ISSN: 2582-6980

AGRI MIRROR
FUTURE INDIA

AIASA Agriculture Magazine

A Voice for Agriculture

ISSUE 06/2023

A Voice for Agriculture

AGRI MIRROR : FUTURE INDIA

Issue 06 | 2023

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INLAND SALINE AQUACULTURE – A NOVEL FOOD PRODUCTION SYSTEM

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Abstract

In many locations of the world, particularly semi-arid and arid zones, wherever the rainfall is low and evaporation is high, saline-affected areas are common. Inland saline waters have a negative impact on the environment and agricultural productivity in many nations, including India. These lands are unfit for either farming or human consumption. Therefore, the saline affected inland areas with saline groundwater can be utilized for aquaculture practices. Inland saline aquaculture is an innovative food production technique needed to increase aquaculture productivity and satisfy the rising seafood demand.

Introduction:

Salt-affected lands cover more than 1000 million hectares worldwide, a major barrier to sustainable agricultural production (Wicke *et al*., 2011). These lands consist of saline and sodic soils, which are primarily generated naturally. However, secondary salinity, which occurs due to rising water tables mainly caused by human activities, including intensive irrigation, affects over 380 million hectares of land in over 20 countries throughout the globe (Ghassemi *et al*.,1995). Many semi-arid and dry parts of the world are affected due to soil salinity, and it is a serious environmental issue that affects freshwater and soil (Munns, 2005). The salt-affected inland areas contain a significant amount of surface and subsurface saline water. Due to the excessive soluble salts in the water, the accessibility of water present in the soil to the crops is highly affected, which affects the productive lands used for agriculture and groundwater used for the irrigation of plants. Consequently, the existence of rural communities, agricultural productivity, food security, economic returns to the country, and the value of agricultural exports will all be significantly impacted (Doupe *et al*., 2003). Using saline groundwater for aquaculture is a fruitful option for salt-affected agricultural lands. Aquaculture conducted on land using saline groundwater is referred to as "inland saline aquaculture". It involves raising various euryhaline fish and shellfish species on saline-affected areas and widely used in many countries, including India (Allan *et al*., 2009).

Saline soil:

The word "saline soil" refers to soils with a significant concentration of soluble salts, such as sodium, magnesium, calcium, sulphates, and chlorides. These salts impact the extent to which the plants can absorb water from the soil. Salinity is typically expressed in terms of electrical conductivity (ECe), which is measured in deci siemens per meter (dS/m) (Chhabra *et al*., 2004).

Table 1. Features of saline soil (Singh *et al*., 2014)

Electrical conductivity	**> 4 ds/m**
Exchangeable sodium percentage	< 15%
pH	< 8.5

Inland saline water:

The salinity (> 0.5 to 165 ppt) and ionic content of inland saline water vary from region to another. According to Jana *et al*. (2004), inland water has a salinity range of 10 to 25 ppt and a high concentration of divalent cations such as calcium and magnesium, which increases the hardness of the water. Problems associated with the usage of open seawater for aquaculture purposes, such as the presence of microorganisms, entry of unwanted organisms living in seawater into the culture system, requirement of expensive filtration units, and other fluctuations in temperature, can be resolved by using saline groundwater for aquaculture practices (Jana et al., 2004).

Fig 1. Salt-affected inland areas in Punjab (Ansal and Singh, 2019)

Fig 2. Inland saline aquaculture demonstrated by local farmers in Punjab (Ansal and Singh, 2019)

Saline-affected areas in India:

It is estimated that around 8 million hectares of salt-affected lands exist in India, with most of these lands being in the northern states of India, such as Gujarat, Haryana, Maharashtra, Punjab, Rajasthan, and Uttar Pradesh. The states of Karnataka, Tamil Nadu, and Andhra Pradesh are among those in south India that are afflicted by soil salinity (Lakra *et al*., 2014).

Causes for the formation of saline soils:

Inland saline soil formation is mainly a result of natural processes, including decreased rainfall and rock erosion, and secondary salination, which is induced by human activities. The excessive use of chemical fertilizers, excessive irrigation, and inadequate drainage cause secondary salination, which raises the salinity of the soil and ground water in inland areas.

A novel perspective about inland saline aquaculture:

The most rapidly expanding food production system in the world is aquaculture. The production of fish worldwide was predicted to be 178 million tonnes in 2020. 49% of the overall production came from aquaculture (SOFIA, 2022). However, the widespread adoption of aquaculture has frequently been associated with several adverse environmental outcomes. The growth of inland saline aquaculture is essential for halting the further destruction of agricultural areas and water bodies by traditional inland aquaculture practices. Inland saline aquaculture have the capability of increasing aquaculture productivity.

Candidate species for aquaculture in saline-affected areas:

It is crucial to evaluate the survivability, growth, feed intake, and disease occurrence of any species before introducing them into the culture ponds developed in inland saline affected areas. Compared to seawater, inland saline groundwater might have different ionic compositions and adjusting the ionic composition and selecting suitable species for aquaculture is one of the main issues for the development of inland saline aquaculture.

Table 2: Candidate species for inland saline aquaculture

Species	Findings	Reference
Common carp (*Cyprinus carpio*)	Inland saline groundwater with a salinity up to 5ppt can be utilized for the culture of Common carp.	(Iffat *et al*., 2020)
Silver perch (*Bidyanus bidyanus*)	The feasibility of silver perch culture in inland saline-affected areas with high survival and reasonable growth has been recorded.	(Doroudi *et al*., 2007)
Western king prawns (*Penaeus latisulcatus*)	Prawns could survive and grow in potassium-fortified inland saline water but with lower growth rates	(Prangnell and Fotedar, 2006)

	than those reared in marine water.	
Pangasianodan hypophthalmus	Pangasius can withstand salinities as high as 15 ppt in inland saline waters; however, 10 ppt is the ideal salinity for culture.	(Kumar *et al*., 2017)
Nile tilapia (*Oreochromis niloticus*) and spinach (*Spinacia oleracea*)	Nile tilapia exhibited 100% survival rate, a higher feed and protein efficiency ratio, and increased yields when cultured using inland saline groundwater. The integration of spinach and Nile tilapia in an aquaponic system was discovered to work best at 9g/L salinity.	(Thomas *et al*., 2021)
Pacific white shrimp (*Lithogenous vannamei*)	Using low-salinity groundwater, Pacific white shrimp can be cultivated at significantly high yield with good survival.	(Samocha *et al*., 2004)
Freshwater carps (*Catla catla*, *Labeo rohita*, *Cyprinus carpio*)	Higher productivity of carps has been recorded in inland saline water with the salinity range from 4 to 8 ppt.	(Ansal *et al*., 2016)
Barramundi (*Lates calcarifer*)	Asian sea bass larval rearing is possible in saline ground water with sufficient potassium concentration, resulted in improved growth with no negative impacts on survival.	**(**Partridge and Lymbery, 2008**)**

The other recorded candidate species includes *Mugil cephalus*, *Chanos chanos*, *Etroplus suratensis*, *Penaeus monodon*, *Macrobrachium rosenbergii*, molluscs, rotifers, algae, spirulina etc (Jana *et al*., 2004; Raizada *et al*., 2005; Kumar *et al*., 2009; Reddy and Harikrishna, 2014; Sandeep *et al*., 2013; Walsh *et al*., 2008; Lee *et al*., 1997; Awal and Christie, 2015).

Steps to promote inland saline aquaculture in India:

I. It is vital to identify promising sites and assign them to farmers with the required support services to implement suitable inland saline aquaculture practices.

II. Enhancing the accessibility of high-quality seed and feed for farmers starting farming in salt-affected inland areas.

III. Assisting in the marketing of fish as well as providing education about the technologies developed and improving skills of the farmers by conducting various training programmes.

Conclusion:

Agriculture is negatively impacted by the varied salinity of salt-affected soils and groundwater resources in the inland areas. Aquaculture is the most significant method for addressing the issue of soil salinity since it involves the culture of several euryhaline aquatic species in salt-affected areas. Additionally, various technologies and candidate species are claimed to be used to successfully carry out inland saline aquaculture activities. As a result, the underutilized salt affected lands can be effectively exploited to boost the earnings of rural communities and national food security.

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Article ID: 275

ANTIMICROBIAL PEPTIDES (AMPS) IN AQUATIC ANIMALS AND DISEASE

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Abstract

In nature, antimicrobial peptides (AMPs) are widely involved in the innate host defense. Fish are considered a great source of antimicrobial peptides because they represent all the major AMP classes, including cathelicidins, hepcidins, defensins, histone-derived peptides, and piscidins. Antimicrobial peptides (AMPs) are said to be highly promising as a natural alternative to chemical antibiotics. The fish peptides exhibit broad-spectrum antimicrobial action, killing both fish and human pathogens. Moreover, they are immunomodulatory, and their genes are highly reactive against microbes and innate immunostimulatory molecules. Some of the special properties of fish peptides, including their ability to act even in very high salt concentrations, make them good potential targets for development as therapeutic antimicrobials. AMPs have demonstrated diverse biological effects, like endotoxin neutralization, immunomodulating activity, and induction of angiogenesis.

Keywords: Antimicrobial peptides, Fish, Defensin, Cathelicidin, Hepcidin, Piscidin

Introduction

Antimicrobial peptides or proteins (AMPs) are the first line of defense molecules occurring naturally in all multicellular species. The proteins constitute a broad family embracing the ability to destroy yeasts, bacteria, fungi, viruses and even cancer cells, with a broad area of action. AMPs are also referred to as host defense peptides in higher eukaryotic cells. The main groups of AMPs, including cathelicidins, hepcidins, defensins, histone-derived peptides, and a fish-specific class of the cecropin family, called piscidins, fish are an excellent source of these peptides (Masso and Diamond, 2014). AMPs are low molecular weight proteins of about 12–100 amino acid residues, usually positively charged. The structure of AMPs gets folded into an amphipathic structure which makes it soluble in aqueous and lipid because about 40- 50% of AMPs residues are hydrophobic.

AMPs are distinct molecules with the ability to kill targeted microbes using a mechanism in which some cationic peptides create electrostatic attraction for negatively charged phospholipids of microbial membranes and integrate into the cell membrane of microbes resulting in membrane disintegration. AMPs are broad-ranged molecules that have the ability to act against gram-positive and gram-negative bacteria, viruses and fungi. They have other functions like mediators of inflammation, cell proliferation, angiogenesis, wound healing, chemotaxis, immune induction and protease–antiprotease balance. Antimicrobial peptides (AMPs) have been considered a good natural alternative to chemical antibiotics. Fish being eukaryotic organisms, very much relies on their innate immunity, also fish is a good source of AMPs having their application in a wider range.

Fish antimicrobial peptides (AMPs) are small peptides whose activities significantly affect the teleost fish's innate immune response and are considered the first line of protection against a broad spectrum of pathogens. Their antitumor effects make them possible medications to be used in human subjects for oncological therapies. AMPs properties as antimicrobial agents and other concern behaviours, such as cardioprotective (anticoagulant, antihypertensive and antiatherosclerotic) antioxidant, anxiolytic, analgesic, appetite suppressant, antidiabetic, and neuroprotective, have a tremendous potential to be used in many industrial sectors (Shabir et al., 2018). AMPs have enormous potential for mass production for further use, and cloning seems to be uncomplicated because the genes that encode for those peptides are small, and highly conserved regions are usually present (Valero et al., 2020).

A large number of AMPs have been isolated from a wide number of fish species during last years, among which pleurocidin from winter flounder (Pleuronectes americanus) cathelicidins from rainbow trout (Oncorhynchus mykiss), defensins from zebrafish (Danio rerio), piscidins from hybrid striped bass (white bass, Morone chrysops, female, x striped bass, Morone saxatilis, male), dicentracin from sea bass (Dicentrarchus labrax) and hepcidin from channel catfish (Ictalurus punctatus), epinecidin from the grouper (Epinephelus coiodes).

History of AMPs

Lysozyme, the first recorded human antimicrobial protein, was described by Alexander Fleming in 1922 from nasal mucus. Nisin, the oldest yet most common antimicrobial peptide, was first described in 1928. A strain of lactic acid bacteria, Lactococcus lactis, produces this 34-residue peptide. It belongs to antibiotics, a family of bacteriocins that are strongly modified post-translationally. In 1971, the structure was explained. The first major report of alpha-helical AMPs was detecting 'cecropins'. It is reported that a total of 3,240 AMPs have been recorded in the antimicrobial peptide database (APD31) updated on August 24, 2020 (Huan et al., 2020). Teleost fish species express a significant number of AMPs: to date, roughly 117 out of 2,927 AMPs belonging to two prominent families have been isolated, piscidins and hepcidins.

Initially 1980, a toxic peptide, named pardaxin, was characterized from the Moses sole fish (Pardachirus marmoratus), (Primor N and AT Tu, 1980), but until 1996 its antimicrobial activity was not discovered. Afterward, a peptide was isolated from the skin secretions of the fish winter flounder (Pleuronectes americanus), (Cole et al., 1997). Thereafter, the field advanced as with other vertebrate species, with the identification of homologous peptides in the piscidin family along with defensin, cathelicidin, and hepcidin families, which are found in many other species.

Classification of antimicrobial peptides AMPs can be classified in two ways based on

(i) Secondary structure

(ii) Composition in amino acids.

In the first form of classification, AMPs are divided into (a) Linear peptides with an amphipathic helical conformation, (b) Linear peptides with a significant proportion of one residue such as P, R or W, (c) Peptides containing loop structures, (d) β-strand structures containing disulfide bridges. Their amino acid composition can be divided into two main clusters: cationic antimicrobial peptides and non-cationic antimicrobial peptides (Zasloff, 2002 and Narayana and Chen, 2015).

Cationic peptides

Linear α-helix peptides: α-helical AMPs are composed of piscidins, gaduscidins, mononucleosis, grammistins, pardaxin, epinecidin and chemokine.

Disulphide bonded peptides:

Peptides with disulphide brides (one, three or more) as their primary structure are composed of different families. The single disulphide is divided into three groups: linear, distinctive disulphide bonds in their constitution and no homology with the previous ones.

Non-cationic antimicrobial peptides

Their amino acid structure is high in aspartic and glutamic acids and varies significantly from cationic peptides. In fish, most non-cationic AMPs have been isolated during the last decade. They can be derived from oxygen-binding proteins, linear α- helical peptides, those with disulphide bridges, or various other structures.

General Properties of AMPs

Generally, the AMPs are synthesized in exposed tissues, such as skin, intestine, lungs, and red blood cells. As compared to immunoglobulin these peptides are synthesized 100 times at a more rapid rate and at a low metabolic cost, but they can also be stored as a reserve in cells and be released when cells are stimulated by contact with pathogens. These peptides are a fast non-specific way to fight against microorganisms. Bacteriocins are labeled Ribosomally synthetized AMPs from prokaryotes. It has been shown that they are effective tools to protect or secure an ecological niche. It is now well known that the plasmatic membrane is the primary target of these peptides. By disrupting the plasmatic membrane, they operate on the responsive cells.

In order to account for anti-viral activity, three methods have been proposed:

1) By disrupting the lipid bilayers of viral envelopes, immediate inactivation of viral particles.

2) By inhibiting viral-cellular membrane fusion, preventing of viral penetration into the host cell.

