops, like tomatoes, are rich in lycopene, which supports cardiovascular health (Parulekar et al., 2019). Additionally, leafy greens like spinach and amaranth provide antioxidants such as vitamins E and C, supporting heart health and immune function.

Vegetables' potential to mitigate chronic conditions is closely linked to their bioactive compounds, or nutraceuticals. Glucosinolates, found in Brassica vegetables, for instance, have shown substantial anticarcinogenic effects. These sulfur-containing compounds, derived from amino acids, break down into biologically active forms when plant tissues are disrupted, thereby promoting health (Mithen et al., 2000). As research advances, breeding strategies focused on optimizing glucosinolate profiles in Brassicas are gaining traction, with the

ultimate goal of developing crops that provide maximum health benefits with minimized antinutritional effects.

### *Overview of Key Phytonutrients*

Phytonutrients in vegetables, categorized primarily as primary and secondary metabolites, confer specific health advantages. Primary metabolites support basic plant functions, while secondary metabolites, such as phenolics, terpenoids, alkaloids, and flavonoids, offer additional defense and health benefits (Saadullah et al., 2023). Phenolic compounds, notably abundant and structurally diverse, possess strong antioxidant properties that protect against oxidative stress, while flavonoids have demonstrated anti-inflammatory and anticancer effects. Terpenoids, derived from isoprene units, and carotenoids, such as lycopene found in tomatoes, further contribute to disease prevention.

One particularly promising class of compounds, anthocyanins, are pigments produced through the phenylpropanoid pathway that offer both visual appeal and health benefits. Anthocyanins have been linked to reduced risks of metabolic disorders and various cancers, yet their concentrations in most vegetables remain suboptimal. Efforts to increase anthocyanin levels through both transgenic and non-transgenic breeding approaches continue to advance (Chaves-Silva et al., 2018).

In addition to their antioxidant properties, anthocyanins serve as natural colorants, appealing to the food industry as alternatives to synthetic dyes. For instance, purple varieties of cabbage, onion, and eggplant owe their vibrant colors to anthocyanin accumulation in the epidermal tissues, making them attractive as both functional foods and visual enhancers in culinary applications (Petropoulos et al., 2019). Colored root vegetables, such as black carrots and beets, offer unique hues and health-promoting properties, highlighting the potential of these crops to meet rising consumer demand for natural additives.

### *Addressing Global Food Security with Resilient Crops*

As awareness of the health benefits of nutrient-dense vegetables grows, the development of resistant crops suited to diverse environmental conditions is also essential. For example, a leaf vegetable, amaranth, a nutritionally rich and adaptable crop, could provide a valuable resource for addressing food security, particularly in developing regions. Known for its high protein, mineral, and vitamin content, amaranth not only supports human health but also thrives under adverse conditions, tolerating heat, drought, and nutrient-poor soils (Rastogi & Shukla, 2013). Its versatility and resilience make it an ideal candidate for

breeding programs aimed at enhancing food security and nutritional quality in resource-constrained environments.

## **2. Conventional Breeding Approaches**

### *Selection and Hybridization*

Conventional breeding methods, including selection and hybridization, are traditional approaches to improve nutrient density and yield in crops. Through these methods, breeders harness genetic variation to introduce and intensify desirable traits within crops. For instance, selection, either mass or recurrent, works by reinforcing specific traits over multiple generations, though the process can be time-consuming. Hybridization combines genetic material from different lines, leading to new cultivars with enhanced nutrient profiles. However, the simultaneous improvement of yield and quality often faces trade-offs due to competing resource allocation within plants.

### *Limitations of Conventional Breeding*

Despite their successes, conventional methods come with inherent limitations such as the slow pace of progress and challenges in balancing yield and nutritional quality. Approaches like mutation and polyploidy breeding introduce novel genetic variations, enhancing yield and stress tolerance, but they also present complexities, including potential reductions in plant vigor or undesirable mutations.

Conventional breeding methods such as plant introduction, selection, hybridization, mutation breeding, and polyploidy breeding have profoundly influenced the nutritional quality and yield of numerous crop species, including vegetables (Fehr, 1991). By accessing genetic variation within and across species, these approaches have enabled breeders to incorporate traits that enhance resilience and quality. Plant introduction, for example, brings in genetic diversity from different environmental backgrounds, forming a foundation for improved resilience and nutrient-rich traits. Polyploidy breeding, on the other hand, can increase cell size, often boosting yield and tolerance to stress conditions.

A prime example of successful conventional breeding is the Illinois Long-Term Selection Experiment in maize, which has run for over a century and remains the longest continuous genetic study in higher plants. This project has created nine maize populations through recurrent selection, demonstrating notable shifts in grain composition, specifically oil and protein content. Over 100 generations, this experiment has shown both the potential for genetic enhancement and the challenges associated with gradual, long-term improvements through traditional selection. The work also illustrates the gene

complexity within these traits, as many genes, potentially over a hundred, contribute to variations in protein and oil content (Moose et al., 2004).

However, conventional breeding for nutritional improvement is often limited by the slow progress achievable over generations. Traits governed by multiple genes with small individual effects, such as nutrient content, can be difficult to improve. Moreover, conventional breeding frequently encounters trade-offs, as plants must balance resources allocated to growth, yield, and nutrient production. Environmental factors can further influence trait expression, leading to variable outcomes. Extensive screening is often required to identify desirable recombinations, adding to the time and labor demands of this approach. These limitations highlight the value of integrating modern techniques like marker-assisted selection and genetic engineering, which can target traits more precisely, accelerate breeding, and ultimately enhance nutritional quality while maintaining yield stability in vegetable crops.

### **3. Molecular Breeding and Biofortification**

#### *Biofortification of Vegetables*

Biofortification is a strategy aimed at enhancing the nutrient content of crops, providing a sustainable approach to combat malnutrition, especially in regions with high micronutrient deficiencies. This approach includes conventional breeding, agronomic methods, and genetic engineering, with the goal of enriching essential vitamins and minerals, such as iron, zinc, selenium, and provitamin A, in staple foods (Lal et al., 2020). For example, Singh et al. (2020) highlight biofortified vegetables with improved nutrient profiles to address "hidden hunger" and support immunity. This approach is crucial in developing countries, where vegetable intake often falls below the World Health Organization's (WHO) recommendation of 300g per person per day, and can provide essential nutrients alongside nutraceuticals like glucosinolates lycopene, and quercetin, which contribute to disease prevention (Parulekar et al., 2019). Common examples of biofortified crops include provitamin A-rich orange-fleshed sweet potatoes and high-iron beans, each tailored to local dietary needs and nutritional deficiencies (Petry et al., 2015).

#### *Marker-Assisted Selection (MAS)*

Marker-assisted selection (MAS) is a transformative tool in plant breeding, allowing for the precise selection of desired traits through molecular markers (Hatipoğlu and Şensoy, 2022; Uçar and Şensoy, 2022). This technique bypasses the limitations of conventional breeding by targeting specific genes linked to nutrient content and other beneficial traits. Molecular markers, including RFLPs,

RAPDs, AFLPs, SNPs, and microsatellites, identify genetic variations at a high resolution, enabling more efficient and accurate breeding for the studied traits (Inan et al., 2012; Yildiz et al., 2014; Iorizzo et al., 2016; Erdinc et al., 2013, 2017; Ekincialp and Sensoy, 2018; Park et al., 2000; Tiwari et al., 2022; Ibrahim et al., 2023). Unlike morphological markers, DNA-based markers are stable across environments, providing reliability in traits selection (Sensoy et al., 2007). Marker-assisted selection (MAS) has played a critical role in enhancing carotenoid levels in orange carrots (Iorizzo et al., 2016), where the identification of key genes such as *DCAR\_032551* has advanced our understanding of the genetic basis of carotenoid accumulation beyond biosynthetic pathways. Similarly, MAS has been instrumental in increasing lycopene content in tomatoes, aided by advances in sequencing technologies, SNP genotyping, and genome-wide association studies (GWAS), which have enabled precise identification of trait-linked genetic regions and SNP markers (Tiwari et al., 2022). Through this process, breeders can select for high-yield, nutrient-rich varieties by linking genetic loci with traits such as flavonoid and glucosinolate accumulation, offering substantial health benefits and improved crop quality.