3) Inhibition of viral replication in infected cells by suppressing viral gene expression

Antimicrobial peptides isolated from fish

By forming aggregates on the cell's surface at the proper position, AMPs may modify membranes to form transmembrane pores that induce the release of intracellular material. As a consequence of peptide-induced permeation, these peptides bind with each other until they achieve a

Piscidins

A 25-residue peptide isolated and characterized from winter flounder skin mucous secretions, Pleuronectes americanus, named pleurocidin, was the first member of the family (Cole et al., 1997). Piscidins against several microorganisms exhibits potent antimicrobial activity. They are widely active against Gram-positive and gram-negative bacteria species, with the highest antibacterial values obtained against Streptococcus species, Pseudomonas, Bacillus and Vibrio. Chrysophsin-3 was observed to kill the three stages of Bacillus anthracis (sporulated, germinated and vegetative), penetrate and kill the spores without full germination. Piscidins have also shown to possess anti-fungal activity, anti-parasitic activity, and anti-viral activity. Piscidins are primarily present in the gill, skin and intestinal region, and also can be found in the head kidney and spleen. Buonocore et al., (2012) stated that mast cells, rodlet cells, phagocytic granulocytes and eosinophilic granular cells are clearly among the cell types where piscidin are expressed.

Furthermore, pleurocidin (or piscidin) expression is found in winter flounder at 13 days post-hatch, which is suggested to play an essential role in protection during development (Douglas et al., 2001). In addition to microorganisms, picaridin-mediated antitumor activity has been shown by the growth inhibition and killing of many different cancer-derived cell lines such as A549 (adenocarcinoma human alveolar basal epithelial cells). Other appealing characteristics of piscidin include its capacity to maintain antibacterial activity at high salt concentrations, thermostability, as observed in seahorse brooding pouch's piscidin maintained maximum activity after exposure for 30 minutes from 20-80°C, and 20 percent loss of activity observed while boiling for 30 minutes at 100°C. Expression of pro-inflammatory and other immune-related genes can be modulated in fish, such as IL-1β, IL-10, IL-22, IL-26 (Interleukin), TNF-α (Tumour Necrosis Factor-alpha), IFN-γ(Interferon-gamma), NF-κB (Nuclear factor kappa B), lysozyme, NOS2 (nitric oxide synthase 2), MyD88 (Myeloid differentiation primary response 88), TLR4a (toll-like receptor 4), TLR1(toll-like receptor 1), TLR3 (toll-like receptor 3).

Defensins

A general term for cysteine-rich, cationic antimicrobial peptides found in plants, fungi, invertebrates and vertebrates, defensins exhibit a general conformation made by cysteinestabilized α-helical and β–-sheet folds. Fish defensins were first identified through a database mining technique in zebrafish, Fugu, and tetraodon (Zou et al., 2007). Three exons and two introns are found in fish encoding a prepropeptide (including signal peptide, propeptide and mature peptide) consisting of 60 to 77 amino acids and a mature peptide of 38 to 45 cationic amino acids with a pI of approximately 8 (except for olive flounder peptides of approximately 4, indicating an anionic nature) (Nam et al. 2010). Fish β-defensins, albeit with very moderate activity, are active against Gram-negative, Aeromonas hydrophila and Grampositive bacteria, Planococcus citreus with limited MIC (minimum inhibitory concentration) values, are exceptions to these records of MICs in the large µM scale. Also, β-defensins are involved in the treatment of fish-related viruses such as Singapore grouper iridovirus (SGIV), viral nervous necrosis virus (VNNV), Haemorrhagic septicemia virus (VHSV) and Rana grylio virus (RGV) specific to frogs. β-defensins have been shown to show numerous immunomodulatory functions in addition to their antimicrobial activities. Cuesta et al., (2008) observed that a chemotactic behaviour was demonstrated by β-defensins from the gilthead seabream, indicating the ability to recruit head-kidney leukocytes. There are other evidence in zebrafish and rainbow trout of CCR6 (chemokine receptor 6) mammalian orthologs that can help resolve the process. Several factors, including cell wall components such as LPS (Lipopolysaccharide), β-glucans and peptidoglycan stimulate fish β-defensin genes.

Hepcidins

Hepcidins are antimicrobial peptides rich in cysteine and were first found in humans (Krause et al., 2000). Shike et al. 2002 first described and isolated fish hepcidin from the hybrid striped bass and since then hepcidins have been identified in at least 37 fish species. A β-sheet-composed hairpin-shaped with four disulfide bridges (formed by eight cysteines) along with an odd vicinal bridge at the hairpin turn is the general structure of Human Hepcidin, which is also the general structure of fish hepcidin. Two forms of Hepcidin are found in fish, HAMP1 and HAMP2. However, while HAMP1 is present in actinopterygian and non-actinopterygian fish, only actinopterygian fish have been shown to have HAMP2. Like other AMP genes, exposure to both Gram-positive and Gram-negative bacteria can trigger fish hepcidins (Solstad et al., 2008), viruses can also induce hepcidin genes in fish (Yang et al., 2013), and poly I:C (polycytidylic acid) as well as mitogens.

The capacity to affect the viability of cancer cells has also been demonstrated by fish hepcidin. Tilapia hepcidin TH2-3, for example, has demonstrated concentration-dependent inhibition of proliferation and migration of the human fibrosarcoma cell line HT1080a. In addition, in HT1080 TH2- 3 was able to induce cell membrane destruction and findings also show that TH2-3 down-regulates c-Jun contributing to apoptosis. However, beyond the possible antimicrobial and immunomodulatory consequences, Hepcidin is best recognized as a crucial ferroportin-controlled iron regulator capable of degrading its internalization, which decreases the transfer of iron into the blood.

Cathelicidins

The first cathelicidins found in fish were initially isolated from the Atlantic hagfish, Myxine glutinosa, as antimicrobial peptides. Fish cathelicidins can be subdivided into two classes; the linear peptides, and the characteristic disulphide bond. Considerable sequence homology is seen among members of the groups (up to 90%) and minute homology between the classes compared to mammalian cathelicidins. Moreover, a third class (focused on sequence homology between themselves and a lack of homology with any of the other two classes) tends to be the newly described cathelicidins found in cod (Maier et al., 2008). It also shows intense anti-fungal activity against C. albicans. On the other hand, Hagfish cathelicidins are active against Gram-positive and Gram-negative bacteria, but inactive against Candida sp. More precisely, rainbow trout cathelicidins are active against Y. ruckeri, while Atlantic salmon cathelicidins are not. It is studied that, in mammals, cathelicidins showed multiple activities, both as immune and non-immune, as well as in excess of their in vitro antimicrobial activities. Although, in fish, research has not approached this level, a recent study demonstrated that two Atlantic salmon cathelicidins induced the rapid and transient expression of IL-8 in peripheral blood leukocytes. This suggests that the immunomodulatory activities seen by mammalian cathelicidins may be shared by their fish counterparts, and may thus be an evolutionarily conserved mechanism of innate immune regulation.

Histones derived peptides

In a variety of fish species, histone-derived AMPs have been identified with broad-spectrum action against both human and fish pathogens (Noga et al., 2011), including water molds (Robinette et al., 1998) and a parasitic dinoflagellate (Noga et al., 2001). In fish skin, they are expressed and secreted and found in other tissues, including the gill, spleen, and intestines. Robinette and Noga (2001) further showed that they play a part in host fish defense from experiments demonstrating that histone-derived AMP gene expression is triggered in various fish species' particular tissues under stress conditions.

Therapeutics

All AMPs exhibit common characteristics of development as therapeutic antimicrobials which include a wide range of activity against a variety of pathogens; potent activity under a wide range of conditions, including temperature and in secretions such as saliva; and a reduced capacity to the development of resistance by bacteria. The identification and characterization of peptides from fish have provided a unique contribution.

Since many AMPs are sensitive to high salt concentrations while some of the fish AMPs have the ability to kill microbes even at extremely high salt concentrations, such as those found in the marine environment. The AMPs from fishes are active against the pathogens like nervous necrosis virus. Also, AMPs have dual functions for example hepcidins are involved in iron regulation as well Piscidins are present in both phagocytic granulocytes as well as in mast cells. For instance, piscidin 2 concentrations in various tissues of hybrid striped bass are lethal to different ectoparasites and the concentrations of piscidin 4 in gills are fatal to various bacteria.

Table 1. Classification and Function of fish Antimicrobial Peptides (AMPs)

Sl.No.	Name of the Antimicrobial Peptides (AMPs)	Fish Species	Function
Piscidin - AMPs			
1.	Piscidin-1	*Gadus morhua* (Atlantic cod)	Antibacterial
2.	Piscidin-2	*Gadus morhua* (Atlantic cod)	Antibacterial
3.	Piscidin-1	*Morone saxatilis* (Striped bass)	Antibacterial
4.	Piscidin-2	*Morone saxatilis* (Striped bass)	Antiparasitic, Antibacterial, Antifungal
5.	Piscidin-1,-2,-4,-5)	*Morone saxatilis* x *M chrysops* (Hybrid Striped bass)	Antibacterial
6.	Dicentracin	*Dicentrarchus labrax* (European sea bass)	Unknown
7.	Moronecidin	*Morone chrysops* (White bass)	Antibacterial, Antifungal
8.	GAD-1, -2	*Gadus morhua* (Atlantic cod)	Antiviral, Antibacterial
9.	Epinecidin-1	*Epinephelus coioides* (Orange spotted-grouper)	Antibacterial, Antiviral, Immune modulator
10.	Pleurocidin	*Pseudopleuronectes americanus* (Winter flounder)	Antibacterial
11.	Chrysophsin-1, -2, -3	*Chrysophrys major* (Red seabream)	Antibacterial, Hemolytic
12.	asCath-1 asCath-2	*Salmo salar* (Atlantic salmon)	Antibacterial Immune modulator
Defensins - AMPs			
1.	CATH1-SALTR	*Salmo trutta fario* (Brown trout)	Unknown
2.	CATH1-SASLFO CATH2-SASLFO	*Salvelinus fontinalis* (Brook trout)	Unknown
3.	aCath	*Plecoglossus altivelis* (Ayu)	Antibacterial
4.	Grammistins Pp1, Pp2a, Pp2b, Pp3, Pp4a, Pp4b	*Pogonoperca punctata* (Soapfish)	Antibacterial Hemolytic Ichthyotoxic
5.	Grammistins GsA, GsB, GsC, GsD, GsD	*Grammistes sexllineatus* (Golden-striped grouper)	Unknown
6.	Astacidin-2	*Pacifastacus leniusculus* (Crayfish)	Antibacterial
7.	HLP1	*Oncorhynchus mykiss* (Rainbow trout)	Antibacterial
8.	H1	*Morone saxatilis x M. chrysops* (Hybrid striped bass)	Antibacterial
9.	Parasin I	*Parasilurus asotus* (Catfish)	Antibacterial Antifungal

Sl.No.	Name of the Antimicrobial Peptides (AMPs)	Fish Species	Function
Hepcidins - AMPs			
1.	Hbβ P1, P2, P3	*Ictalurus punctatus* (Channel catfish)	Antiparasitic Antibacterial
2.	Astacin-1	*Pacifastacus leniusculus* (Crayfish)	Antibacterial
3.	omBD-1	*Oncorhynchus mykiss* (Rainbow trout)	Antibacterial Antiviral
4.	zfBD1, 2, 3	*Danio rerio* (Zebrafish)	Unknown
5.	Defensin	*Epinephelus coioides* (Orange-spotted grouper)	Antibacterial Antiviral
6.	Hepcidin	*Sparus aurata* (Gilthead seabream)	Antibacterial
7.	Hepcidin	*Morone saxatilis x M. chrysops* (Hybrid striped bass)	Antibacterial
8.	Sal -1, Sal - 2	*Salmo salar* (Atlantic salmon)	Unknown
9.	LEAP - 2	*Ictalurus furcatus* (Blue catfish)	Unknown
Cathelicidins - AMPs			
1.	apoA - I apoA -II	*Cyprinus carpio* (Common carp)	Antibacterial
2.	NK -lysin	*Ictalurum punctatus* (Channel catfish)	Unknwon
3.	JF -NK - 2	*Paralichthys olivaceus* (Japanese flounder)	Antibacterial
4.	NK -lysin	*Danio rerio* (Zebrafish)	Unknown
5.	MAPP	*Misgurnus anguillicaudatus* (Loach)	Antibacterial
6.	Myxinidin	*Myxine glutinous* (Hagfish)	Antibacterial
7.	Misgurin	*Misgurnus anguillicaudatus* (Loach)	Antibacterial Antifungal
8.	From pepsin hydrolysate	*Setipinna taty* (Scaly hairpin anchovy)	Antibacterial

Conclusion

Antimicrobial peptides are found as host defense molecules in all animals from primitive prokaryotes to most evolved eukaryotes. As examined fishes are challenged many times by a variety of pathogens which not only affect their health but also increase the risk of becoming resistant to conventional antibiotics which severely affects the aquaculture industry. Therefore, AMPs can be considered as the potential proteins for developing therapeutic agents in the field of aquaculture. AMPs are examined to have diverse biological effects like endotoxin neutralization, immune modulating activity, and induction of angiogenesis and therefore, they are considered as very vital and attractive therapeutic tools. For the future research study should be done on different human health benefits and therapeutic effects of AMPs which can be used for treating diseases like microbial as well as cancerous. Synthetic drugs are either least effective or completely ineffective and have lesser safety margins, these endogenous peptides will prove to be potent and effective, thus the health benefits of AMPs need to be evaluated.

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Article ID: 276

RICE BREEDING BREAKTHROUGH COULD FEED BILLIONS, APOMIXIS IN HYBRID RICE

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Introduction

Day by day increase in global population which can have a detrimental impact on global food security. That's why farmers will need to increase crop yield, either by increasing the amount of agricultural land, adopting new technology like precision farming and high yielding varieties. But most Indian farmers are small land holder that's why only high yielding varieties is the best available option for increasing crop yield. Heterosis is exploited by plant breeders to produce elite high yielding varieties, but beneficial phenotypes are lost in subsequent generations owing to genetic segregation (Wang *et al.,* 2018).

Rice, the staple food for more than half of mankind, has a naturally low outcrossing rate and the production of F_1 hybrid seeds relies on the implementation of complex male sterility systems, resulting in a high seed cost. Thus, the dissemination of hybrid rice is essentially restricted to areas with efficient and well-established seed production and distribution systems. As a result, the benefits of hybrids have not yet reached to large number of rice farmers.

A potentially revolutionary alternative to male sterility systems is the propagation of F_1 seeds over generations in an immortalized manner through an asexual, clonal mode of reproduction called apomixis. But introgression of apomictic genes from wild relative to cultivated one is very difficult task and therefore found unsuccessful. Engineered synthetic apomixis can provide a low-cost, immortalized source of F_1 seeds that will allow their use by small farmers. Now synthetic apomixis development is observed feasible in rice through the inactivation of three genes (*MiMe*), which results in the conversion of meiosis into mitosis in a line ectopically expressing the *BABYBOOM1* (*BBM1*) parthenogenetic trigger in egg cells (Vernet *et al.,* 2022).

Major limitations of hybrid seed production using male sterility and self-incompatibility

- Maintenance of parental lines
- Synchronization in the flowering
- Regular rouging
- Isolation distance
- Laborious, and expensive approach
- Environment influence

Major advantages of apomixis

- Hybrids to be produced once and propagated through seeds for multiple generations.
- Obligate apomixis permits fixation of heterosis in the hybrids

Features of apomictic plant

- Apomictic reproduction essentially involves three components, *viz*. apomeiosis, parthenogenesis and functional endosperm development.
- Apomictic species are almost perennials, polyploid and often use a vegetative reproduction.
- Gametophytic apomixis is more common and most apomicts produce viable pollen.
- Evolutionary point of view, apomixis may be regarded as a consequence of sexual failure rather than as a recipe for clonal success.