### *Transgenic Approaches*

Genetic modification offers another pathway to enhance nutraceutical traits in vegetables (Koukounaras et al. 2022). Transgenic approaches involve introducing new genes or modifying existing ones to increase the content of health-promoting compounds or enhance nutrient availability. Examples in vegetable crops include the genetic modification of tomatoes to boost lycopene levels and the development of anthocyanin-rich purple carrots. These modifications allow for targeted enhancement of nutraceutical compounds like antioxidants, flavonoids, and polyphenols, which contribute to health benefits beyond basic nutrition. However, genetic modification in vegetables faces regulatory, public acceptance, and environmental challenges, which can limit its widespread application.

The convergence of biofortification, MAS, and transgenic methods creates powerful opportunities to address nutritional deficiencies. Biofortification through MAS and transgenic approaches holds promise for developing high-yield, nutrient-dense vegetable varieties with enhanced health benefits, supporting a shift toward nutritional security globally.

## 4. Genomic Approaches and Biotechnology Tools

### *High-Throughput Omics Technologies*

High-throughput omics technologies, encompassing genomics, transcriptomics, proteomics, and metabolomics, have emerged as powerful tools to enhance the understanding of complex genetic traits related to nutraceuticals in vegetables. Traditionally, improving the nutraceutical value of vegetables has been a slow process reliant on conventional breeding methods. However, advancements in omics enable precise identification and quantification of key bioactive compounds, such as phytoestrogens, carotenoids, and polyphenols, accelerating breeding processes and reducing costs. Karthiga et al. (2023) emphasize that the integration of omics allows the development of nutritionally superior vegetable cultivars with enhanced health benefits. Zhu et al. (2018) exemplify this potential in their study on tomato breeding, where a multi-omics approach involving hundreds of tomato genotypes revealed substantial alterations in metabolite profiles, linked to genes selected for traits like fruit size and color. The study highlighted that while certain genetic selections, such as pink fruit color, resulted in beneficial metabolites, others influenced anti-nutritional factors, showcasing the utility of omics in shaping the nutritional profiles of vegetables for improved health outcomes.

### *CRISPR/Cas and Other Gene Editing Tools*

Gene editing tools, particularly CRISPR/Cas systems, have revolutionized the field of plant biotechnology by enabling precise genetic modifications that enhance nutrient profiles and reduce antinutritional factors. Singh et al. (2021) illustrate the impact of CRISPR/Cas9 in elevating anthocyanin and lycopene levels in vegetables, which are critical for nutritional enhancement. Naeem et al. (2023) detail how CRISPR/Cas9 and CRISPR/Cas12a have been applied to manipulate carotenoid pathways in tomatoes, improving both aesthetic and nutritional qualities. This includes fine-tuning genes like *PSY1*, which is involved in the carotenoid biosynthesis pathway, to enhance pigments and secondary metabolite accumulation beneficial to human health. Such advancements not only boost nutritional value but also provide an edge for market acceptance through appealing color traits. The European Union's progressive regulation approach, based on the gene-editing process rather than the final product, highlights the growing support for CRISPR-based innovations in agriculture.

Recent work by Qian et al. (2022) demonstrates how CRISPR/Cas9 can target aroma-related genes, such as *GmBADH2* in vegetable soybeans, to generate aromatic cultivars by developing functional markers for these traits. The study confirmed that the aroma trait follows a recessive inheritance pattern, providing

a foundation for marker-assisted breeding of flavor-enhanced vegetable varieties. This example highlights CRISPR/Cas's capability to precisely modify crop traits to align with consumer preferences, offering significant potential for commercial applications in vegetable breeding.

#### *Transcriptomics and RNA Interference (RNAi)*

Transcriptomics and RNA interference (RNAi) provide essential insights into gene expression and regulation for nutraceutical-related genes, offering a fine-tuning mechanism to optimize the nutrient profiles of vegetables. By analyzing gene function and expression through RNAi and transcriptomic approaches, breeders can selectively silence genes that contribute to undesirable traits or enhance the expression of beneficial ones. Sun et al. (2024) highlight the integration of RNAi with QTL mapping and gene editing, enabling more targeted crop improvements that transcend traditional yield-focused goals to address specific nutraceutical outcomes.

Transgenic approaches also play a role in enhancing the nutraceutical content of vegetables. Koukounaras et al. (2022) achieved a significant increase in L-ascorbic acid (AsA) content in tomatoes by overexpressing *GGPI* and *GPP* genes. This gene manipulation not only tripled AsA content but also led to beneficial metabolic shifts, including reduced ethylene production and improved sugar and carotenoid levels. Such findings highlight the potential for transgenic techniques to elevate vegetable nutritional quality, though regulatory barriers persist in many regions. Nonetheless, advancements in transcriptomics, RNAi, and gene-editing technologies present promising avenues for improving vegetable crops with nutraceutical enhancements that align with modern dietary needs.

## **5. Genetic Resources and Germplasm Utilization**

### *Germplasm Evaluation: Importance in Nutrient Content and Resilience*

Genetic diversity is foundational for nutritional breeding, as it offers a spectrum of traits that enhance resilience, productivity, and nutrient density, essential for sustainable agriculture. The historic Irish Potato Famine, exacerbated by dependence on a genetically uniform crop, highlights the dangers of low diversity, which left potatoes highly susceptible to blight. Such events exemplify the critical role of genetic variation in safeguarding crops from pathogens, pests, and environmental stresses (Majeed et al., 2022). By preserving genetic diversity, breeders can develop crops with better nutritional profiles and resilience, reducing external input reliance and preventing major crop failures, contributing to global food security.

Genetic evaluations have proven invaluable in identifying bioactive and nutraceutical potentials within germplasm collections. For example, Siddiqui et al. (2014) explored bioactive properties in tomato hybrids containing the dark green (*dg*), old gold crimson (*og*), and ripening inhibitor (*rin*) genes. Their research compared hybrid tomatoes developed from Berika and BCT-115 (carrying the *dg* gene) to those from BCT-119 and BCT-111 (carrying *og* and *rin* genes, respectively). The findings revealed that the hybrids with the *dg* gene exhibited superior nutritional potential. These hybrids showed enhanced nutritional profiles with higher concentrations of bioactive compounds like ascorbic acid, lycopene,  $\beta$ -carotene, flavonoids, and phenols, suggesting potential applications in nutraceuticals and functional foods. Similarly, germplasm collections provide essential resources for breeding nutrient-rich vegetables. Erdinc et al. (2018) examined miniature tomato cultivars under various fertilization regimes, revealing cultivars like Black Zebra and Sweet Pea Currant with significant levels of bioactive components under combined organic and biochemical fertilization. This variability highlights the importance of germplasm in enhancing vegetable nutritional quality.

In addition, diverse germplasm is crucial for conserving wild species with high nutraceutical potential. For instance, Erdinc et al. (2021) analyzed the genetic diversity of wild rhubarb (*Rheum ribes L.*) using iPBS molecular markers. They identified high polymorphism levels, indicating substantial genetic diversity, which could benefit breeding and conservation efforts. Similarly, Cakmakci et al. (2021) studied the nutraceutical potential of *Allium vineale*, a wild species from Turkey, and found considerable variation in bioactive compounds among accessions, showcasing its value for phytonutrient enhancement.

Wild species in genetic resources can also offer unique antioxidants. Altıntaş et al. (2019) investigated wild asparagus species native to Türkiye, identifying species with high total phenolics, flavonoids, and antioxidant activity, with *Asparagus persicus* showing particularly strong results. These wild species provide a valuable genetic pool to improve cultivated asparagus' genetic diversity and could aid breeding efforts focused on enhancing bioactive compounds.

Germplasm evaluation is also essential for maintaining nutritional quality under stress conditions, which can influence bioactive component levels. Kipcak et al. (2019) studied salt stress effects on bean genotypes from the Lake Van Basin, revealing substantial differences in nutrient content and antioxidant capacity among genotypes. This variability highlights the potential of identifying salt-tolerant genotypes with retained nutritional quality for adverse conditions.

### *Breeding Programs and Varietal Development: Advancing Nutraceutical Quality*

Nutraceutical-focused breeding programs have led to significant progress in enhancing vegetable health benefits. For instance, Ranga et al. (2024) examined heterosis and combining ability in okra (*Abelmoschus esculentus* L.), identifying combinations with strong polyphenol content and yield improvements. Utilizing a line  $\times$  tester mating design, they revealed cross combinations with enhanced nutritional and medicinal properties, highlighting breeding's potential to produce resilient, high-yielding cultivars.

Non-transgenic approaches have also shown potential in enhancing flavonoid content in tomatoes, as shown by Willits et al. (2005). By reintroducing flavonoid biosynthetic pathways using wild *Solanum pennellii*, they achieved high flavonoid levels in both peel and flesh of cultivated tomatoes, demonstrating the value of wild genetic resources for nutrient enhancement without genetic modification.

## **6. Case Studies in Nutraceutical Breeding**

Nutraceutical breeding in vegetables has seen significant progress, driven by both traditional breeding and modern biotechnological innovations. Several case studies highlight the successful application of these approaches to enhance the nutritional and health-promoting qualities of crops, aiming to produce foods that are both nutritious and commercially viable.

- *Lycopene-Rich Tomatoes*

The development of tomatoes rich in lycopene, a powerful antioxidant, has been a crucial point of genetic improvement. Lycopene, found predominantly in red tomatoes, has been associated with numerous health benefits, including the reduction of cancer risks and heart disease. Recent advancements have focused on breeding varieties with higher lycopene content through both conventional and molecular techniques. Cammareri et al. (2024) reviewed the genetic and biotechnological strategies for improving anthocyanin content in eggplants and tomatoes, including the development of purple-fruited tomatoes, a newer innovation. Genetic studies revealed variability in anthocyanin profiles, which may not always correlate with visible traits. These findings open avenues for enhancing tomato lycopene content through both traditional breeding and cutting-edge genetic engineering tools, such as CRISPR-based gene editing, providing precise enhancements in both fruit nutritional quality and resilience to environmental stresses.

- *Glucosinolate-Enriched Brassicas*

Brassica crops such as broccoli and kale are valued for their high glucosinolate content, which contributes to their anticancer properties. Dong et al. (2019) conducted a comprehensive analysis of  $\beta$ -glucosidase genes in *Brassica rapa*, revealing insights into glucosinolate production. The study identified critical genes involved in flavonoid metabolism and glucosinolate regulation, opening new opportunities for breeding nutraceutical-rich Brassica cultivars. This research highlights the importance of genetic and genomic resources in developing Brassica crops with enhanced concentrations of health-promoting compounds like glucosinolates, which could have a profound impact on public health by improving the nutritional quality of widely consumed vegetables.

- *Genotypic and Environmental Effects on Chili Peppers*

Tripodi et al. (2019) explored how both genotypic and environmental factors influence the agronomic traits and bioactive compound profiles of hot pepper varieties, focusing on their antioxidant properties and capsaicin content. The study highlighted the complex genotype  $\times$  environment interactions that affect the pungency and antioxidant capacity of chili peppers. This research provides valuable insights for the breeding of chili peppers tailored to specific market needs, such as varieties with high antioxidant capacity or high capsaicin content for different culinary and medicinal purposes. It highlights the challenge of breeding for quality traits in crops with complex genetic-environment interactions, calling for precision breeding strategies that consider both genetic and environmental factors.

- *Genomic Variation in Cultivated Pumpkins*

Lee et al. (2023) analyzed the genomic variation within cultivated pumpkin species, focusing on seed traits, which are rich in protein, oil, and vitamins, contributing to their high nutraceutical value. The study mapped loci associated with seed traits using genome-wide association studies (GWAS) and identified genes regulating seed size. The findings provide a genetic framework for breeding pumpkins with enhanced seed characteristics, which are crucial for both nutritional content and commercial viability. These genomic insights will aid molecular marker-assisted selection (MAS) in accelerating the development of nutritionally improved pumpkin cultivars, ultimately enhancing the crop's contribution to human diets.

- *Polyacetylenes and Bitterness in Carrots*

Dunemann et al. (2022) investigated the genetic basis of polyacetylenes (PAs), which contribute to both the bitter flavor and potential health benefits in carrots. By identifying quantitative trait loci (QTLs) linked to PA levels, the researchers identified key genes involved in the biosynthesis of PAs, such as fatty acid desaturase 2 (*FAD2*) and ECERIFERUM1 (*CER1*). These findings provide a valuable resource for carrot breeding programs aimed at adjusting PA levels to enhance either the health benefits or consumer preference for milder-tasting carrots. Understanding the genetic mechanisms of PA production is essential for tailoring carrot cultivars with optimized bitterness and health-promoting properties.

- *Aroma in Sponge Gourd*

Chaubey et al. (2022) studied the inheritance of aroma in sponge gourd, a trait controlled by a recessive gene. This trait, which is linked to specific volatile compounds, can enhance the sensory and nutritional appeal of the vegetable. The study found that aroma production requires significant energy investment, which can reduce overall yield, a common challenge in breeding aromatic crops. However, the research opens possibilities for breeding sponge gourd cultivars with enhanced aroma profiles through genetic selection, ultimately contributing to the development of more flavorful and nutritionally valuable varieties.

These case studies exemplify the integration of genetic approaches, including genome-wide association studies (GWAS), gene editing, and traditional breeding, to enhance the nutraceutical potential of various vegetable crops. Each study highlights how understanding the genetic basis of key bioactive compounds - such as lycopene, glucosinolates, polyacetylenes, and aroma - can lead to the development of improved cultivars with better nutritional profiles, offering significant benefits for both human health and agricultural sustainability.

## **7. Future Directions and Challenges**

The advancement of genetic approaches and nutraceutical breeding in vegetables has the potential to significantly enhance nutritional quality. However, there are several future directions and challenges that need to be addressed for effective progress in this field. These include integrating genomic and phenotypic data, understanding consumer preferences and market acceptance, and navigating regulatory and ethical considerations.

### *Integrating Genomic and Phenotypic Data*

One of the primary challenges in modern breeding is the integration of diverse datasets to enhance the precision and efficiency of breeding programs. The combination of genomic, phenotypic, and multi-omics data is transforming the scope and scale of plant breeding, enabling the identification and selection of traits related to flavor and nutritional quality. As Ferrão et al. (2023) discuss, Artificial Intelligence (AI) plays a pivotal role in this integration by managing complex datasets, automating phenotypic and genotypic analyses, and advancing high-throughput phenotyping techniques. Through predictive models and data integration, AI allows breeders to simulate outcomes and optimize selection for crops with enhanced nutritional content. This process benefits from AI-driven tools that facilitate the methods such as High-Performance Liquid Chromatography (HPLC), aiding in nutrient profiling and streamlining the identification of beneficial compounds in vegetables. Furthermore, AI models that incorporate genomic, metabolomic, and transcriptomic data are crucial for addressing the intricate relationships between genetics, environment, and trait expression (Ferrão et al., 2023). These AI applications enable breeders to more accurately predict flavor profiles and target specific health-related metabolites or flavor-enhancing compounds, providing unprecedented opportunities to improve both the nutritional and sensory qualities of vegetables.