Ideal apomictic system

- Obligate apomixis: All progeny should be apomictic.
- Apomictic genotype should be male fertile and self-incompatible and reproduce *via* pseudogamy.
- Pollen production is essential for transfer of genes controlling apomixis in normal sexual genotype.
- In diplospory, chromosome should not recombine and restitution must occur at or just after first meiosis.
- Dominant over sexual reproduction and should not be affected by environmental changes

Types of Apomixis

- **Gametophytic**: Embryo develops from the egg cell of a well-developed embryo sac, without fertilization.
    - Apospory: Apomeiosis pathway where a diploid embryo sac develops from a somatic ovule cell that is not the megaspore mother cell, called the aposporous initial (AI) cell.
    - Diplospory: Diploid megaspore mother cell is directly converted into the mature embryo sac through mitosis and omitting miosis
        - Parthenogenesis: Development of an egg found in a diplosporous or aposporous embryo sac into an embryo without fertilization.
        - Apogamy: Development of mature embryo from Synergids and Antipodals cell.
    - Androgenesis: Development of whole plant from male gamete using tissue culture protocol.
- **Sporophytic**: Direct formation of one or more embryos from somatic diploid ovule cells termed embryo initial cells.
    - Adventitious embryony: development of mature embryo sac from chalaza, nucellus and integuments.

Breeding approaches

- **Introgression**: Wide hybridization between a crop species and an apomictic relative, followed by a backcross breeding, *e.g. Tripsacum dactyloides* to *Zea mays*, *Pennisetum squamulatum* to *P. glaucum* (Pearl millet), *Elymus rectisetus* to *Triticum* (Wheat)
- **Apomictic recombinants from interspecific crosses**: Progeny of interspecific or intergeneric hybrids of two sexual species, *e.g.* Intergeneric hybrids between *T. aestivum* and *Avena sativa, Hordeum vulgare* and *T. aestivum/turgidum*
- **Mutation**: Mutation that produce an egg with an unreduced number of chromosomes (Meiotic mutants) & that develop embryo without fertilization (Parthenogenetic mutants), *e.g.* Sorghum R473
- **Genetic transformation**: exogenous and endogenous genes that control the expression of apomixis.
- **Genome editing**: In Rice (Oryza sativa) create mutation in *OsSPO11-1, OsREC8, OsOSD1,* and *OsMATL* through a CRISPR/Cas9 system.
- Identification of candidate genes governing apomixis through study of ovule transcriptome.
- Molecular tagging and transcriptomic approaches are being employed to find genes governing apomixis using natural variants, mutants, introgression lines.
- Isolation of RNA from ovules of sexual and apomictic plants and transcriptome sequencing.
- qRT-PCR analysis for the validation of differentially expressed apomictic transcripts.

Methods to identify apomictics

- **Phenotypic identification**
- **Cyto-embryological detection**
  - Pistil-clearing technique
  - Callose fluorescence
- **Flow cytometry analysis**
- **Biochemical techniques**
- **Molecular markers**
- **Histological technique**
- **Whole genome resequencing**

Problems associated with apomixis

- Complex and tedious mechanism.
- Estimation of the level of apomixis difficult.
- In the absence of linked morphological markers, identification is very difficult task.
- Maintenance of apomictic stock become difficult (Facultative apomixis).
- Nuisance when the breeder desires to obtain sexual progeny.

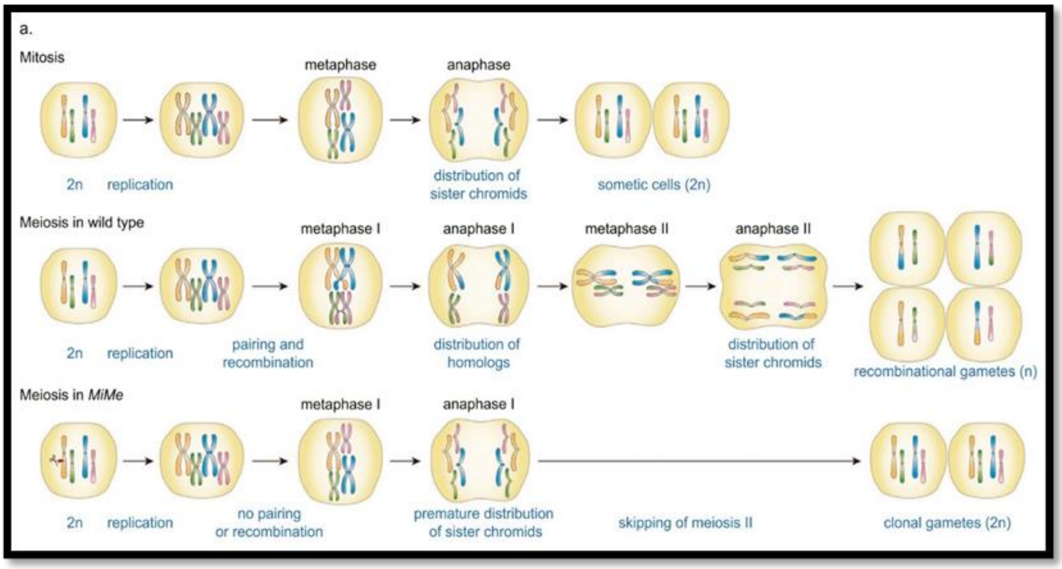
- Apomictic genes could escape into wild relatives and cause genetic erosion.

Engineering synthetic apomixis

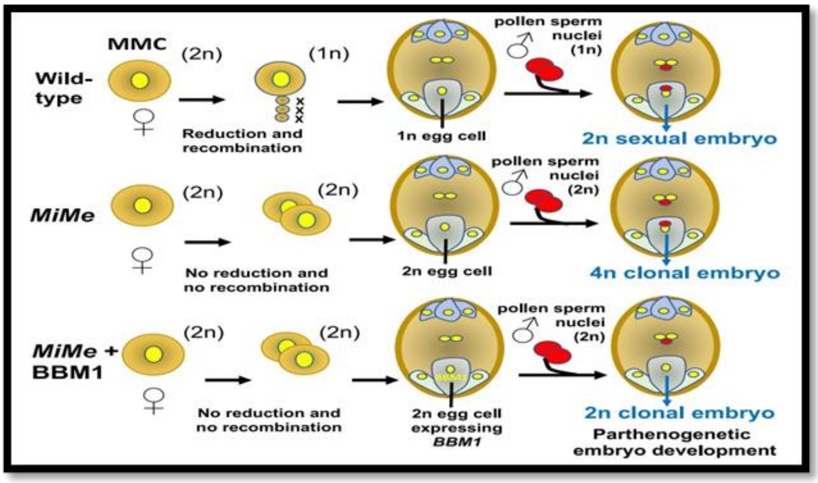
- Engineering synthetic apomixis has proven feasible in inbred rice through the **inactivation of three genes (MiMe)**, which results in the **conversion of meiosis into mitosis** in a line ectopically expressing the ***BABYBOOM1* (*BBM1*) parthenogenetic** trigger in egg cells.
- Three critical steps
  1. Formation of functional female gametophytes without meiosis (apomeiosis).
  2. Formation of embryos in the absence of fertilization (parthenogenesis).
  3. Formation of viable endosperm either *via* autonomous or pseudogamous mechanism.

MiMe strategy

- MiMe combines three mutations that occur meiosis into a mitotic-like division (apomeiosis)
  1. Abolishing meiotic recombination.
  2. Separating the sister chromatids.
  3. Skipping the second division.



Xiong *et al.* (2023)



Vernet *et al.* (2022)

Advantages of synthetic apomixis

- Provide a low cost, immortalized source of F_1 seeds that will allow their use by smallholder farmers.
- Without using male sterile sources, we can develop diversified synthetic apomictic hybrids.
- Maximum potential improvement in grain quality, which has been a limiting factor in F_1 hybrids.
- Easily overcome the limitations of CMS based hybrid seed production
- Minimum time requirement for fixation of available heterosis

Future thrust

- Effective genome combinations by using bioinformatics to exploit dispensable rice genome for developing future climate-smart apomictic hybrids.
- The identification of QTLs/genes through new mapping technologies, like comparative mapping, linkage disequilibrium mapping, deletion mapping and high throughput sequencing methods, will help to reveal core apomixis chromosomal regions.
- Novel technologies can expose or remove the evolutionary genetic load of genes and epigenetic modifications related to apomixis and can be further utilized in agriculture as a tool to fix elite genotypes that are important for food security.
- High-throughput, fast, and accurate selection techniques to distinguish clonal seeds or plants among the offspring must be developed. Recently, an efficient and accurate visual identification system *via* anthocyanin has been established in maize, accuracy as high as 99.1%, Therefore, apomixis system, which contains the genome elimination genes, may adopt the same visual strategy that distinguished the clonal seeds based on the presence or absence of paternal genome.
- Additionally, apomixis system, engineered with parthenogenesis genes, may employ the fluorescence detection system, which ectopically expressed a fluorescent gene and parthenogenetic gene simultaneously.

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Agri Mirror: Future India
ISSN: 2582-6980

Article ID: 277

REVOLUTIONIZING AGRICULTURE: THE POWER OF AUTOMATION IN IRRIGATION

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ABSTRACT

Agriculture stands at the forefront of technological transformation, with automation in irrigation emerging as a pivotal innovation. This article explores how automated irrigation systems are revolutionizing farming practices by addressing the challenges posed by traditional methods. It investigates the historical evolution of irrigation, highlighting the limitations of manual approaches in the face of climate change and water scarcity. The article then delves into the core principles of automation, emphasizing the role of sensors, precision water delivery, and remote monitoring and control in optimizing resource usage. Real-world case studies from regions such as California's Central Valley, Israel, and India illustrate the tangible benefits of automated irrigation. Moreover, the article scrutinizes the challenges associated with automation, including initial investments, technology adoption, and sustainability concerns. It concludes by emphasizing the imperative role of automation in ensuring a sustainable and food-secure future for global agriculture.

Introduction

In an era where technology is transforming every aspect of our lives, agriculture is no exception. Automation in irrigation is at the forefront of this transformation, reshaping the way we cultivate crops and manage water resources. This revolutionary approach to farming not only boosts productivity but also conserves water, reduces labour costs, and ensures food security in an ever-changing climate.

The Challenge of Traditional Irrigation

Traditional irrigation methods have been the backbone of agriculture for centuries, but they come with their fair share of challenges. Manual irrigation systems often involve inefficient water distribution, overuse of water resources, and heavy reliance on human labour. These methods are also susceptible to human errors, weather fluctuations, and changing water availability due to climate change.

Traditional irrigation has served humanity for millennia, but it comes with inherent flaws. In this section, we examine the limitations of manual irrigation systems, from inefficient water distribution to labor-intensive practices. We explore how these shortcomings have driven the need for automated solutions.

The Solution: Automated Irrigation

Automated irrigation systems have emerged as a game-changer in modern agriculture. These systems leverage cutting-edge technology to precisely control when, where, and how much water is delivered to crops. Here are some of the key benefits of automation in irrigation:

1. Water Conservation: Automated systems use sensors to monitor soil moisture levels and weather conditions in real-time. This data enables them to deliver the precise amount of water needed, reducing water wastage significantly.

2. Increased Productivity: By ensuring that crops receive the right amount of water at the right time, automation leads to healthier plants and higher yields. Farmers can also cultivate more efficiently, saving time and resources.

3. Labor Savings: Automated irrigation reduces the need for manual labour, allowing farmers to allocate their workforce to other critical tasks such as pest control, harvesting, and maintenance.

4. Improved Crop Quality: Consistent and controlled irrigation enhances the quality of crops, making them more marketable and less prone to diseases related to overwatering.

5. Weather Resilience: Automated systems can adjust irrigation schedules in response to changing weather patterns, reducing the risk of overwatering during rainy periods or underwatering during droughts.

6. Remote Monitoring and Control: Farmers can manage their irrigation systems remotely through smartphones or computers, giving them greater flexibility and control over their operations.

7. Sustainability: Automated irrigation contributes to sustainable agriculture by minimizing the environmental impact of excessive water use and reducing energy consumption.

The Power of Automation

The principles and technologies that underpin automated irrigation systems. We explore the following key aspects:

Sensors and Data: Automated systems rely on a network of sensors to gather data on soil moisture levels, weather conditions, and plant health. We discuss how these sensors work and their role in optimizing water usage.

Precision Water Delivery: The core of automation lies in precision – delivering the right amount of water at the right time to each plant. We examine how automation achieves this level of precision, even in large-scale farming operations.

Remote Monitoring and Control: One of the most significant advantages of automation is the ability to manage irrigation systems remotely. We discuss how farmers can access and control their systems via smartphones or computers, ensuring flexibility and efficiency.

Real-Life Success Stories

Several real-life examples showcase the transformative power of automated irrigation:

1. California's Central Valley: Farmers in this drought-prone region have adopted precision irrigation techniques, reduced water use while maintaining high crop yields.
2. Israel's Drip Irrigation: Israel, a country with scarce water resources, has pioneered drip irrigation technology, enabling farmers to grow crops efficiently in arid conditions.
3. Indian Agriculture: In India, where agriculture is a significant contributor to the economy, automated irrigation has helped millions of farmers increase their income while conserving water.

Challenges and Considerations

Despite its numerous benefits, automation in irrigation also faces challenges such as initial investment costs, technology adoption barriers, and the need for skilled technicians to operate and maintain these systems. Additionally, there is a risk of overreliance on technology, which could lead to neglect of traditional farming knowledge.

While automated irrigation presents numerous advantages, it also faces challenges and considerations:

1. **Initial Investment**: We discuss the upfront costs associated with implementing automated irrigation systems and strategies to overcome them.

2. **Technology Adoption**: Automation requires a shift in mindset and skillset. We explore the barriers farmers might face when adopting this technology and how education and training play a crucial role.

3. **Sustainability**: As with any technology, there is a risk of overreliance on automation. We examine the importance of striking a balance between traditional farming knowledge and modern technology.

Conclusion

Automation in irrigation is a compelling example of how technology is reshaping agriculture. By conserving water, increasing productivity, and improving crop quality, automated irrigation systems are essential tools for modern farmers. As we continue to face climate change and water scarcity, embracing these innovations will be crucial for the sustainable future of agriculture, ensuring food security for generations to come.The potential of automated irrigation to drive a sustainable future for agriculture. As we face climate change and water scarcity, embracing automation is not just an option but a necessity. We emphasize the role of automation in ensuring food security and the prosperity of farming communities worldwide.