### *Consumer Preferences and Market Acceptance*

A significant challenge for the widespread adoption of genetically enhanced nutraceutical crops is consumer acceptance. Despite the potential benefits of biofortified and genetically modified vegetables, public perception remains a barrier to their market acceptance. Consumer resistance often stems from concerns about safety, environmental impact, and the ethical implications of genetic modifications. The success of nutraceutical breeding depends not only on scientific advancements but also on the willingness of consumers to accept and adopt these crops. Addressing these concerns through transparent communication, education, and demonstrating the tangible benefits of these crops - such as improved health outcomes - will be crucial in gaining broader market acceptance.

### *Regulatory and Ethical Considerations*

The regulatory landscape surrounding genetically enhanced nutraceutical crops is complex and varies across regions. In Europe, for example, recent developments in the regulatory framework, such as the European Court of Justice's ruling to regulate gene-edited crops based on the process rather than the

final product, have shifted how genetically modified organisms (GMOs) are evaluated. This decision allows crops created using gene-editing technologies to be assessed under existing GMO laws (Directive 2001/18/EC), potentially providing a more flexible and efficient regulatory pathway (Naeem et al., 2023). While these regulatory changes present opportunities for the adoption of gene-edited crops, further refinement of global regulatory frameworks is needed to facilitate the deployment of genetically enhanced nutraceutical crops. Additionally, ethical considerations regarding the modification of plant genomes, particularly in the context of public health, must be carefully evaluated to ensure that these crops are safe and beneficial for consumers.

In conclusion, the future of genetic approaches and nutraceutical breeding in vegetables is promising, yet it faces significant challenges related to data integration, consumer acceptance, and regulatory frameworks. Addressing these challenges through technological innovation, transparent communication, and regulatory reform will be crucial for advancing the development and adoption of genetically enhanced nutraceutical crops, ultimately improving global food security and health outcomes.

## **8. Conclusion**

### *Summary of Advances in Nutraceutical Breeding*

Genetic approaches in nutraceutical breeding have made significant strides in enhancing the nutritional quality of vegetables. Techniques such as traditional breeding, marker-assisted selection, and gene editing have been employed to increase concentrations of bioactive compounds, such as vitamins, antioxidants, and polyphenols. These advancements have the potential to improve human health by offering vegetables with enhanced nutritional profiles, promoting disease prevention, and combating malnutrition. Artificial Intelligence (AI) and multi-omics approaches have further enhanced breeding precision, enabling the identification of desirable traits more efficiently and accurately. AI has also supported the integration of genomic, phenotypic, and metabolomic data, helping breeders model flavor and nutrient profiles, which is crucial for consumer acceptance.

### *Future Prospects*

Looking ahead, several promising technologies and methodologies could further elevate the nutraceutical content in vegetables. The integration of AI with advanced genomic and phenotypic data, alongside multi-omics approaches, holds great potential for precision breeding, enabling breeders to target specific health-related metabolites and flavor-enhancing compounds. Gene-editing technologies,

such as CRISPR, are likely to play a larger role in creating crops with tailored nutritional profiles. However, the success of these advancements will depend on overcoming challenges related to regulatory frameworks, consumer acceptance, and ethical considerations. Moving forward, regulatory reforms that facilitate the adoption of genetically modified and gene-edited crops will be crucial. Furthermore, addressing consumer concerns about genetically modified organisms (GMOs) through education and transparent communication will be necessary for broader acceptance.

No matter how advanced and cutting-edge the technologies and methodologies employed, it is crucial to remember that plant breeding is not only a scientific discipline but also an art. The breeder's artistic touch, combined with the potential to create innovative products through the lengthy breeding process, is what ultimately enables success in the market. To thrive, a product must address the minimum needs of consumers, producers, and intermediaries, all within a delicate balance. The complexity of nutritional content, particularly nutraceutical properties, may present challenges, often requiring breeders to make tough decisions between yield and other traits. Striking the right balance between these factors is a key aspect of successful breeding, as it involves harmonizing both scientific rigor and the creativity necessary to meet diverse demands.

Overall, the continued development of nutraceutical crops through genetic innovation holds great promise for improving global health and ensuring food security.

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## CHAPTER 15

### Stem Cell-Based Seafood Production in Aquaculture

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## INTRODUCTION

World demand for fish, crustacean and mollusk products is the largest of all terrestrial animal food products and the aquaculture is the fastest growing sector in the global (Maulu et al. 2021). This sector supports 17% of the global demand for animal protein. Seafood, including freshwater, marine fish, crustaceans, and mollusks demand is predicted to increase 140 metric tons by 2050 due to the anticipated increase in the human population ( Costello et al. 2020). Recently, one of the greatest concerns of the world is to provide a healthy diet for the increasing human population, which, is expected to reach 9.1 billion people by 2050, and calculations show that 70% more food needs to be produced to feed the projected population (Doering and Sorensen, 2019).

As stated by the Food and Agriculture Organization of the United Nations (FAO, 2024), global fisheries and aquaculture production has extended to increase, reaching 223.2 million tons of which 130.9 million tons were produced in farms and ponds. Of these, 94.4 million tons were aquatic animals (fish, crustacean and mollusk). Moreover, fish, crustacean and mollusk production is predicted to expand by 10% by 2032 to reach 205 million tons. Although, universal marine and freshwater fish production has increased by 500% since the late 1980 (FAO, 2018), with the current state of fish farming and fishing productions, there is going to be a substantial gap between the supply and demand of seafood. However, the expansion of freshwater and marine fish farming and fishing industries to supply the demand for seafood is strongly linked to a number of environmental, public health and animal welfare issues (Ong et al. 2021). The increased demand for seafood elevates overfishing. In addition, freshwater, marine fish and crustacean farming have negative effects on aquatic ecosystem (Tripathi and Agarwal, 2023). There are some other restrictive barriers such as: global warming, threatened fish stock due to over fishing, environmental toxicity, competition with terrestrial animal production, fishmeal and oil shortage for use in fish feeds, parasitic worms in raw seafood. Thus, in the near future, fisheries and aquatic animal production will not be sustainable. These, global warming has the most devastating effects on global seafood production (Cheung et al., 2023). In order to calculate the effects of global warming on aquatic animal production Reverter et al. (2020) and Engelhard et al. (2022) have carried, some studies. However, Reid et al. (2019) and Naylor et al. (2021) stated that, it was extremely difficult to validate these measurements. The fatalistic effects are complex, some are direct, and others are indirect. Of these are, changes in the amount, duration, intensity, and seasonal distribution of rainfall, rising temperature forest loss due to droughts

and wildfire, acidification in the ocean (Elsheikh, 2021). While some fish farms in some regions may experience limited benefits from global temperature rising, overall world food productions are projected to decrease 10 per cent by 2050 (Barange et al. 2014). At 4 °C temperature rising, nutrient availability is predicted to decline by -30% by 2100 (Cheung et al. 2023) and the aquatic animal production's long-term sustainability is challenged by the consequences of global temperature rising, environmental toxicity, foodborne diseases and fishmeal and oil shortage.

In addition, the covid-19 pandemic spotlighted the threats to global food system security and the importance of sustainable and resilient animal protein production systems and suggested to use new technologies for sustainable global food security (Elias and Jambor, 2021). For sustainable freshwater, marine fish, crustaceans, and mollusks production, a freer pathway on novel technologies is necessary. Of these technologies, stem cell-based freshwater, marine fish, crustaceans, and mollusks meat production discovery could offer new means of sustainable aquatic animal production. In this technology, fresh meat cultivated from stem cells such as adult and pluripotent cells in sterilized laboratory conditions. This technique recommended as an innovative approach to enhancement the traditional marine, freshwater fish, crustaceans, and mollusks production. Thus, stem cell-based aquaculture technology proposed as a solution to complement sustainable seafood production (Farzad, 2021; Elias and Jambor, 2021).