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Article ID: 278

BIOFERTILIZERS IN MODERN AGRICULTURE: BENEFITS, APPLICATIONS AND FUTURE PROSPECTS

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Abstract

With the continuous growth of the global population, there is a pressing need to expand food production to meet the rising demand. Moreover, arable land has gradually lost its natural nutrients over time, leading to a reliance on fertilizers, pesticides, and herbicides to replenish soil fertility and increase agricultural output. While significant efforts are being directed towards fulfilling this food demand and ensuring food security, it is equally critical to guarantee food safety at every stage of the food production process. One fundamental strategy involves shifting from chemical or synthetic fertilizers, pesticides, and herbicides to the use of biofertilizers and plant growth promoters, which pose fewer risks to human and animal health. Biofertilizers competitively establish themselves in plant root systems, thereby enhancing nutrient absorption, boosting productivity and crop yields, enhancing plant stress tolerance and resistance to pathogens, and promoting plant growth through mechanisms like the activation of vital elements, nutrients, and plant growth hormones. Biofertilizers are both economical and environmentally friendly, and their consistent application contributes to the long-term improvement of soil fertility.

Introduction:

Biofertilizers are described as formulations that consist of living microorganisms or dormant cells with the ability to establish themselves in the root systems of crop plants. They enhance plant growth by enhancing the availability and uptake of nutrients. When liquid biofertilizers are produced, it is essential to follow specific protocols to protect the organisms and ensure their effective transportation to the intended locations, thereby enhancing their biological effectiveness. This requires formulating a mixture of microorganisms within a suitable medium to maintain their vitality for a specified duration, ultimately leading to an augmentation of biological activity at the specified site. The formulations of biofertilizers are crafted through the amalgamation of powerful microorganisms with carrier-based inoculants. Carriers serve as beneficial carbon sources for the integration of microorganisms, enhancing the biofertilizers efficiency by extending their shelf life. Bacterial inoculants encompass rhizobia, nitrogen-fixing rhizobacteria, and plant growth-promoting rhizobacteria (Naveed *et al.,* 2008). An ideal carrier material should be non-harmful to both the bacterial strains used as inoculants and the plants possess excellent moisture-absorbing properties, be easy to process and sterilize, cost-effective, and readily accessible. It is also beneficial for carriers to exhibit strong adhesion to seeds and maintain good pH and buffering capacity. Demonstrated effective carrier materials include rice husk, farmyard manure (FYM), charcoal, peat, and lignite.

Benefits of biofertilizers

Biofertilizers, consisting of microbial inoculants, present an environmentally friendly alternative to chemical fertilizers. They play a significant role in safeguarding the lithosphere, improving the biosphere by mitigating air, water, soil pollution, and eutrophication, and enhancing agricultural yields. These biofertilizers aid in enriching the soil with both macro- and micro-nutrients although also releasing plant growth regulators. Especially, the essential plant nutrients, nitrogen, phosphorus, and potassium, are effectively catered to by these microbial inoculants in biofertilizers, which stimulate plant growth and yield by secreting antibiotics, enzymes, antifungal, and antibacterial substances, and/or by releasing hormones that help plants combat diseases and stress (Stewart and Roberts, 2012). Biofertilizers can be categorized into various types, including nitrogen-fixing, phosphate-solubilizing, phosphate-mobilizing, potassium-solubilizing, potassium-mobilizing, and sulfur-oxidizing varieties.

The formulation of liquid biofertilizers involves specific conditions for preserving the organisms and facilitating their delivery to the intended areas, ultimately enhancing their biological effectiveness. This entails creating a consortium of microorganisms within a suitable medium to maintain their viability for a specified duration, thereby promoting increased biological activity at the target location. The development of liquid formulations is an emerging technology in India, characterized by distinct and unique production methods. Liquid biofertilizers are microbial products that consist of particular beneficial microorganisms with the ability to fix, solubilize, or mobilize plant nutrients through their biological activities.

These can be generally categorized into three main groups:

1. Nitrogen-Fixing Microbes (NFM)
2. Phosphorus-Solubilizing Microbes (PSM), along with Phosphate-Mobilizing Microbes, and
3. Potash-Mobilizing Microbes

The liquid bio-fertilizers have a shelf life of two years and can withstand high temperatures of up to 55°C and exposure to ultraviolet radiation. The microbial count remains consistently high at 109 colony-forming units per milliliter (C.f.u/ml) throughout this period. Therefore, applying just 1 ml of these liquid bio-fertilizers is equivalent to using 1 kilogram of carrier-based bio-fertilizers that are 5 months old, making it 1000 times more effective. The liquid form makes field application straightforward and hassle-free, allowing for various application methods such as hand sprayers, power sprayers, drip irrigation systems, and mixing with basal manure like farmyard manure (FYM), among other options.

Other benefits of Liquid Bio-fertilizers:

1. Incorporation of special cell protectants or elements that promote the development of dormant spores or cysts.
2. Utilization of specialized nutrients to guarantee extended shelf life, improved viability on seeds and in the soil, and resilience in adverse environmental conditions.
3. The liquid formulation ensures ease of handling and application.
4. Microorganisms are maintained in a stable state during production, distribution, and storage, with their activity being enhanced upon contact and interaction with the target crops.
5. Enhanced potential to effectively compete with the native microbial populations.
6. Dosage requirements are ten times lower as compared to carrier-based bio-fertilizers.
7. Remarkably high enzymatic activity is achieved due to minimal contamination.

Application of biofertilizers:

When applying microbial bio-fertilizers, it's essential to keep in mind that the majority of these microorganisms are heterotrophic. This means they are unable to synthesize their own nutrients and rely on organic carbon present in the soil for their energy and growth. Consequently, they either establish colonies in the rhizosphere area or engage in symbiotic relationships within the roots of higher plants. Bacteria that colonize the rhizosphere zone derive their organic carbon compounds from the root exudates of higher plants, while symbiotic bacteria obtain organic carbon directly from the plant roots. Therefore, it is crucial to apply microbial inoculants in a manner that ensures these bacteria adhere to the root surface. When transplanting the crops, through the roots of plants the inoculants are applied. Whereas for crops sown directly in the field, the inoculants are applied via the seeds. This strategy ensures that the inoculants can establish themselves in the rhizosphere area as young roots emerge following seed germination.

Limitations of biofertilizers:

1. Biofertilizers should not be combined with chemical fertilizers.
2. Biofertilizers should not be applied simultaneously with fungicides or plant ash.
3. Avoid direct sunlight exposure.
4. Store within the temperature range of 0 to 35°C at room temperature.

Future prospects:

In response to the challenges faced by a rising global population and food scarcity, various agricultural practices have been employed. These include the widespread use of chemical or synthetic fertilizers, pesticides, and insecticides to achieve high crop yields in a short time and protect crops from insect and pest damage during and after harvesting. However, the utilization of these fertilizers and pesticides has raised substantial public concerns regarding the sustainability and safety of our food supply. Research has indicated the presence of significant pesticide residues in food items long after they have left the farm and entered the human food chain. The global increase in population and the depletion of natural resources have given rise to a range of environmental and agricultural challenges. Addressing these challenges necessitates the adoption of sustainable agricultural and environmental practices. Innovative methods are crucial to meet current food demands and sustain crop production. Achieving optimal plant growth, increased productivity, and a balanced supply of nutrients are key factors in promoting sustainable agriculture (Daniel *et al.,* 2020). Therefore, there is a pressing need for alternative approaches, such as biofertilizers, to ensure food safety and security. In many developing countries, soil infertility is a major impediment to crop productivity. Large-scale production of biofertilizers has the potential to transform not only the agricultural sector but also the livelihoods of farmers. Biofertilizers represent products that, with the provision of sufficient knowledge to producers and farmers, hold great long-term commercial promise. The global adoption of biofertilizers is poised to yield not only economic benefits for sustainable agriculture but also make significant contributions to fostering a sustainable ecosystem and enhancing the overall welfare of humanity.

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Article ID: 279

BETACAROTENE BIO-FORTIFICATION IN SWEET POTATO

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Sweet potato *(Ipomoea batatas)* belonging to the family of convolvulaceae, is one of the mostly consumed tuber and security food crop for tropical regions. Over 400 varieties of sweet potato are frown across the world. The major consumer of sweet potato in the world is China computing for 66% of global consumption. Many of the commonly cultivated sweet potato varieties are less in vitamin A and generally white and yellow-fleshed. Sweet potato is a relatively easy crop to grow, and conventional wisdom correctly classifies it as a crop of the poor. Most dominant varieties are white-fleshed, having no beta-carotene. Biofortification breeding programmes aims to enhance the vitamin A nutrient in sweet potato.

In world level sweet potato area and production was 9.20 million hectare and 112.83 million tonnes. Over 24.2 million tons production of sweet potato is produced in South Africa (1). Beauregard is the most commonly cultivated high yielding sweet potato variety in world and it is orange fleshed sweet potato and has fairly good amount of vitamin A. Apart from the conventional breeding methods used in biofortification new biotechnological tools are being used (2).

VITAMIN A

The deficiency caused by micronutrients is called hidden hunger and vitamin A deficiency leads to hidden hunger. Vitamin A deficiency causes weak immunity, blindness, ill heart health and poor skin. People of Africa and South-East Asia face serious health issues, due of vitamin A deficiency. For the last 20 years, the biological fortification is the important aim in biotechnology. Transgenic breeding is aimed to transfer the high beta carotene gene (*IBOr-Ins*) from orange fleshed varieties to white fleshed varieties. The orange fleshed sweet potato varieties are rich in Vitamin A and efficient in improving the vision, good immune power and protect the body for germs. Eradicating Vitamin A deficiency reduces child mortality rates. The carotenoids content in the sweet potato is 1.1 to 26.5 μg/g fresh weight. The measuring unit of provitamin A is RAE (Retinol Activity Equivalents) (3). Daily dietary requirement of vitamin A for preschoolers is 400 μg RAE. The cooked portion of sweet potato 125g/day contain greater than 10% Zn, Fe, Mg and vitamin C required for school-aged children .Different biotechnology strategies are used, first strategy to enhance the β-carotene in tubers by using the strategies of targeting overexpression of specific catalysing enzymes, second strategy to increasing the pigmentation by using gene silencing, third strategy to accumulation of carotenoids by overexpression of gene (4). Last strategy to reduce the post-harvest losses of carotenoids is by using cold storage. To increases the nutrient quality in sweet potato by using desired traits such as resistance to virus diseases, pigmentation etc. to agronomic characters in breeding program. In Sub-Sahara Africa 158 varieties were released in sweet potato, out of which 98 varieties are orange-fleshed sweet potato. And also 60% of the African community is affected by Fe deficiency and 1/3th of the community is affected by Zn deficiency. The iron content of orange fleshed sweet potato is 0.63-15.26 μg/100 g. The popular orange-fleshed sweet potato varieties are Beletech (192026 II), Birtukanie (Saluboro), Kulfo (Lo-323), Tulla (CIP 420027) and Awassa-83 (5). Apart from the nutritional benefits orange fleshed sweet potato has medicinal values too. In sweet potato, conventional breeding has not been too victorious because of male sterility problem and cross-incompatibility. Molecular breeding and genetic engineering is used for biofortification (6).

PLANT BREEDING STRATEGIES TO ENHANCE BETA CAROTENE IN SWEET POTATO

International Potato Center (CIP) is actively involved in biofortification of sweet potato with the support from CGIAR. Traditionally bred Orange Sweet Potatoes containing PVA carotenoids were the first biofortified crop developed and released by CIP, HarvestPlus and its partners. Delvia, Chiwoko, Olympia, Chumfwa, Kokota, Ejumula, Kakamega, Kabode, Gerald and Joweria are the important biofortified sweet potato varieties released by CIP under harvest plus programme (7).

Consumer's preference is the basic requirement for the success of any variety. The major constraint of lack of critical mass involved in sweet potato breeding has been overcome, high dry matter orange fleashed varieties (OFSP) are available, the accelerated breeding method validated and the sweetpotato *speedbreeders* are using common protocols and analytic tools as a successful and growing community of practice. In 2016, 30 breeders working in 14 countries signed a memorandum committing to the principle that at least 50% of the varieties they submit for release will be orange-fleshed. With appropriate water management systems, OFSP clearly has the potential to expand into more semi-arid regions where its dual purpose potential for feed and food is appreciated by communities with sizable livestock assets (8). Early maturing OFSP varieties integrate easily into many food systems facing changing climatic conditions because it produces quickly (3–4 months) within a broad temperature range, can produce on marginal soils, and as a rotation crop with grains, legumes or vegetables, helps manage pest and disease incidence (9).

There is no doubt that the transformation opportunities for sweetpotato into processed products will only continue to expand, the pace depending on the price of sweet potato relative to other ingredients. Improving productivity through better varieties, more efficient seed systems, and improved crop management and post-harvest practices will help lower overall price and drive expanded industrial use.

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Article ID: 280

AN OVERVIEW OF EXTENSION TRAINING FOR FARMERS AND EXTENSION PROFESSIONALS

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DEFINITION

"Activities which strengthen individual knowledge, abilities, skills, and behavior and improve institutional structures and processes so that the organization can efficiently meet its mission and goals in a sustainable manner" is how capacity building is described. One of the crucial elements of capacity building is training.

TRAINING

- Training is becoming an integral component of HRD. It has developed into one of the elements that allows any institution to produce its personnel who are the most productive and qualified.
- Training is the art of improving a worker's knowledge and ability to do a certain profession.
- A generally permanent change in behavior that results from experience is what training is intended to achieve.
- According to Milton Mall (1980), training is the process of assisting employees in becoming more effective in their current or future employment by the development of suitable thinking, action, skill, knowledge, and attitude habits.
- According to Krietner (1989), training is the process of modifying an employee's behavior, attitudes, or views.
- According to Milkovich and Boudreau (1998), training is a systematic procedure that modifies the behavior, knowledge, and/or motivation of current employees to better match their skills with job requirements.

Need for Training

- Training has largely caught up, especially in industries. This is due to the abrupt and competitive shift that is taking place in the globe. However, the following factors can be used to determine the requirement for training:
- Rapid advancements in technology and employment.
- A short-term and long-term scarcity of skills
- Changes in workforce expectations and demographics
- Market pressure and competition are driving improvements in product and service quality.

Types of training given to extension personnel – This is of broadly two types

Pre-Service Training

- It is a procedure through which people are prepared to enter a certain type of professional career, such as one in engineering, medicine, or agriculture. Prior to any appointment, an individual must complete professional training designed to prepare them for starting a new line of work.
- According to Swanson (1984), it is a course of instruction that gets someone ready for a career in extension and typically results in a diploma, certificate, degree, or other credential in one or more of the following fields: agriculture, fisheries, forestry, animal and/or veterinary science, or home science.
- For admittance into their extension services, the state departments of agriculture now prefer university graduates, and the veterinary department prefers to only accept veterinary graduates who have just graduated from an accredited veterinary college.

In-Service Training

It is intended for employed applicants who are currently in service. In-service training is a procedure for staff development that aims to enhance an incumbent's performance in a position with specific work duties. It encourages people to further their careers. The chance to establish a feeling of purpose is offered through in-service training, which is a problem-centered, learner-oriented, and time-bound set of activities. raise the participants' capacity to learn new things and improve their technical proficiency.

Even for university graduates, continuing education is necessary after completing a formal education, according to Arnon (1987). He said that the following goals guide the in-service training.

1. To stay current with research by holding collaborative colloquia, regular meetings with researchers and extension workers, etc.

2. To transmit fundamental information in areas such as sociology, economics, psychology, and others that are not just immediately relevant to agriculture.

3. To enhance extension techniques by ongoing method review, collaboration between research findings and extension methods, and experience sharing.