The production of laboratory cultured terrestrial animal meat began with the attempt to culture terrestrial animal tissue through stem cell technology (Post, 2012; Post et al. 2020). Similarly, laboratory cultured freshwater, marine fish, crustacean and mollusk meats were mainly produced by stem cell technology using 3D bio printing, micro carriers, and scaffolds to form edible tissues (Lee et al. 2023). Thus, in this book chapter, history, classification of stem cells, basic methodology, commercialization of stem cell based seafood, challenges and acquiescence of this technology in the area of sustainable aquatic animal production will be updated.

### **History of Stem Cells**

Cohnheim et al. (1868) conducted first pioneering works in blood stem cell in 1868. Thereafter, German zoologists Theodor Boveri and Valentin Hacker used the term stem cell in 1892. Following these scientists, the key properties of a stem cell were first defined by McCulloch and Till in the Ontario cancer institute, at the university of Toronto (1963). They discovered the blood-

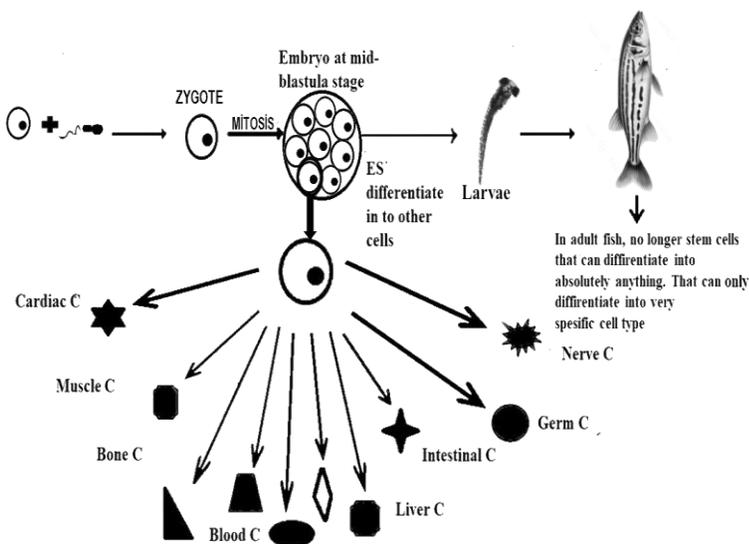
forming, hematopoietic stem cells in mice animal model. In the study, the authors injected bone marrow cells into irradiated mice and observed colonies of cells in the spleens of the mice that were linearly proportional to the number of the bone marrow cells injected. They concluded that each colony was a clone arising from a single marrow stem cell. In 1981, embryonic stem cells (ES) were first isolated and cultured in mouse (Evans and Kaufman, 1981). Following years, human embryonic stem cells were first isolated and cultured by American scientists James Thomson (1998). However, the origin of stem cell-based meat production dates back to 1930 when a British writer Frederick Edwin Smith first proposed the idea. Following years Winston Churchill, who was a close friend of Frederick Edwin Smith has, appropriated his ideas in 1932 by writing about how one could grow chicken breast from living cells instead of farming chickens. Churchill's words are widely quoted whereas Smith's idea have been largely forgotten (Ford, 2011). Nevertheless, stem cell based meat production remained only an idea for over 40 years due to the technical limitations of the time (Ye et al. 2022). In 1971, Ross (1971) investigated the growth of smooth muscle and the formation of elastic fibers cultured *in vitro* using smooth muscle obtained from the inner media of guinea pig aorta.

Research on fish stem cells started 60 years ago in 1961. Kirsche and Kirsche (1961) noted that the optic tetum of crucian carp (*Carassius carassius*) could regenerate by producing of new cells. These cells were involved in the regeneration (Kawakami, 2010). Kimmel and Varga (1986) have identified that, cells at mid-blastula stage are not differentiated yet and have the power to differentiate into embryonic stem cells (ES). Following Collodi et al. (1992) established methods for the culture of cells derived from mid-blastula stage zebrafish embryo. Thereafter, cell cultures derived from early zebrafish embryos differentiated *in vitro* into neurons and astrocytes in zebrafish (Gosh et al. 1997). Hong et al. (2000) reviewed status and perspectives of fish embryonic stem cells. Benjaminson et al. (2002) successfully cultured dorsal abdominal skeletal muscle mass tissue from common *Carassius carassius*, goldfish in petri dishes for Space travelers. Their goal was to establish the feasibility of an *in vitro* muscle protein production system. The authors pointed the way to an innovative, viable means of supplying safe, healthy, and nutritious food to Space voyagers on long journeys. That study was the pioneering study of today's stem cell based seafood production technology.

## **Classification of Stem cells**

Stem cells are a class of undifferentiated cells from which all other cells with specialized functions are generated. Under optimum conditions in the microenvironment of body and /or a Petri dish, these cells have the power to reproduce themselves by mitotic cell division. These cells become either new stem cells or differentiate into specialized cells with a more specific function, such as blood, brain, heart, muscle, bone cells, spermatogonia and /or oogonia (Çek, 2006; Çek-Yalnız and Yaraş, 2019; Yaraş and Çek-Yalnız, 2021). In the human and animal body, no other cells have the ability to generate new cell types. The behavior of these cells is regulated by the microenvironment and they persist for a long time (Figure 1).

Research on stem cells, increased gradually in all countries in the world. Publications of manuscripts in this field are also steadily increasing in the last 40 years. Since it was first defined in 1963, substantial amount of academic research has been performed on stem cells. In Google Scholar 4240000 paper were indexed to the term stem cell. The numbers of these studies are increasing rapidly. When these publications observed there is no world widely accepted classification of these cells among scientists. However, there are two defining quality of a stem cell. The first one is self-renewal by mitotic division and the second one is the power to differentiate into a specialized adult cell type. These cells are classified based on their source and their power to differentiate into as many as other cells in the body of an organism. As a whole, these cells are divided into three types. This division is based on their origin (Çek et al. 2016). These are embryonic stem cells, Adult stem cells and Germ stem cells. Stem cells are also can be divided into 6 stages based on their differentiation power (Figure 1). These are totipotent, pluripotent, multipotent, oligopotent, unipotent and nilpotent. A fertilized egg cell or zygote has the genetic information to generate a whole organism is defined as totipotent. Collodi et al (1992) derive first totipotent stem cells in fish.



**FIGURE 1:** Classification of Fish embryonic stem cells, which have the power to create an adult fish. Following fertilization, cells divide and increase in number. Subsequently, creating an embryo at mid-blastula stage, haploid cell in the embryo at mid-blastula stage has the ability to produce an adult fish. ES are able to differentiate into other body cell types. Updated based on Çek-Yalınz and Aydın, 2023.

A pluripotent stem cell has the power to differentiate into many cells in the fish body, however has no potency to create a whole organism. Multipotent stem cells can be differentiated into less number of body cells than pluripotent stem cells. E.g. mesenchymal or fibro-adipogenic stem cells which are finally differentiate into muscle, cartilage, lipid cells. Oligo potent stem cells can differentiate into less number of cells than the multipotent stem cells. E.g. hematopoietic stem cells. Unipotent cells are those with the least ability to differentiate. Such as adult muscle, stem cells like satellite cells and myoblasts (Post 2020). These cells from animal tissues are easy to obtain and can differentiate into the necessary mature cell types existing in meat and seafood, but their proliferative ability or maintenance in vitro is not yet sufficient.

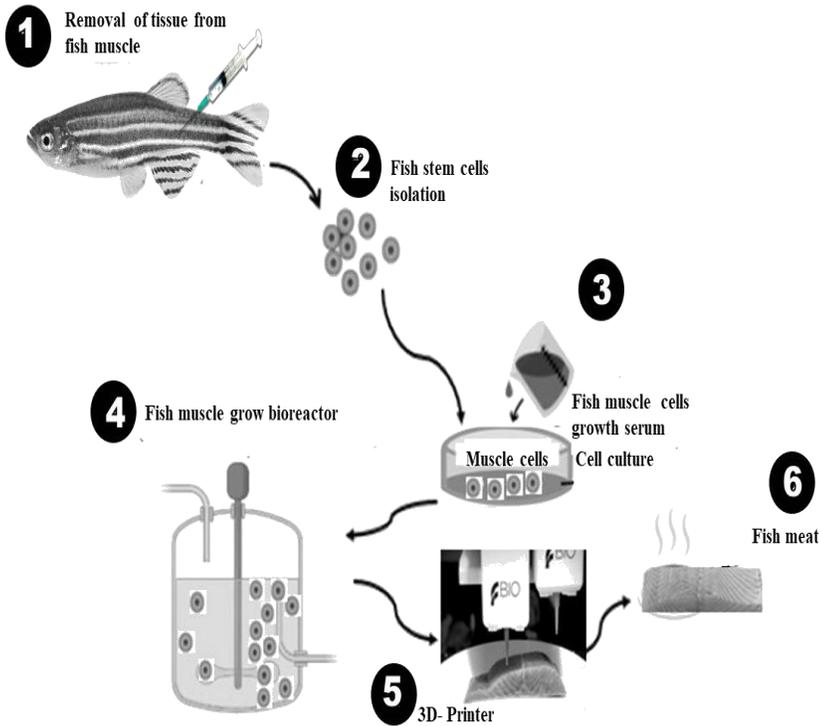
Musgrove et al (2024) did a comprehensive review on the crustacean stem cell based seafood production. The authors indicated the lack of cell lines in stem cell based crustacean meat production. They highlighted three potential non-lethal primary stem cell resources that could be utilized in stem cell based crustacean meat production. Those were indicated as muscle tissue within fully-grown crustacean limbs such as claw or walking legs; tissues within actively

regenerating limbs and finally; hematopoietic stem cells circulating within the hemolymph. Selecting the best starting stem cell types for cell-based seafood production is one of the most important issues. It seems that embryonic stem cells (ESCs) and induced pluripotent stem cells (iPSCs) are the best candidates for the best starting to cultivate stem cell-based seafood. Bomkamp et al. (2023) reviewed advantages and disadvantages of the different cell types considered for stem cell based seafood production, most of which were of mesodermal origin. The authors indicated that, the most critical cells for the final product were muscle and fat cells, the precursors to these cell types; mesenchymal stem cells, satellite cells, fibro-adipogenic progenitors, and preadipocytes were the most likely candidates among the adult stem cells to be used for stem cell based seafood production. Muscle fibers and adipocytes cell lines were developed for Yellow croaker fish species (Xu et al. 2023). Stem cell-based seafood production is less developed than stem cell based mammalian meat production. Mechanism and signaling pathways involved in the formation of muscle, fat and other relevant tissue are relatively well investigated in the Zebrafish. ESCs and iPSCs lines are best defined in this small aquarium species. This same process is not well established in other fish species, crustacean and mollusk (Bomkamp et al. 2023). Among the cell types used for the stem cell based seafood production, muscle cells have been reported more frequently compared to lipid and fibroblast cells.

### **Methodology of stem cell based seafood production**

Chandrababu and Puthumana, (2024), reviewed basic methodology of stem cell based seafood production. This technology involves culturing cells from fish, mollusks and crustaceans, rather than catching or farming fish, mollusks and crustaceans. Stem cell- based seafood is produced by cultivating isolated cells, such as muscle, lipid and fibroblast cells, derived from living aquatic animals (Yang et al. 2024). These isolated muscle cells are then cultured under sterile and optimal conditions to encourage growth and differentiation. Subsequently, a three-dimensional tissue culture is conducted using scaffolds or micro-carriers to develop fish, mollusks and crustaceans meat. The product of this process is termed as cell based cultured fish or cell- cultured seafood. In another words, the production of stem cell-based freshwater, marine fish, mollusks and crustaceans meat have to be extracted from fish, mollusks and crustaceans, muscle cells, followed by their isolation and culture in optimum environment inside a bioreactor. Muscle and lipid stem cells from fish, crustacean and mollusk are grown on an edible scaffold that is designed to give

them the structure and texture of wild caught meat (Figure 2, Table 2). This laboratory-cultured seafood should be indistinguishable from wild-caught fish, mollusk and crustacean meat. An edible scaffold is conducted to give the cells the structure and texture of wild-caught and hatchery cultured freshwater and marine fish meat. The cells grow on the scaffold. Mean expectation is that seafood produced by stem cell culture technology is indistinctive from conventional seafood which wild caught and hatchery cultured meat.



**FIGURE 2:** Demonstration of stem cell-based freshwater, marine and/or ornamental fish meat production. Modified from, Çek-Yalnız and Aydın, 2023.

**TABLE 2:** Basic methodology of stem cell based seafood production. Cell isolation, culture, differentiation and collecting the final seafood are the basic steps.

Key Steps	Step 1 Cell Isolation	Step 2 Cell Culture	Step 3 Cell Differentiation and Maturation	Step 4 3D tissue culture and collecting the final product
1	Anesthetization of fish, mollusk and crustacean	Prepare stock solution of culture medium. (Use DMEM and/or L-15, and/or AIM-V)	Transfer cells into a growth tank (cultivator)	Maintained cell viability, proliferation, differentiation and maturation
2	Disinfection of fish, mollusk and crustacean with 70% ethanol Sterilization of all equipment (dissection set, lids, tubes, glass bottles etc.) This equipment's are kept in a continuous burner to prevent possible contamination.	Prepare flask collagen coated	Enrich culture medium with differentiation agent	Promote cell fusion (Scaffolds and Microcarriers+volume addition)
3	Biopsy of muscle/ lipid/ fibroblast cells	Add, growth factors, HEPES, antibiotics, nutrients, Insulin, Fish serum,	Add serum, extracellular matrix,	Hydrogel Crosslinked
4	Separation of these cells from other cells like blood cells and tissues (Use mechanical, enzymatic and explant)	Maintain pH at certain degree Maintain temperature at certain degree Maintain osmolality at certain degree	Add a network of proteins and other molecules	Collect the edible muscles
5	The tissue samples were chopped into very small pieces and for proper enzymatic digestion and maximum dispersion of the tissues, Trypsin-EDTA (ethylene diamine tetra acetic acid), or collagenase type XI depending on concentration and duration of digestion.	Check the cells daily, under an inverted microscope	TNF- $\alpha$ , LIF, IGF1, IGF2,FGF, HGF, GSK3b Cytokines	Remove off the non-edible scaffolds.
6	Wash cells with Hang 'Buffer Solution and /or PBS	Dilute the cells daily	Specialized technique, such as gene transfer may require in some cases.	Edible muscles are stored and ready for distribution
7	Prevent contamination in each stage	Feed the cells (cell feed contains water, oxygen, nutrients and growth factors)		Serve as fresh seafood or process further
8	Terminate the digestion process, add DMEM and/or L15 supplemented with 10% Fetal Bovine Serum and/or fish serum			
9	After digestion, avoid contamination, For example use antibiotics and antifungal reagent namely, penicillin-streptomycin, tetracycline			
10	Aspirate and filtered out the cells with a 40 $\mu$ m cell strainer			
11	Centrifugated the cells			

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12	After centrifugation count the cells with a hemocytometer and/or automated cell counter
13	Separation of Satellite cells (Using magnetic beads, cell sorting, Gradient centrifuge with percoll, ficoll, histopaque).

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Marine and freshwater stem cells may be advantageous compared to terrestrial species. These cells require culturing at lower temperatures (19-30°C), they have a broader pH tolerance (6.8-7.6) and do not require CO<sub>2</sub>, taking all together could translate to lower manufacturing energy consumption (Shang et al. 2015; 2018; Çek-Yalınz and Yaraş 2019; Yaraş and Çek Yalınz, 2021; Musgrove et al. 2024). In addition, most fish species and crustacean display continual growth throughout life have impressive regeneration abilities and high telomerase expression, they may have potential for greater rates of cellular proliferation and differentiation (Specht et al. 2021; Musgrove et al. 2024).