In-Service training are of different types, some of them are as follows:

i. Orientation Training

This training is given usually to newly appointed extension personnel. It provides an introduction to public employment and provides answers to questions which a newly recruited person is likely to ask. This term is also used for training in-service extension personnel in a new responsibility likes a new operational programme so that personnel are appropriately oriented towards meeting the requirements of new situation.

ii.Induction / portal / vestibule Training

Induction training is given to new extension personnel immediately after they have been employed and before they are assigned to work in particular area usually as an Assistant Agriculture Officer or Agriculture Officer, or Extension Officer.

iii. Maintenance or refresher training

This training is originally started for trainers of the training institutes and Universities for refreshing their knowledge and skills for imparting them to trainees. The term indicates any new training for updating professional competence of extension personnel notably in the subject matter area of specialization. This training is usually imparted in the later career of extension personnel. This training is having considerable importance to extension personnel as it relates to updating to technical knowledge and competence of extension personnel. This deals with new information and new methods and review of older materials. This type of training is given to the employees to keep them at their peak performance level and also prevent them from getting into a rut.

iv. Retraining

It refers to the efforts designed to prepare an individual for a new assignment or a broadened aspect of the old specialty.

v. Career or development training / Training for professional qualification

This type of training is designed to upgrade the knowledge, skills and ability of employees to help them assume greater responsibility in higher positions. This training may lead to the acquisition of higher degree (undergraduate or postgraduate) or diploma by the employees, to motivate them to move up higher levels of administrative hierarchy (promotions).

Training to Farmers

There is a regular farmer training programme in all agricultural universities. There are training centres for young farmers. In some states, they also arrange short courses for the farmers. The training includes crop raising, animal feeding and management, plant protection. For such training the following points should be considered.

Time of holding the training: It should be at the convenience of the farmers i.e., when they are comparatively free from such of the agricultural operations. This will differ according to the seasons and climate. In case T.N March to May for Kharif crop and August to September for rabi crop is ideal time for conducting training courses in Agriculture.

Duration of course: For farmers who are engaged in farming, a one week course is sufficient for special topics such as use of irrigation facilities and water management, operation of implements and plant protection etc, it may be of two or three days duration.

Venue of course: Besides physical facilities, the appropriate environment under which the course is to be conducted i.e, where the farmers car see the actual crop, method demonstrations, operations with some machines and implements or some treatments such as fertilizer application, venue has to be given due considerations.

Production cum demonstration camps and discussion groups of the farmers: These should be arranged in the villages because the farmers cannot afford to remain away from their farms and homes. These should be organized before each main crop. The duration should be 1-2 days only, and the trainees or participants should be from the same village or groups of nearby villages, so that the farmers can walk back to their home the same evening. This will provide technical knowledge to the farmers right in their villages, and the topics can be related to their local problems

Training Process

In case of training, the focus will be on a person-on-the job-in the organization. Whereas in the case of training process, the focus will be both at the starting point and at the end with difference. The application of what a person has learned during training process is called the effectiveness of training.

The training process has three phases as follows:

- Pre-training
- Training
- Post-training

Pre-training phase

- Pre-training process starts with understanding the situation, which calls for behavior that is more effective.
- Key aspect of the process is analysis of situation and job on which improved performance is to be achieved.
- Pre-training begins with description of the job to be changed by it.
- The technical requirement of the job is not enough but also knowledge on operational description of the job is required so that the training programme can be designed to meet out those requirements.

Training phase

Most of the training programmes would be for a session or an evening course or a residential program.

In the training program, the trainee is exposed to a new subject matter, new people, new atmosphere and the participant would be at unease for a while, later when the subject which would be useful and stimulating is taught the participant would focus his attention on the subject of his interest and would be in line with other participants.

There would be several questions in his mind, such that he is lacking, the skill required for his job or is it an opportunity given for his sincere work in the organization or is it a plan of the organization to keep him away from the organization so that it would implement the programme which he had strongly opposed.

With all such questions in his mind, there will be no guarantee that the trainee will learn what he has chosen to learn. His mind would deviate and he would learn something of his interest from the training program provided. This error in selection would be due to the lack of necessary capabilities of the trainee or irrelevant training design and methodology followed by that training institution etc.,

Finally after overcoming all the hurdles in the initial stage of training programme, the participant would explore in training situation what interests him the more. After exploring, if he finds it useful he tries it again and checks for its effectiveness and satisfaction. There would be several trials repeatedly.

If he is satisfied with the results, he decides to incorporate it in his organization, but if he finds it to be not useful he discards it and tries some other variant, in some cases he may discontinue his learning.

Post-training phase

Here the situation changes, the participant goes back to his work place, meets his colleagues, family members etc. He goes prepared with some anticipation; as he had been away from them for a while and also had come back learning some new ideas.

Newly learned skills undergo modifications to fit in with the work situation. If the organization were encouraging and helping, the participant would use his training for the betterment of his organization. Some organization would offer support to the participants to have contact with the training institution even after the training program.

On the other hand, if the organization resented his absence and if his table is loaded with work, he would feel extra burden and would work to make up for lost time. He would loose his interest to make use of his training and the contact with the training institution is also broken off.

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Article ID: 281

SMART FARM MECHANIZATION: REVOLUTIONIZING AGRICULTURE FOR A SUSTAINABLE FUTURE

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Abstract:

Smart farm mechanization is a paradigm-shifting strategy that combines cutting-edge technologies into agriculture to increase production, sustainability, and efficiency. These technologies include robotics, sensors, artificial intelligence, and the Internet of Things (IoT). This article examines the idea of smart farm mechanization, emphasizing its essential elements and advantages. Smart farm mechanization tackles important issues facing contemporary agriculture by maximizing resource utilization, minimizing waste, and facilitating data-driven decision-making. Although it has many benefits, there are also drawbacks to take into account, including start-ups fees and data security. Smart farm mechanization has the potential to change agriculture in an era of rising global population and environmental concerns, providing food security and a sustainable future. With the combination of quantum computing and cutting-edge genetic engineering, smart farm mechanization is set to progress even further in the future, enabling crops and livestock to be precisely adapted to climatic conditions. A further benefit of the widespread adoption of smart farm mechanization is that it may encourage data sharing and collective intelligence for the sustainable management of agricultural resources on a global scale by creating networks of cooperative, interconnected farms.

Keywords: Agriculture technology, Artificial intelligence, Robotics, Internet of Things (IoT), Food security.

Introduction

Agriculture has come a long way from its humble beginnings, evolving into a highly complex and technology-driven industry. One of the most significant advancements in modern farming is smart farm mechanization, a transformative approach that harnesses cutting-edge technologies to optimize agricultural processes (Malhotra & Firdaus, 2022). In a world where the global population continues to grow, and climate change poses unprecedented challenges, smart farm mechanization is a critical tool for ensuring food security, sustainability, and increased productivity (Hanjra & Qureshi, 2010). This article explores the concept of smart farm mechanization, its benefits, and its potential to reshape the future of agriculture.

Agriculture, the cornerstone of human civilization, has undergone a remarkable transformation throughout history (Barker, 1985). From the humble beginnings of manual labor and basic tools to the modern, technologically advanced practices of today, agriculture has continuously evolved to meet the ever-growing demands of a burgeoning global population. Now, in the 21st century, a new chapter in the agricultural revolution is unfolding, driven by a convergence of cutting-edge technologies and innovative practices. Smart farm mechanization, a concept at the forefront of this revolution, stands as a beacon of hope for addressing the complex challenges that agriculture faces in the modern world.

Smart farm mechanization represents a paradigm shift in the way we approach agriculture. (Blackmore et al., 2005) At its core, it is the integration of advanced technologies into every aspect of farming, from planting to harvesting and beyond. This integration extends beyond the use of machines; it encompasses a comprehensive network of sensors, artificial intelligence, data analytics, and connectivity that allows for the intelligent management of agricultural processes.

As our world grapples with critical issues like climate change, resource scarcity, and the need to feed a population projected to reach nearly 9 billion by 2050, the role of agriculture in ensuring a sustainable future cannot be overstated (Chartres & Noble, 2015). The traditional practices that sustained us for centuries are increasingly unsustainable in the face of these challenges. Smart farm mechanization emerges as a transformative solution, offering a way to maximize agricultural efficiency, minimize waste, and reduce the environmental footprint of farming (Malhotra & Firdaus 2022).

In this comprehensive exploration of smart farm mechanization, we will delve into the core principles, technologies, benefits, challenges, and the future prospects of this ground-breaking approach to agriculture. It is a journey into the heart of a revolution that holds the promise of not only feeding the world but also doing so in a manner that safeguards our planet's fragile ecosystems for generations to come.

Understanding Smart Farm Mechanization

Smart farm mechanization refers to the integration of advanced technologies into agricultural machinery and processes to improve efficiency, productivity, and sustainability. This approach leverages various technologies, including robotics, sensors, artificial intelligence, big data analytics, and the Internet of Things (IoT), to create intelligent and automated farming systems. These systems can perform tasks such as planting, harvesting, irrigation, and pest control with precision and minimal human intervention.

Precision Agriculture: Precision agriculture involves the use of GPS technology, sensors, and data analytics to optimize farming operations. Farmers can collect data on soil conditions, crop health, and weather patterns to make informed decisions about planting, fertilizing, and irrigating, resulting in reduced resource wastage and increased crop yields (Zhang et al., 2002).

Autonomous Machinery: Autonomous tractors, drones, and robots are becoming increasingly common on smart farms. These machines can perform tasks with remarkable accuracy, working around the clock if necessary, and reducing labor costs while enhancing efficiency (Ghobadpour et al., 2022).

IoT Sensors: IoT sensors are strategically placed throughout the farm to gather real-time data on soil moisture, temperature, and humidity. This data allows farmers to make timely adjustments to irrigation and other operations, conserving water and energy (Pal et al., 2023).

Artificial Intelligence (AI): AI algorithms process vast amounts of data to provide insights into crop and livestock management. These systems can predict disease outbreaks, optimize feeding schedules, and even detect and control pests and weeds (Mishra, & Mishra 2023).

Benefits of Smart Farm Mechanization

Increased Productivity: Smart farm mechanization enables farmers to produce more with less. Optimized processes, precision farming techniques, and the use of data-driven decisions result in higher crop yields and greater efficiency.

Resource Conservation: The precise application of resources such as water, fertilizers, and pesticides minimizes waste, reducing the environmental impact of farming. This is especially crucial in the face of increasing water scarcity and environmental concerns.

Labor Savings: Automated machinery and robotics reduce the need for manual labor, addressing the labor shortages often faced by the agricultural industry. This can make farming more attractive and sustainable in the long term.

Improved Sustainability: By optimizing resource use, minimizing waste, and reducing the carbon footprint of farming operations, smart farm mechanization contributes to greater sustainability and environmental responsibility.

Data-Driven Decision Making: Access to real-time data allows farmers to make informed decisions about their operations, leading to better outcomes and higher profitability.

Challenges and Considerations

While smart farm mechanization offers numerous benefits, it also presents challenges and considerations. The initial cost of implementing these technologies can be high, potentially limiting access for small-scale farmers. Additionally, there are concerns about data security, as the collection and storage of sensitive agricultural data become more prevalent.

Conclusion

Smart farm mechanization represents a significant step forward in the evolution of agriculture. By harnessing the power of advanced technologies, farmers can increase productivity, conserve resources, and reduce environmental impact. This approach is not only essential for meeting the growing global demand for food but also for addressing the challenges posed by climate change and resource scarcity. As smart farm mechanization continues to advance, it holds the promise of a more sustainable and prosperous future for agriculture. It's not just a technological revolution; it's a necessity for the survival of our planet and future generations.

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Article ID: 282

EFFECT OF GUAVA LEAF ON DIABETES

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Introduction:

P. guajava, popularly known as guava, is also called “the apple of the tropics” or “poor man’s fruit” (as shown in fig 1 and 2), as it thrives in various soils, propagates easily and bears fruit relatively quickly. Guava is the only fruit of the Myrtaceae family with significant commercial. In addition to being a food source, is also an alternative to medicine due to its various herbal properties.

Psidium guajava (L.) is an important fruit in tropical areas like India, Indonesia, Pakistan, Bangladesh and South America. Various studies have shown that guava possesses excellent antioxidant activity due to its high content of ascorbic acid, in addition to having the highest natural concentrations of folic acid, niacin, pyridoxine and dietary fiber and the leaves of the guava plant have many health benefits (anticancer, antidiabetic, antioxidant, antidiarrheal, antimicrobial, lipid-lowering and hepatoprotection), which are attributed to their plethora of phytochemicals, such as quercetin, avicularin, apigenin, guaijaverin, kaempferol, hyperin, myricetin, gallic acid, catechin, epicatechin, chlorogenic acid, epigallocatechin gallate and caffeic acid (Kumar *et al*., 2021).

Table. 1. Nomenclature of *Psidium guajava* L. (Guava):

Superorder	Rosanae
Order	Myrtales
Family	Myrtaceae – myrtles, myrtacces
Genus	*Psidium* L. – guavas
Species	*Psidium guajava* L. – guava, abas, goyavier, guabang, kautonga, kuahpa

Fig. 1. Guava Fruit

Fig. 2. Guava Leaves

Table. 2. Nutritional composition of Guava:

Parameters	Leaves	Seeds	Pulp
Moisture (%)	82.47 ± 2.10^{b}	46.22 ± 1.50^{a}	52.18 ± 1.60^{ab}
Ash (%)	3.64 ± 0.05^{c}	3.15 ± 0.10^{b}	2.42 ± 0.20^{a}
Fat (%)	0.62 ± 0.23^{a}	7.94 ± 1.23^{b}	6.54 ± 1.11^{b}
Protein (%)	18.53 ± 2.29^{b}	13.31 ± 1.34^{a}	10.64 ± 1.66^{a}
Carbohydrates (%)	12.74 ± 1.87^{c}	3.06 ± 1.47^{a}	8.57 ± 1.52^{b}
Vitamin. C (mg)	103.05 ± 4.59^{b}	87.43 ± 5.72^{a}	116.17 ± 6.32^{c}
Total phenolic compounds (mgGAE/g)	1717 ± 6.43^{b}	344 ± 3.77^{a}	383 ±9.32^{a}
Antioxidant activity (%)	234 ± 7.57^{b}	89 ± 6.11^{a}	365 ± 8.65^{c}

(Source: Shabbir et al., 2020)

Chemical composition:

Leaves of guava hold essential oil with the presence of core components such as “α-pinene, limonene, isopropyl alcohol, terphenyl acetate, selinene, caryophyllene, β-pinene, longicyclene, β-bisabolene, farnesene, caryophyllene oxide, humulene, β-copanene, cardinene, menthol and curcumene”. Vital oil from leaves of guava has been identified with the presence of guavavolic acids and as well as with the presence of ursolic, nerolidiol, β-sitosterol and crategolic acid.

Guava leaves include 0.37 % volatile oil, 6.0 % fixed oil, 3.14 % resin, 8.51 % tannin, and other fixed compounds, as well as resin 3.14 %, tannin 8.51 %, and other fixed substances. guava leaves include fat, mineral salts, resin, chlorophyll, tannin, cellulose and volatile oil, with eugenol, mallic acid and tannin ranging from 8-16 % in essential oil. The leaves also contain a vital oil high in cineol, 4 triterpenic acids and 3 flavonoids: quercetin, its 3-L-4-4 arabinofuranoside (avicularin), and its 3-L-4-pyranoside (Rehman *et al*., 2022).

Bioactive compounds and health benefits:

P. guajava is a plant of multipurpose health activities and economic utilizations. Guava and its bioactive compounds are associated with many different and useful behaviours against chronic diseases, such as diabetes, hypertension, and dyslipidaemia as shown in fig. 3. The use of aqueous extract from the leaf of this plant has cardioprotective effects against myocardial ischemia/reperfusion injury. Quercetin maintains the effective functioning of the immune system due to its antioxidant properties. The leaf extract can significantly prevent lipolysis (LPS) induced by by nitric oxide and prostaglandin E2.

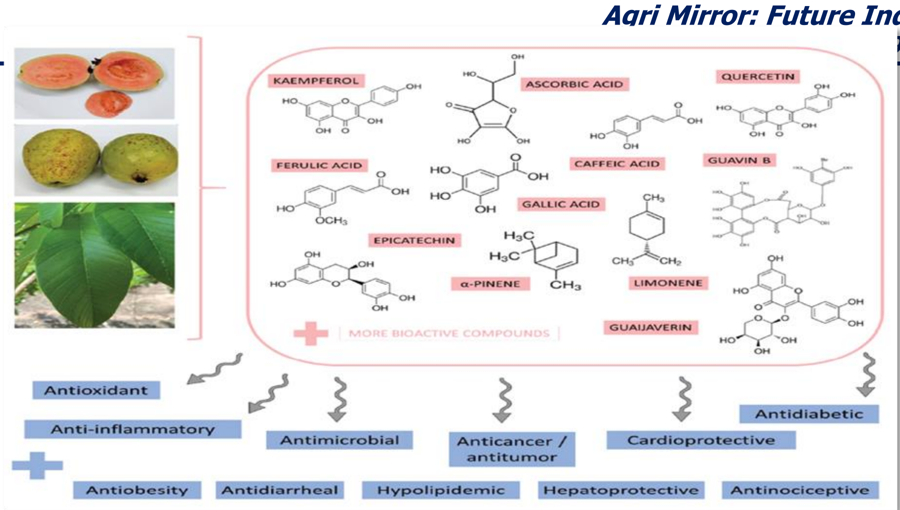

Fig. 3. Representation of Bioactive compounds present in the guava leaf

Role of guava leaf on type II diabetes:

Guava is widely used to treat diabetes. Because it has high levels of triterpenoids in the plant leaves, it is possible to reduce the appearance of diabetic peripheral neuropathy in rats, improving insulin resistance (IR) in adipocytes. Corosolic acid (higher leaf triterpenoid) is a potent inhibitor of α-glucosidase, which delays the breakdown of carbohydrates and reduces the postprandial blood glucose surge (beneficial for the treatment of Diabetes mellitus (DM).

Ethanol leaf extract also has an antidiabetic and protective effect on altered glucose metabolism, in which it reduces blood glucose levels of HbA1c and increases plasma insulin levels.

Zhu *et al*. (2020) Evaluated the guava leaf flavanoid's anti-hyperglycemic and liver protective effects with a high-fat diet and a low-dose streptozotocin induced diabetic mouse model. Flavonoids supplementation significantly decreased fasting plasma glucose, glucose tolerance, and the insulin resistance index in diabetic mice. Results showed that guava leaf flavonoids had significant anti-diabetic and liver protective activities in diabetic mice.

Tella *et al*. (2022) Investigated, the anti-diabetic potential of *Psidium guajava* leaf in streptozotocin-induced diabetic rats. Normal and diabetic animals were treated with 400 mg/kg body weight of guava leaves aqueous extract for a period of 14 days. The leaves extract reduced glycogen phosphorylase expression in diabetic animals. The results from this study suggested that the antidiabetic effects of guava leaf may be due to modulation of glycogen metabolism mediated by phenolic compounds and triterpenes present in the extract.

Conclusion:

Guava leaf extract is proven to be effective to treat diabetes and oxidative stress in animals (Mice/Rats). However, there are little or no human clinical studies that represent anti-hyperglycaemic or anti-diabetic activity of the guava leaf extract, except for traditional uses. Moreover, its underlying therapeutic mechanisms and safety in terms of interaction with other medicines remain unclarified. So further experimental and clinical trials are required to discover the dosage, toxicity and effect of guava leaf extract on humans.

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Article ID: 283

EFFECT OF WATER STRESS ON BIOCHEMISTRY OF HORTICULTURAL CROPS

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Abstract:

Climate change is intensifying both abiotic and biotic stressors in agriculture, with drought emerging as a prominent threat. Drought adversely affects plant growth and crop yields, compounding challenges in food production. Understanding the biochemical responses of plants to water stress is essential. Abscisic acid (ABA) plays a pivotal role in regulating these responses, leading to significant alterations in gene expression and the production of metabolites like amino acids, sugars, indoles, phenolics, and glucosinolates. To combat nutritional deficiencies, there's a growing imperative to enhance phytochemical levels in crops, achievable through exposing them to mild environmental stress during their growth stages. Carbohydrates, specifically starch and sugar content, are critical determinants of crop quality. Water stress can reduce starch accumulation due to lowered sucrose levels and enzymatic activity. Conversely, total soluble solids and soluble sugars accumulate under water stress, serving as osmotic agents and antioxidants. Lipids, vital for cell membranes and storage, are vulnerable to oxidative damage under drought, potentially altering membrane fluidity and protein function. Mineral concentrations in crops are influenced by factors such as genetics, environmental conditions, and soil properties, all influenced by drought. Lastly, secondary metabolites like polyphenols, pigments, ascorbic acid, and tocopherol play various roles in plant stress adaptation, with their levels responding to drought depending on factors like crop type and timing of stress. Understanding these dynamics is crucial for improving crop quality and nutrition in a changing climate.

Keywords: Water Stress, ROS, Structural carbohydrate, Carotenoid

1. Introduction

Abiotic and biotic stress factors have a significant negative impact on agricultural yields and are spreading more widely as a result of the effects of climate change. The main abiotic element is drought, which has an impact on plant growth and development and limits the production of agricultural crops. In the coming years, problems with water shortages and declining irrigation water supply may become more prevalent due to rising agricultural water needs, contamination of natural water resources, and climate change scenarios.

The effects of water stress on plant have several biochemical changes. The primary regulator of these pathways is the phytohormone abscisic acid (ABA), which accumulates in response to osmotic stress (Wang *et al*., 2015). Plants respond to drought by activating a number of pathways, which results in a significant change in gene expression and plant metabolome as a result, some metabolites, including amino acids, sugars, indoles, phenolics, and glucosinolates, have their production significantly enhanced. It is now more necessary than at any time to improve the nutritional and health benefits of food crops to prevent dietary deficits. So, increasing some phytochemicals and improving the nutritional value of crops while maintaining crop yields may be possible by introducing mild environmental stress during the crop growth season.

2. Carbohydrates

Among carbohydrates, two components play a particular role as determinants of the quality of the final product: (i) starch and (ii) sugar concentration (Wang and Frei, 2011).

2.1 Starch

Drought stress during vegetative growth lowers the amount of carbohydrate accumulated in the stems and leaves determining a seed mass constraint, since the biomass produced in such organs from the vegetative to reproductive stages is needed to build up enough resources for translocation; water limitation occurring during the reproductive phases directly constrain seeds development, causing a premature grain desiccation (Fábián *et al*., 2011). The starch accumulation is strongly dependent on the seeds' sucrose concentration and on enzyme activity (e.g., sucrose synthase) involved in starch synthesis (Saeedipour and Moradi, 2011). This suggests that, under water stress, both a low sucrose content and decline in enzymatic activity are responsible for starch reduction.

In starchy tuber crops, a decrease in starch concentration in response to drought has been observed for cassava (Vandegeer *et al*., 2013) and potato (Ballmer *et al*., 2012) where the inhibition of starch biosynthesis determines a higher level of sugars (2- to 8-fold) and a two- to three-fold stimulation of sucrose synthesis in tubers, indicating a change of AGPase expression as a direct inhibition of cell-wall synthesis.

2.2 Total soluble solids and soluble sugars

Water stress has been proven to induce an accumulation of total soluble solids (TSS) in many crop plants as an important adaptation mechanism (Babita *et al*., 2010). Compatible soluble like sugars, glycerol, proline, or glycine betaine can also contribute to turgor maintenance (i.e., accumulation of osmotically active solutes) as fundamental physiological means for lowering the negative effects of drought (Aldesuquy *et al*., 2012).

Among the compatible soluble, soluble sugars can act: as osmotic agents and as osmoprotectors (Huan *et al*., 2014). As an osmotic agent, water stress-increased sugar was significantly correlated with osmotic adjustment. As osmoprotectors, sugars stabilize proteins and membranes, substituting the water in the formation of hydrogen bonds with polypeptide polar residues and phospholipid phosphate groups; besides, under drought soluble sugars accumulate to also former reserve assimilates for seed filling.

At low moisture conditions, sugar levels increase in several crop species such as tomatoes (Wu and Kubota, 2008), cucumbers (Huang *et al*., 2009), and grapes (Deluc *et al*., 2009). In red beet (*Beta vulgaris* var. *conditiva* Alef) a decrease in total sugar concentration has been reported under moderate and severe stressful conditions (Stagnari *et al*., 2014a), to support vegetative tissues' current growth rather than storage of reserves.

3. Lipids

Lipids represent both structural components of cell membranes and storage products. Under drought conditions, the production of reactive oxygen species (ROS) is detrimental to lipids as they induce the production of various oxidative radicals that cause oxidation (Ahmadi *et al*., 2014). As a consequence, the composition of lipids in the plasma membrane varies strongly, especially in terms of modifications in the fatty acid (FA) composition of membranes (Laribi *et al*., 2009). These changes affect the fluidity and also intrinsic-membrane protein activities as a result of alteration in the lipidic environment in which they are embedded (Thomas *et al*., 2013). In addition, water limitation can also influence the synthesis of the essential oils and their composition (Laribi *et al*., 2009). Consequently, the interaction between water stress and oil concentration of crops has been principally investigated in crops that contain oils as well as essential oils (Wang and Frei, 2011).

4. Mineral

According to (Martnez Ballesta *et al*.2010), genotypes, environmental growing circumstances, soil characteristics, and crop maturity at harvest all affect the amount of minerals present in crops. While numerous research has concentrated on the connections between the availability of soil minerals and the mineral concentration of the crop harvested organs, the impact of environmental abiotic stress (non-nutritional stress) on the mineral concentration of crop has largely gone undiscovered. Drought is crucial because the availability of water in the soil directly affects how plants absorb minerals. On the other hand, it can inhibit acropetal translocation of nutrients due to decreased transpiration rates, impair active transport, and damage membrane permeability (Misle *et al*., 2014). In particular, (i) arid zone soil is often calcareous and characterized by high pH values (e.g., Mediterranean regions) or rich in aluminum and iron oxides with low pHs (e.g., semi-arid tropics), and (ii) a great P fixation is observable in these soils. (iii) Water content in soil is directly correlated with soil $K+$ mobility. Due to these factors, the regulation of fertilization has been the focus of the majority of investigations.

5. Secondary metabolites

More than 100,000 secondary metabolites (Edreva *et al*., 2008) are specific compounds of some taxonomic groups that are not required for an organism to survive but play a role in how the cell (organism) interacts with its environment. They are distinguished by a wide chemical diversity, including aliphatic, aromatic, hydroaromatic, and heterocyclic; unique carbon skeletons occur along with multiplicity of functional groups. Their quantities in plant products are highly influenced by the environment during growth, particularly stressful conditions like drought (Kannan and Kulandaivelu, 2011). To increase productivity and recover "phytochemicals," it is crucial to understand how water availability affects growth, biomass partitioning, and secondary metabolite concentration in plant tissues and organs.

5.1 Polyphenols

Polyphenols originate from the phenylpropanoid biosynthetic pathway and are involved in plant adaptations to biotic and abiotic stresses. They are known to participate in the defense against reactive oxygen species (ROS) having strong antioxidant activities and, thus, are almost always produced when environmental stresses impair photosynthetic reactions, through the stimulation of the phenylalanine ammonia lyase (PAL) or other key enzymes (Oh *et al*., 2009).

In *Brassicaceae* species the influence of water deficit on phenolic content is strictly related to the time at which the stress condition occurs. For broccoli, significant increase of polyphenolic concentrations by 35.5% due to drought (Fortier *et al*., 2010), even though the influences of genotype, maturity at harvest, weather, cultivation, and storage conditions are significant (Podsedek, 2007). Conversely, under greenhouse conditions, the content of phenolic compounds seems to be lower by low soil water content. Young lettuce plants (4 weeks old) exposed to mild drought respond with a significant increase in their total phenolic concentration and antioxidant capacity (Oh *et al*., 2010); this was also found for various lettuce genotypes reaching commercial maturity (Eichholz *et al*., 2014). Plants of *Cucumis sativus* L. exposed to different water regimes result in significantly increased anthocyanin content (Sonnenberg *et al*., 2013) due to the stimulation of anthocyanin hydroxylation, by upregulating the gene encoding its enzyme, or as a result of increased early accumulation of sugars (Castellarin *et al*., 2007).

5.2 Pigment

The effects of water constraint on carotenoid accumulation have been extensively investigated due to their protective role against ROS, since they are very efficient physical and chemical quenchers of 1O2 (Esteban *et al*., 2015) however, to date, tomato is the most studied species (Stefanelli *et al*., 2010), due to its lycopene content in fruits (Dumas *et al*., 2003). This is because of the protective role that carotenoid accumulation plays against ROS and the fact that they are very effective physical and chemical quenchers of $_1O^2$ (Esteban *et al*., 2015). In general, water availability restrictions regularly raise the concentrations of lycopene and total carotenoids in fruits but have no impact on the contents of -carotene or xanthophyll. The amount of lycopene and beta-carotene in processed tomato increases dramatically with longer watering intervals and a reduced irrigation regime.

In earlier studies, moderate water stress had already been shown to enhance lycopene, especially in the outer pericarp of red and pink large-fruited tomatoes (Serio *et al*., 2006). sweet potatoes also showed a similar pattern (Rautenbach *et al*., 2010). On the other hand, other research shows that as moisture stress increases, the concentrations of carotenoids and lycopene in tomato fruits generally decrease (Riggi *et al*., 2008). However, betaxanthins (yellow-orange betalains) and betacyanins (red-violet betalains) have recently attracted attention for their antioxidative activities that were proven to surpass even the values of classical dietary antioxidants such as ascorbic acid, rutin, and catechin. While the betacyanin/betaxanthin ratio appeared unaffected by water restrictions, red beet was found to increase in both betaxanthin and betacyanin concentration (Stagnari *et al*., 2014). This finding suggests that betalains accumulate because they may serve as osmolytes to support physiological processes similar to the amino acid proline.

5.3 Ascorbic acid and Tocopherol

Compared to phenolics, fewer data are available on the effects of water limitation on ascorbate (vitamin C) accumulation in crops and many of them are often inconsistent.

According to various research on tomatoes, a lack of water has led to an increase in ascorbate levels (Patanè *et al*., 2011). The concentration of vitamin C increases in particular when irrigation threshold is lowered to 20 and 40 mm (Favati *et al*., 2009). However, it should depend on the cultivar because prior studies show that under drought, yield increases and vitamin C and soluble solids content decreases (Dumas *et al*., 2003). It's probable that the more apparent canopy present in tomato plants that have received full watering will produce a more suitable fruit cover and shadowing, which is associated with a drop in the ascorbate content of fruits.