Recently Chan et al. (2024) have made a comprehensive review of stem cell based seafood production methodology and concluded that the isolation of mesenchymal stem cell was more straightforward approach than other stem cell sources for stem cell based seafood production. Isolation of stem cell niches for crustaceans and mollusks and optimization of culture conditions remains a challenge. Serum free media development involves resource-intensive profiling of serum replacements, and determination of optimal concentrations, but may yield cost efficiencies in large-scale stem cell based seafood production. Tackling these challenges is instrumental to the long-term sustainability of stem cell based seafood production.

### **Commercialization of stem cell based seafood**

Stem cell-based hamburger was first tasted in 2013. Since then, researches on stem cell-based meat have increased rapidly. However, culturing seafood from aquatic organism's stem cells has received very little attention all over the world (Rubio et al. 2019). Most effective projects on stem cell-based seafood production are ongoing in Singapore. Countries worldwide, including UK, USA, Netherlands, China, Portugal, Canada Hong Kong, Israel are actively involved in the production of stem cell-based seafood. More than 200 companies are competing to prepare for marketing stem cell-based terrestrial and aquatic animal meat production in the world. Of these 61 are attempting to cultivate cell-based freshwater, marine fish, crustacean and mollusk meat production (TABLE 2).

**TABLE 2** List of enterprise working on cell-based freshwater, marine fish, crustacean and mollusk meat production. Updated based on Çek-Yalınz and Aydın, 2023

No	Enterprise Name	Location	Seafood Type	Date Estab lished
1	Thai Union Group	Thailand	Tuna and mahi-mahi	1977
2	ArtMeat	Russia	Sturgeon	2015
3	Memphis Meats (UPSIDE Foods)	USA	Sea foods	2015
4	Atlantic Fish Company	USA	Cultivated Seafood's	2021
5	Finless Foods	USA	Bluefin tuna and carp	2016
6	WildType	USA	Salmon	2016
7	BlueNalu	USA	Mahi-mahi and Bluefin tuna, to start.	2017
8	SeaFuture	Canada	Fish	2017
9	Biftek.co	Türkiye (The company located in USA)	Currently only beef, no seafood Founder: Can Akçalı and Erdem Erikçi, Kerem Erikçi	2018
10	Avant Meats	Hong Kong	fish maw and undisclosed fish fillets	2018
11	Shiok Meats	Singapore	crustaceans- shrimp, crab and lobster have already been unveiled	2018
12	Umami Bioworks	Singapore	Seafood	2018
13	Magic Caviar	Netherlands	Caviar and oocytes (world's first)	2019
14	Cell MEAT	South Korea	Shrimp, lobster and other high-value seafood varieties.	2019
15	Cell Ag Tech	Canada	White fish	2019
16	Sophie's Bionutrients	The Netherlands (Company located in Taiwan)	Crabmeat	2019
17	SoundEats	USA	White Fish	2019
18	CellulaREvolution	United Kingdom	Improving bioprocessing technology in seafood and meat	2019
19	LiveMatrix Biotech	USA	Seafood, meat, ingredients	2019
20	Sustineri Piscis	Brazilia	Seafood	2020
21	Umami Meats	Singapore	Exotic, crab, shrimp	2020
22	CellX	China	Seafood1/3 world 'demand	2020
23	Cultured Decadence	USA	Crustacea	2020
24	Bluu Biosciences	Germany	Salmon, trout and Carp	2020
25	Unicorn Biotechnologies	United Kingdom	Standardize, digitize and automate stem cell culture workflows	2020

26	Another Fish	Canada	Fish	2021
27	Bluefin Foods	USA	Bluefin tuna	2021
28	Fisheroo	Singapore	Fish	2021
29	SeaWith	South Korean	Fish	2021
30	Sea-Stematic	South Africa	Sea foods	2021
31	Sea2Cell	Israel	Seafoods	2021
32	Ambrosia Sciences	Singapore	Seafood	2021
33	Cellqua	South Korea	Seafood	2021
34	Meatosis	Israel	Seafood	2021
35	Mermade Seafoods	Israel	Seafood	2021
36	Simple Planet	South Korea	Seafood	2021
37	E-FISHient Protein	Israel	Fish (Particularly Tilapia)	2021
38	Wanda Fish Technology	Israel	Seafood	2021
39	Forsea Foods	Israel	Freshwater Eel (Unagi)	2021
40	Pearlita Foods	USA	Oyster and mollusk	2022
41	Extracellular	United Kingdom	Developing media, cells and scaffolds for seafood and meat production	2022
42	Pacifico Biolabs	Germany	Seafood particularly white fish	2022
43	Cell Agritech Snd Bhd	Malaysia	seafood	2022
44	Cell4food	Portugal	seafood	2022
45	Cell Tec Systems GMBT	Germany	Seafood, meat	2022
46	Noubio	USA	Seafood, meat and dairy, serum replacement	2022
47	Reel Foods	USA	Seafood	2022
48	Sound Eats	USA	Seafood	2022
49	Fishway BV	Belgian	Seafood	2022
50	ImpacFat	Singapore	Seafood and meat	2022
51	Klever Meat	India' first	Seafood	2022
52	Meatosys	Germany	Seafood, meat	2022
53	MyriaMeat	Germany	Seafood , meat	2022
54	NeatMeatt (Pvt Ltd)	India	Seafood , meat, ingredients	2022
55	Upstream Foods	Netherlands	Seafood	2022
56	Arta Bioanalytics	Australia	Eggs, Seafood	2023
57	Biokraft Foods	India	Seafood	2023
58	BOBFoodTech	Irael	Seafood	2023
59	Marinas Bio	USA	Seafood	2023
60	Nuna	Israel	Seafood	2023
61	Deco Labs	USA	Seafood meat	2024

Singapore is the first country in the world to allow stem cell based meat grown in laboratories to be sold and eaten by public. Currently one restaurant

serving stem cell-based meat is operating in this country. After Singapore's approval in December 2020, the company, 'Eat Just' has succeeded in to get its stem cell based produced chicken meat for public sell in Singapore (Ye et al., 2022). The company claimed that the meat was ethical, clean, and green and the texture and taste was indiscernible from farm-produced meat. Following this announce, dozens of local and international companies have initiated operations in an effort to bring these products to market and currently there are 13 stem cell-based meat production companies in Singapore (Stevens and Ruperti, 2024). However, Singapore is a very small island nation and it lacks natural farmland and resources. Its 'land area is 720 square kilometers and holding a population of 5.9 million. Less than one percent of Singapore's land is currently used for agricultural food production (Stevens and Ruperti, 2024). Consequently, it imports more than 90% of its meat from more than 170 countries and regions. The country wants to change the situation and aimed at producing stem cell-based meat and seafood developing stem cell technologies (Matwick, 2024). Countries like Türkiye, which has large natural farmland and seawater resources, may not need cell-based meat and seafood production.

America is the second country approving meat grown in laboratories as safe consumption and sale. The Israeli Ministry of Health announced its regulatory approval for stem cell-based meat production on January 2024 as well. It seems that, the number of countries, which are allowing stem cell-based meat and seafood, are increasing rapidly in the world.

Chandimali et al. (2024), made a comprehensive review on seafood commercialization and concluded, the need for these stem cell based seafood without relying solely on conventional seafood. The most attractive motivation of consuming stem cell-based seafood includes good color, smell, flavor and texture (Chan et al., 2024). Stem cell-based freshwater, marine fish, crustacean and mollusk meat have to be better than caught and farmed fish, crustacean and mollusk meat. Currently, its color, texture, taste, smell has not met yet the accepted qualities. Its price, approval and availability are other challenges. Wang et al. (2023) reviewed three-Dimensional Scaffolds in laboratory-grown seafood. The authors concluded that, laboratory-grown seafood could not completely replace aquaculture.