In table grapes the partial root zone drying is an effective practice to increase vitamin C content by 15–42% (Du *et al*., 2008) as well as the concentration of ascorbic acid and ellagic acid in strawberry (Dodds *et al.,* 2007). Ascorbic acid content builds up in mandarins when deficit irrigation is induced during phase II of the fruit growth (Navarro *et al*., 2010). These changes to the allowing for variable ascorbate concentration findings revealed that the complexity of the Plants' ascorbate metabolism is controlled by two (i) De novo synthesis of ascorbate from its precursor glucose-6-phosphate using GDP-mannose as an intermediary and l-galactose; (ii) ascorbate recycling through Dehydroascorbate is converted from its oxidized form to its reduced form via the ascorbate-glutathione cycle (Sade *et al*., 2013).

Conclusion

Crop quality attributes are impacted by drought in conjunction with genetic, physiological, and environmental factors. Additionally, a multifactorial interaction happens in every experiment: For instance, a water shortage is frequently connected to greater salt levels or with a high temperature. As a result, in the corresponding results are not always there in many circumstances. However, a clear general stimulation by a lack of soluble sugars, proteins, and secondary metabolites, or lower quantities of starch and lipids, may be deduced regarding the accumulation of primary and secondary metabolites. Water-stressed plants may be a source of bioactive chemicals, notably concentrated in tissues and with a limited biomass production, in relation to the growing interest in functional meals. This knowledge can be used to develop agronomic solutions (such as deficit irrigation) that will increase crop quality traits while minimizing adverse impacts on yield or to improve the quality of crops originating from drought-stressed areas.

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Article ID: 284

GENETIC IDENTIFICATION AND MASS PROPAGATION OF ECONOMICALLY IMPORTANT SEAWEEDS

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Abstract:

Seaweeds are a primary source of hydrocolloids, which can be processed into various food additives, cosmetics, and pharmaceuticals. The inability of current commercial seaweed farming projects to meet industrial demands is underscored by a plethora of challenges, which include the lack of high-quality germplasm with the desired cultural characteristics. Molecular markers have become increasingly relevant to the selection of a diverse range of wild varieties for domestication, and this augurs well for strain identification. The development of high-density linkage maps based on molecular markers offers an avenue for the implementation of molecular breeding strategies based on quantitative trait loci (QTLs). Concurrently, the productivity of existing varieties can be enhanced by the analysis of exogenous factors known to affect the growth and survival of tissue-cultured seedlings. The application of photobioreactors for tissue culture is another important development, which will be digressed upon. In addition to this, quality control which focuses on the comparison of chemical and physical qualities of the tissue cultured and conventional cultivated seaweeds will become increasingly relevant to the development of industry standards for sustainable seaweed production to fulfil the increasing demands of seaweed-related industries.

Keywords: Seaweed, Acclimatization, Genetic marker, Marker-assisted selection, Photobioreactor, and Tissue culture.

1. Seaweeds and their economic importance

Seaweeds are marine macroalgae, Generally found living in oceans and coastal areas throughout the world. The classification of seaweeds has been typically based on their phenotypical features, which include pigmentation and photosynthetic properties. Since the mid-nineteenth century, seaweeds have been empirically distinguished into three main divisions based on their colour; Red algae (phylum: Rhodophyta) consists of about 6000 species, Brown algae (phylum: Ochrophyta) consists of about 1750 species, and Green algae (phylum: Chlorophyta) consist of about 1200 species. Recently, a wide variety of seaweeds and their products have been studied for their industrial, culinary, and renewable energy applications, which include cosmetics, chemistry, paint, medicine, biofuel, etc. Increasing global demand for seaweed resources and overexploitation of natural seaweeds have highlighted the need for sustainable seaweed cultivation to significantly increase captive production in mariculture systems.

Among all the seaweed based products, hydrocolloids viz. carrageenans, agar, and alginates continue to be the principal extracts, which received commercial attention through their application in various industries. *Kappaphycus* and *Gracilaria* are the two most important red seaweeds in the world trade market, which have been reported to contribute a significant amount (60–80%) of the world's carrageenan and agar resources, respectively. Seaweed hydrocolloids are mainly used in food processing industries as is evident from their applications in health care products such as carrageenan gel capsules and alginate microbeads. Apart from the hydrocolloid industry, seaweed natural products are also a promising source of biologically active compounds with medical and pharmaceutical applications and nutritional supplements for animals and plants.

2. Current limitations of conventional seaweed breeding and alternative solutions

Emerging applications of hydrocolloids in the food industry and other hydrocolloid-related industries have led to an enhancement of the economic values of red algae including *Kappaphycus, Eucheuma*, and *Gracilaria* species. Production of seaweed biomass has globally increased from 4 million wet tones in 1980 to 20 million wet tones in 2010 with 95.5% of biomass produced from artificial cultivation in mariculture program. The economic viability of the seaweed industry is dependent on the production of high-quality germplasm with the desired traits viz. high growth rates, amenability to treatment with fertilizers, and resistance to diseases for incorporation into the breeding program. The high variation of morphological features in the wild and the lack of diagnostic morphological characters have resulted in the misidentification of cultivated seaweed strains, which in turn have contributed to a decline in the productivity and quality of cultured seaweeds.

Current practices of seaweed cultivation are predominantly based on traditional methods, whereas seaweeds are exposed to environmental challenges and pathogens. Most of the seaweeds are cultivated using seedlings produced by vegetative propagation from cultured germplasm. Through this practice, parasites or pathogens from the harvested seaweeds may be reintroduced and subsequently reduce the productivity of the farm. The other logistical problems faced by conventional seaweed farmers include the identification of appropriate sites for farming, and labour intensive tasks such as inspection, disease, and seedling losses resulting from extreme weather conditions and water quality. In order to increase productivity, modern biotechnology via tissue culture can be considered as one of the best options to overcome the conventional breeding challenges including shortage of raw material for planting and seedlings destruction by epiphytes, subsequently facilitating the propagation of high-quality seaweeds.

3. Application of molecular markers in seaweed breeding

The establishment of an effective seaweed breeding program is founded on the selection of strains of seaweed with desirable cultural characteristics such as high growth rates, high carrageenan content, disease resistance, and accelerated growth in response to supplemental fertilizers. Phenotypical identification methods are currently the standards by which specific seaweed strains are selected. Although invaluable, morphological characterization can be time-consuming and requires a high level of expertise to discriminate key morphological features indicative of the seaweed species. In addition to this, the physical characteristics of seaweeds tend to be variable as they are directly influenced by environmental factors. Most of these seaweeds cannot be distinguished on the basis of one or collection of specimens. Using morphological characters alone, and an exhaustive taxonomic study is essential before the variety can be identified. For example, high morphological plasticity within the Hawaiian Eucheuma seaweeds has led to the misidentification of three introduced eucheumatoid species. Commercially important seaweed species are selected on the basis of the types of biopolymers that they synthesize, where the infrared spectroscopy of their gels has become a measurement to differentiate among genera and species. Nowadays, morphological data have to be complemented with molecular data in order to characterize the desired species of seaweed. Molecular markers are an ideal tool for the classification of cultivated and wild seaweeds independent of their morphological appearance and growth stage. Their application can be extended to Marker Assisted Selection (MAS) and the development of isogenic strains for application in current and future propagation programs. The development of molecular markers for germplasm will be useful for species and variety specific identification, plant variety protection, and interspecific and intergeneric crosses development for economically important seaweed species.

4. Genetic marker for identification of commercially important seaweed species

The application of different genes for the genetic identification of seaweed species is widely carried out, where the targeted DNA regions are the nuclear, plastid, and mitochondrial DNA (mtDNA). Most molecular characterization targets seaweeds with economic value such as *Palmaria palmate* (Dulse), *Porphyra umbilicalis* (Nori), *Gracilaria changii*, *Kappaphycus alvarezii, K. striatus*, *Eucheuma denticulatum* (carrageenophytes), and many more. Currently, large scale DNA barcoding such as Red Algal Tree of Life Initiatives (RedToL) is analysing the phylogenetic relationship of 471 red seaweeds using two nuclear, four plastids, and two mitochondrial encoded gene markers. China has also conducted a large scale phylogenomic analysis of marine red algae revealing evolutionary lineages for rhodophytes. The quality of DNA barcoding locus should have adequate internal variability to enable differentiation at the species level and contain flanking regions that are conserved enough to study routine amplification across highly divergent taxa.

Nuclear ribosomal regions, which include sequences of large subunit (28S), small subunit (18S), and the intergenic transcribed spacers (ITS1, ITS2), can be served as target sites for molecular markers because the ribosomal DNA (rDNA) genes contain both highly conserved and variable regions that can be used as diagnostic tools for certain organisms. The small subunit (18S) and the large subunit (28S) regions are the most used regions for marker development as they are best suited for inference at high taxonomic levels. However, ITS region is often targeted for intraspecific genetic studies in Chlorophyceae (Codium fragile), Phaeophyceae (*Fucus serratus, F. evanescens*), and Rhodophyceae (*Chondrus crispus, Ulva intestianlis, U. compressaa*) on account of its high rate of nucleotide substitution, permitting comparison between relatively diverged taxa. *Hu et al*., 2010. had also used the ITS1 region to study the intraspecific relatedness among 59 Porphyra yezoensis (Nori) for a selective breeding program of economically important nori crops.

mtDNA has a higher mutation rate that gives rise to variation in its DNA sequence. mtDNA is usually used to analyze the phylogenetic relationships of groups within a species or individuals that are closely related. The gene map of mtDNA of the red alga, *P. purpurea*, is available, where all the different genes have been successfully sequenced. The mtDNA loci, which are generally targeted in seaweed identification, are cytochrome oxidase subunit I (cox1), cytochrome oxidase subunit 2–3 intergenic spacer (cox2–3 spacer) and cytochrome b (cytb) genes. Tan et al.,2012. had evaluated the effectiveness of three mtDNA markers, cox1, cox2, and cox2–3 spacer in barcoding the two commercially important carrageenophytes, *Kappaphycus* and *Eucheuma* seaweeds, and has determined that the cox2–3 spacer DNA marker is more suitable as a barcoding gene because its widespread use. Recently, Lim et al. had found higher species diversity of Kappaphycus seaweeds in Southeast Asia (Malaysia, Indonesia, Philippines, and Vietnam) using the mitochondrial cox1 and cox2–3 spacer.

Loci derived from the chloroplast genome (cpDNA) can be used for the identification of seaweed species due to the low frequency of structural changes and low sequence evolution rate of cpDNA. The cpDNA loci that are routinely used for seaweed identification are the ribulose-1, 5-bisphosphate carboxylase/oxygenase (RuBisCo) gene, specifically the large subunit of the RuBisCo (rbcL) and the RuBisCo spacer. In a study conducted by Geraldino et al. molecular phylogeny of 23 specimens of red alga, Hypnea flexicaulis from three countries (Korea, Taiwan, and the Philippines), was successfully studied based on the plastid rbcL region. Guillemin et al. utilized the RuBisCo spacer region to identify six species of Gracilariaceae: G. gracilis, G. conferta, G. dura, G. multipartite, G. vermiculophylla, and G. longissima, which exhibit a high degree of phenotypic similarity.

Identification of commercially important seaweeds based on standard DNA barcodes or single marker amplification has proven to be useful as the phenotypic plasticity of the species can confound traditional taxonomic approaches. Table 1 showed the summary of nuclear, mitochondrial, and plastid DNA regions that are used for the identification of rhodophyta. Molecular markers are still valuable, despite the increasing popularity of next-generation sequencing technologies, where the identity of an unknown seaweed species can be acquired based on a simple polymerase chain reaction amplification and a single sequence read (two sequence reads if both strands are sequenced). Examples of DNA markers used to identify commercially important seaweeds are given in Table 2.

Table 1. Summary of nuclear, mitochondrial and plastid DNA regions used for identification in Rhodophyta

Sl.No.	DNA Regions	Abbreviation	Size (bp)
	Nuclear DNA		
1.	Small subunit ribosomal DNA	SSU	1800
2.	Internal transcribed spacer ribosomal DNA	ITS	650–1100
3.	Large subunit ribosomal DNA	LSU	2700
	Plastid DNA		
4.	Photosystem I P700 chlorophyll a apoprotein A1	psaA	1600
5.	Photosystem I P700 chlorophyll a apoprotein A2	psaB	1250
6.	Photosystem II thylakoid membrane protein D1	psbA	950
7.	Plastid LSU (23S) domain V	UPA	370
8.	Ribulose-1,5-bisphosphate carboxylase large subunit	rbcL	1350
	Mitochondrial DNA		
9.	Cytochrome c oxidase subunit I DNA barcode region	COI-5P	664
10.	Cytochrome c oxidase subunit I extended fragment	COI	1232
11.	Cytochrome b	COB	940
12.	Cytochrome oxidase subunit 2-3 intergenic spacer	cox 2–3	350–400

Table 2. Examples of DNA markers used for seaweed identification

Sl.No.	DNA Marker	Primer	Primer sequence
	Nuclear DNA		
1.	SSU ribosomal DNA	Forward primer Reverse primer	5′-CAACCTGGTTGATCCTGCCAGT-3′ 5′-TGATCCTTCTGCAGGTTCACCTAC-3′
2.	ITS ribosomal DNA	Forward primer Reverse primer	5′-TCGTAACAAGGTTTCCGTAGG-3′ 5′-TTCCTTCCGCTTATTGATATGC-3′
	Mitochondrial DNA		
3.	cox1	COXI43F COXI1549R	5′-TCAACAAATCATAAAGATATTGGWACT-3′ 5′-AGGCATTTCTTCAAANGTATGATA-3
4.	cox2-3 spacer	Cox2_for Cox3_rev	5′-GTACCWTCTTTDRGRRKDAAATGTGATGC-3′ 5′-GGATCTACWAGATGRAAWGGATGTC-

			3′
	Plastid DNA		
5.	rbcL	F7 R753	5′-AACTCTGTAGTAGAACGNACAAG-3′ 5′-GCTCTTTCATACATATCTTCC-3
6.	RuBisCo spacer	Forward primer Reverse primer	5′-TGTGGACCTCTACAAACAGC–3′ 5′-CCCCATAGTTCCCAAT-3

5. Marker-assisted selection (MAS)

Marker-assisted selection is defined as an indirect selection method of an individual with desired traits in a breeding program based on DNA markers. The important of MAS in a seaweed breeding program is to obtain basic genetic knowledge of the chosen commercially important seaweed. Some desired seaweed traits, such as crop yield or phycocolloid content, may be controlled by one gene or a group of genes. Therefore, it is beneficial to develop markers for a range of commercially important seaweed species to provide the foundation needed for MAS in the seaweed breeding program.

In seaweed farming, specifically for the phycocolloid industry, the desired traits of seaweed would be disease resistance, suitable carrageenan content, high productivity, and yield. These desired characteristic or traits can be genetically simple, where only one gene is involved. However, most economically important crops tend to have traits that are genetically complex, where it is controlled by many genes (QTL) and the environment. For example, *Babu et al*.,2003. had detected a total of 47 QTLs for drought resistance traits from various plant water stress indicators to increase the production and yield of rice in rainfed agriculture ecosystems. To date, there are no reports in the literature on the application of MAS in seaweed breeding programs. Recently, *Maili et al*.,2016. He had successfully developed eight out of 112 single loci DNA markers to discriminate between varieties *of K. alvarezii, K. striatus*, and *E. denticulatum* seaweeds, where the markers could be applied in MAS and hybrid development. In future, application of DNA markers in MAS could be used as a tool that can help seaweed breeders to select more efficiently for desirable traits for the improvement in the culturing method of seaweed.