### **Challenges of stem cell-based seafood production**

There are some challenges of stem cell based fish (freshwater and marine), crustacean and mollusk meat production. These challenges includes: developing

the new product and improving the production methodology, introducing to market, making it cost competitive and getting consumers to willingly substitute it for wild-caught fish, crustacean and mollusk

Fish stem cells may be advantageous compared to terrestrial species. In culture conditions, requiring lower temperatures, having a broader pH tolerance and not needing CO<sub>2</sub> may be good qualities for these cells (Shang et al. 2015; 2018; Çek-Yalınz and Yaraş 2019; Yaraş and Çek Yalınz, 2021; Musgrove et al. 2024). Nevertheless, currently there is no internationally accepted methodology for each species cell lines and the future of laboratory grown seafood depends on the evolution and depiction of appropriate fish, crustacean and mollusk muscle cell lines from commercially important and highest income-generating aquatic animal species, (such as; shrimps, yellow tail, crayfish, oyster). There are some basic challenges that need to be overcome in next decade. These are optimization of cell culture micro environmental conditions, universally available serum free culture media, and mass production of cells in bioreactors.

Its price, approval and availability are other challenges. The price of a kilogram of shrimp meat from a company in Singapore is around 50\$. Stem cell-based meat is still not close to competing with meat from caught and farmed fish meat. When it comes to the approval of stem cell-based meat, countries in Asia are far more open to the technology than European countries. Despite building some regulatory approval on stem cell-based aquatic animal meat production, there is no systematic approach for developing a comprehensive safety plan for laboratory-cultured seafood. Valuable detailed information regarding to safety plan can be found in Ovissipour et al. (2024).

Laboratory cultured, fish, crustacean and mollusk meat production technology has been proposed as a solution to complement environmental conservation (Rubio et al. 2019; Chandimali et al. 2024). In a more realistic way, the links between cell-based fish (freshwater and marine), crustacean and mollusk meat production and environmental conservation may be weaker than they first appear. Because, stem cell-based seafood production has to be in large quantities so that fishing declines, resulting in threatened stock recovery. However, currently this issue has not been solved yet.

One of the biggest future challenges is to get the consumers to make the switch to cell-based fish, crustacean and mollusk, and making them believing that they translates to reduce fishing of threatened species. Stem cell-based freshwater and marine meats have to be better than caught and farmed fish meat. Currently, its color, texture, taste, smell has not met yet the accepted

qualities. Religious consumers might even reject the idea of alternating stem cell based meat production. Its production must be kosher for Jewish consumers, halal for Muslim consumers and must be defined as edible based on Holy Bible for Christian consumers (Benny et al. 2022). In a word, it is obvious that consumer prejudice is a difficult problem that needs to be overcome.

Nevertheless, laboratory-grown seafood cannot completely replace aquaculture. Because of unavailable fish crustacean and mollusk muscle cell lines to produce stem cell, based seafood and currently high production price which is not affordable and finally incomplete methodology for production of stem cell based seafood. The trails, for stem cell-based fish, crustacean and mollusk production to have a benefit to fisheries and aquaculture is difficult, long and narrow way to go

### **Acquiescence of stem cell-based seafood production**

Cell based seafood is declared to be free from antibiotic residues, zoonotic bacteria, viruses and other contaminants associated with cultured aquatic species (Rao et al., 2024). This claim is not always the case. During the culturing of stem cells contamination is sometimes a problem and antibiotic use is inevitable (personal observation). Thus, stem cell-based seafood is not completely antibiotics free. However, it may not contain zoonotic bacteria or viruses. A parasitic worm in raw seafood, which has been reported to increase 283-fold in the past 40 years is not exist in stem cell based seafood production (Fiorenza et al. 2020).

Stem cell based seafood production claimed to be produced at any location on the earth. This claim is also may not be the case. Because, there are many countries, which rejects the production of stem cell-based seafood due to religious beliefs. Nevertheless, cell-based seafood can be produced throughout the year regardless of season (Rao et al., 2024). If the methodology developed, it can be produced from any aquatic animal species including threatened/endangered and even from ornamental fish species.

Currently, it is still difficult to farm most of deep-sea species. Deep-sea species are also difficult to catch. In addition, it needs significant land area and financial investment to farming; therefore stem cell based seafood production may be beneficial (Tsuruwaka and Shimada, 2022). Moreover, some cannibalistic shrimps, crabs and lobster species are extremely difficult to farm and stem cell based seafood production may reduce cycle times and shorten the production duration and cannibalism is out of the question. It may take stem cell-based seafood cultures weeks to months to produce edible seafood.

Whereas, it takes years to farm fish, crustacean and mollusk. For example, Bluefin tuna reach sexual maturity at about 5-10 years; it takes at least one year to bring it to the market. Regular salmon reach market size at 18 months (Waltz, 2017; 2020). Lobster reach sexual maturity at five to eight years (Bianchini et al. 1998) and freshwater crayfish reach sexual maturity at least in tree year (Aydın, 2010). Stem cell based seafood production may reduce the duration of marketable seafood.

Maturation of fish (Moblely et al. 2021), crustacean (Mesquita et al. 2020) and mollusk (Cerros-Cornelio et al. 2023) is also differs between the sexes. One sex mature earlier than the other sex thus, artificial production in hatcheries and production in every day of the year may not be possible whereas in stem cell based seafood production this problem is not an issue.

Other claim is that stem cell based seafood production minimizes the production area of seafood. This claim may have the right reason since stem cell based seafood production can be made in a lab and does not require large breeding ponds or fish farms. Countries like Singapore which has hardly any of its own farmland and natural resources, may benefit from stem cell based meat and seafood production.

Stem cell-based seafood declared to be identical to the muscle cells that are present in conventional farmed seafood. Currently, it has not yet identical to farmed and caught fish meat. Taste and texture of stem cell-based seafood is also needs to be improved.

The presence of micro plastics in mollusk is an important issue (Gamarro et al. 2020) and this issue does not exist in stem-cell based seafood production. Another food safety benefit of stem-cell based seafood production is claiming to be the lack of heavy metals (Chandimali et al. 2014).

The aquaculture of crustacean, freshwater and marine fish was found to emit more carbon emissions than the cultivation of poultry. Therefore cell-based seafood production is defined as an eco-friendly process compared to conventional farmed seafood in terms of greenhouse emissions, land and water use (Marhawa et al. 2022).

Vegetarians who refrain from eating meat out of concern for the well-being of fish and the climate crisis are may interested in stem cell-grown meat (In cruelty-free meat). However, there is no studies on the vegetarians 'will to consume stem cell based produced fish meat.

The conversion of mangroves to shrimp farm has resulted in the loss of approximately 1.5 million hectares of mangroves since 1980 (Eid et al. 2019).

The study has shown that stem cell based seafood production use considerably less land, water, and energy than traditional production.

## **Conclusion**

The consumption of freshwater, marine fish, mollusk and crustacean are rising, as the global population. Thus, stem cell based fish, crustacean and mollusk meat production is a novel technology for increasing aquaculture production. Challenges and acquiescence of this novel technology are summarized above. There are some basic challenges in the methodology, application, and acceptance of this technology. Such as; the difficulties of application methods, opportunity of fish, crustacean and mollusk muscle cell line to produce laboratory-grown seafood, and consumer resistance to consume seafood produced by this technology. Once these problems are resolved, the future of this technology will be bright. This technology has the principal goal of cultivating fish, crustacean and mollusk meat in the laboratories for human consumption and the number of companies developing cell based aquatic animal meat production are increasing rapidly. In 2023, the number of these companies was 30. This number increased and reached 61 in just one year. It seems that this technology can make the aquaculture production sustainable. It may even contribute the decreasing of the global temperature, in terms of greenhouse emissions, replenish fish stock due to over-fishing, ensuring safe food, which are free from foodborne diseases, plastics, antibiotic residues, heavy metals, zoonotic bacteria, parasitic worms in raw seafood. The potential economic benefits of this technology to aquaculture are obvious and in the next decade, seafood produced by this technology will be on the sell in many countries.

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