6. Mass propagation of seaweed seedlings via tissue culture

Repeated vegetative propagation applied in conventional seaweed cultivation was found to decrease the genetic variability of seaweeds and subsequently contribute to the decrease in growth rates and yields and increased susceptibility to diseases. Micropropagation via tissue culture technology has been proposed as an alternative method compared to conventional breeding to resolve the seedling shortage problem and increase the productivity of seaweed raw materials. Micropropagation is a versatile tool to produce a high number of uniform specimens from selected strains with desirable characteristics and increase seed stock production in a shorter period of time. However, challenges including lack of optimized protocols to obtain axenic cultures and regeneration of explants have limited the widespread use of tissue culture technology in commercial seaweed production. The efficacy of seaweed tissue culture depends on the effective manipulation of endogenous (age, source, developmental stage, and physiological state of explants) and exogenous (media composition, light, salinity, pH, and temperature) factors. Current research has been strategized to improve the culture conditions for the mass production of high-quality laboratory seedlings to enhance the overall productivity of seaweed cultivation.

7. Preparation of axenic cultures

Explants have to be sterilized in order to obtain the axenic cultures for mass propagation in tissue culture. Seaweed samples collected from the wild are associated with a significant level of biological contamination, which is likely to be commensal or symbiotic; therefore, it is necessary to surface sterilize the explants with general disinfectants as well as targeted antibiotics prior to cultivation. Povidone iodine and alcohol are common disinfectants used for surface sterilization as they have a localized activity compared to the narrow spectrum antibiotics with their functionality limited to specific classes of microbes. Surface sterilization of seaweeds is difficult as they lack of thick protective surface, and therefore, sodium hypochlorite and similar agents can easily damage the tissues, especially newly regenerated thallus. Prolonged exposure of explants to excessive disinfectants (e.g., more than 5 min in 2% betadine and more than 72 h in 5% antibiotic mixture) was reported to cause patches of damaged surface on thallus and explants.

8. Media composition

Culture media commonly used for rapid propagation of rhodophyte are reported to be Provasoli's enriched seawater (PES), seawater supplemented with von Stosch (VS) solution, and seawater enriched with half strength "f medium" (F/2). The selection of culture media for seaweed propagation is highly dependent on the nutrient level, ambient water, and cultured species. The optimized culture medium for economically important *K. alvarezii* was discovered to be seawater enriched with 50% of PES solution, whereas enrichments with 50% of VS and 50% of F/2 solutions were found not effective for *K. alvarezii* cultures. Besides, *G. changii* cultured in 25% of PES-enriched seawater was revealed to propagate well and demonstrated a promising growth rate. Although some rhodophytes have been reported to grow well in VS and F/2 media, the difference in media used may be due to the source of different explants or different genotypes of explants. PES medium has a low concentration of nutrients, whereas F/2 and VS media have a higher concentration of salts which may interfere with the growth of *K. alvarezii*. The F/2 medium is literally formulated for growing coastal marine algae, especially diatoms, while the VS medium is developed for culturing and investigating the life cycle of the freshwater red algae *Bangia atropurpurea*

9. Plant growth regulators

The addition of plant growth regulators and their role in seaweed tissue culture have been extensively reviewed. Cellular competence to plant hormones in cultivated seaweeds is significant only if the cells possess the ability to perceive, transduce, and respond to the hormonal signal. The common plant hormones used in seaweed tissue culture are auxins (indole-3-acetic acid, 2,4-dichlorophenoxyacetic acid), cytokinins (benzyadenine, isopentenyladenine, kinetin), and gibberellins (gibberellic acid). The presence of phytoregulators in tissue culture medium is known to be able to stimulate tissue elongation and contribute to overall plant growth. Generally, auxins are used to increase protein synthesis, induce morphogenesis, and elicit changes in the genetic expression of explants, while cytokinins are used to stimulate cell division, enhance metabolic activities, and affect cell differentiation in seaweed tissue cultures. A combination of auxins (α-naphthalene acetic acid and phenylacetic acid) and cytokinins (N6 -(2-isopentenyl) adenine and 6-benzyl amino purine) has been reported to induce the highest callus growth in *K. alvarezii*, whereas indole-3-acetic acid and 6- benzylaminopurine in their combination have been revealed to stimulate the regeneration process of *K. alvarezii*

10. Organic fertilizers and biostimulants

The organic requirements for axenic seaweed culture are remained unclear although additions of organic complexes (coconut milk, yeast, and algal extracts) to increase the growth rates of seaweed tissue have been reported. Three commercially available formulated fertilizers and biostimulants in the global market for seaweed cultivation are Acadian marine plant extract powder (AMPEP), Gofar600 (GF), and natural seaweed extract (NSE). AMPEP is extracted from Ascophyllum nodosum, while GF and NSE are the mixture extracts of brown seaweeds including A. nodosum, Sargassum sp., and Laminaria sp. in different ratios of concentration. The use of AMPEP was first reported to successfully induce the regeneration of young plants from different varieties of Kappaphycus seaweed. Other studies have also highlighted the positive influence of AMPEP application on the growth and health of *K. alvarezii* cultures both in vitro and in the field. Brown seaweed extracts as contained in AMPEP have been discovered to potentially activate the natural defense system of *K. alvarezii* against pathogens and ameliorate the negative impacts of exposure to oxidative bursts, which may result in bleaching of the thallus.

11. Exogenous factors affecting seaweed tissue culture

Studies on optimizing the growth of economically important seaweeds, especially *Kappaphycus* and *Gracilaria spp*., in tissue culture conditions can help to mass propagate these viable species for continuous, steady, and defined production, while circumventing the barriers of seasonality and environmental vagaries in seedlings production. Several protocols for callus induction and thallus regeneration of a wide variety of seaweeds are available in the works of literature. A number of studies have also reported the direct regeneration of micro-propagules from the explants of red algae for maintenance and clonal propagation of maricultural stock. Apart from media composition and supplementation of phytoregulators and fertilizers, the abiotic factors determined to have significant effect on the growth of seaweed tissue cultures are reported to be light intensity, aeration activity, salinity, and pH. The daily growth rate (DGR) of seaweeds in tissue culture optimization was measured and calculated as DGR = $[(W_t / W_0)^{1/t} - 1] \times 100\%$, where W_0 is the initial fresh weight, and W_t is the final fresh weight of the seedlings after t days of culture.

12. Light intensity

Light source is one of the most important parameters to be optimized in seaweed cultivation. The intensity, wavelength, and spectral quality of light, all influence the photosynthetic productivity of algae. Different strains or varieties of seaweed may exhibit different optimum growth ranges and tolerance to different light resources. *K. alvarezii* strains from Sabah, Malaysia, have been discovered to achieve optimum growth under photon flux density of 75 μmol photons m^{-2} s^{-1} with DGR of 4.3 ± 0.5% day^{-1}. Various forms of *K. alvarezii* and *E. denticulatum* from the Philippines have been reported to grow under irradiances of 25–160 μmol photons m^{-2} s^{-1} with optimum growth in 60 μmol photons m^{-2} s^{-1}, while two different strains of *E. denticulatum* and *K. striatus* from Southern Japan have been revealed attained highest growth rate under light irradiance of 145 μmol photons m^{-2} s^{-1}. Light intensity in the range of 5–100 μmol photons m^{-2} s^{-1} is commonly used for tissue culture of *K. alvarezii* and other phycocolloid yielding seaweeds. Explants were found to respond to higher light intensity by producing more buds and subsequently leading to an increase in biomass. However, a further increase in light intensity was found to be a detriment to the growth of seaweeds as this might be due to the effect of photoinhibition. Nitrogen concentration in seaweed tissues was reported influenced by light intensity, where the increase of light intensity beyond a critical level has resulted in the decrease of crude protein content in seaweeds.

13. Aeration activity

Aeration is an ideal method of energy transfer; whereby, atmospheric carbon dioxide is diffused into the culture medium. Continuous aeration is an important process to provide enough carbon dioxide for carbon fixation in seaweed metabolism. Meanwhile, carbon source can be provided in the organic form such as glycerol yet the addition of glycerol in the culture medium was found to reduce the morphogenetic capacity and totipotency of the explants. Continuous aeration (at 30.0 L h^{-1}) was found to significantly enhance the growth of *K. alvarezii* with an optimum DGR of 4.2 ± 0.3% day^{-1}. Growth of *Gelidium pulchellum*, another red alga, was reported to achieve maximum rate under continuous light with the support of aeration activity. Mixing of the culture can be accomplished by different mechanisms such as aeration, sparging, pumping, mechanical agitation, or a combination of these depending on the scale and type of cultivation systems. Agitation by mixing is considered as the key parameter for the equal distribution of nutrients and cells in the liquid phase, and maintenance of their uniform concentration to increase the mass transfer rate. Growth of cultures is normally enhanced under aerated conditions provided the other culture requirements (light, photoperiod, salinity, pH, temperature) are remained constant. Appropriate aeration activity may increase thallus exposure to light, eliminate nutrient diffusion barriers, improve gas exchange, and facilitate heat transfer to avoid thermal stratification.

14. Salinity

Salinity is reported to be one of the factors affecting the growth and exerting strong influences on the photosynthetic capacity of the cultured seaweeds. Prolonged exposure to low salinity may induce stress that led to reduced photosynthetic efficiency, inhibited cell division, and subsequently result in stunted growth and declined in growth rate. K. alvarezii explants cultivated in the salinity range of 25–35 ppt have been reported to achieve higher DGR of 4.2 ± 0.4 to 4.7 ± 0.5% day−1 as compared to those cultivated in salinities of 20 and 40 ppt with DGR of 2.6 ± 0.3 and 2.2 ± 0.4% day−1, respectively. While Gracilar- ia seaweeds have been found growing well in a wide geographical range with salinities from 15 to 60 ppt, their optimal growth performance was still reported in salinities around 30 ppt. *K. alvarezii, E. denticulatum*, and *G. changii* explants were observed to be unhealthy in the exposure to hyposaline conditions (below 20 ppt) with the formation of ice-ice whitening and bleaching throughout the branches leading to fragmentation and completely damage of the branches [80, 120]. Extremely low salinity may cause oxidative stress in which peroxide may be accumulated in the explants and lead to loss of thallus rigidity as observed in *Gracilaria corticata* under exposure to 15 ppt. Moreover, *K. alvarezii* explants treated in hypersaline conditions (above 40 ppt) have also been reported to exhibit lower growth rate as did those treated in hyposaline conditions, where the growth metabolism may be sacrificed near the salinity tolerance limits to carry out osmoregulation for survival in a short period of time.

15. pH

The Ordinary seawater is slightly alkaline (pH ∼8) with bicarbonate ions ($HCO^{3\ -}$) constituting about 91% of total dissolved inorganic carbon (DIC), followed by 8% of carbonate ions ($CO3^{2-}$), and 1% of dissolved CO_2. Alterations in seawater pH may vary the equilibrium of the carbonate system and change the concentration of inorganic carbon species, subsequently affecting the growth of seaweeds which depend on the supply of inorganic carbon for photosynthesis. The pH range for normal growth of most seaweed cultures was reported to be 7–9 with optimum growth in between 8.2 and 8.7. *K. alvarezii* explants were discovered to attain higher growth rates when cultured in the alkaline conditions (pH 7.5 and 9.5) with respective DGR of 5.5 ± 0.7 and 4.7 ± 0.6% day−1 as compared to the acidic condition (pH 5.5) with DGR of 1.2 ± 0.4% day−1. The increased of hydrogen ions (H+) concentration and decreased of photosynthetic carbon source (HCO3-) under acidified conditions may severely limit the photosynthesis process of explants and reduce their growth rate. Proteins are the primary effector molecules potentially influenced by environmental conditions and associated with the response to various abiotic stresses. Enzymes involved in biological activities are generally respond immediately to the change of pH and achieve their highest performance under the optimum pH range. The low growth rate of *K. alvarezii* explants in acidic conditions may also be due to the denaturation of proteins beyond the tolerance limit, which in turn hinders the cellular physiological and biological processes of seaweeds. However, better growth of purple *K. alvarezii* morphotype in slightly acidic conditions (pH 6.7) was reported indicating different Kappaphycus varieties may respond differently to pH conditions.

16. Optimal growth of seaweed micro-propagules in tissue culture and photobioreactor

In order to maximize the growth of micro-propagules and enhance the productivity of seaweed propagation, the incorporation of optimized parameters in their combination in the tissue culture system (Figure 5a) and the application of a photobioreactor with optimal growth conditions are highly recommended. Maximum DGRs of *K. alvarezii* and *G. changii* have been reported to achieve 5.5 ± 0.7% day−1 and 6.6 ± 1.5% day−1, respectively, under optimized tissue culture conditions. The growth of *K. alvarezii* micro-propagules has further been increased to 6.5 ± 0.2% day−1 in a customized airlift photobioreactor with the incorporation of optimized growth parameters. These growth rate achievements were found significantly higher than the earlier reports of 3–4% day−1 for the growth of *K. alvarezii* in tissue culture and 1–1.5% day−1 for the growth of G. dura in vertical polythene-tube-column culture.

Although macroalgal tissue culture is underdeveloped relative to that of land plants, there are more than 85 species of seaweeds from which tissue culture aspects including successful callus formation, plant regeneration, somatic embryogenesis, and thallus development have been reported. The exploitation of seaweed organogenetic potential for the isolation of superior clones has been initiated since the late twentieth century to improve the performance of cultivated species including *Chondrus, Gigartina, Gracilaria,* and *Kappaphycus*. Studies on optimizing the growth of commercially important seaweeds in tissue culture can help to mass propagate these viable species and open up new opportunities to produce and recover seaweed products from cell and tissue aggregates in a photobioreactor. The development of bioprocess engineering including bioreactor design and identification of strategies for secondary metabolite production can expedite the production of valuable compounds from seaweeds and subsequently derive the maximum benefits from photobioreactor-grown cultures for various industrial applications.

17. Acclimatization of tissue cultured seedlings prior to farming

While the studies of seaweed tissue culture and micro-propagation have been reported from various literature, information about acclimatization and successful out-planting of tissue-cultured seedlings are still limited to date. Acclimatization to ex vitro conditions (nursery or glasshouse) is necessary to provide a buffer condition to the seaweed cultures for suitable adaptation before their exposure to the complex open sea environment. Direct planting out of tissue-cultured seaweeds without going through the acclimatization phase may cause stress and shock to the seedlings due to sudden changes in environmental conditions]. Therefore, an effective acclimatization process is considered to be a key element in enhancing the survival rate of tissue-cultured seaweeds after they have been out-planted to the open sea. Transferring of micro-propagated *K. alvarezii* seedlings from an in vitro flask culture to a partially in vitro tank culture prior to their acclimatization to an outdoor nursery has been recommended to improve their survival capability and growth performance. Investigation of factors or parameters affecting the DGR of micro-propagated K. alvarezii during their acclimatization in outdoor nurseries has also been carried out and reported.

Through the observation, *K. alvarezii* seedlings were found to achieve optimum growth with DGR of 7.14 ± 0.30% day−1 when acclimatized in seawater enriched with mixed-algae fertilizer as formulated in NSE, under daily replenishment of seawater and culture density of 0.40 g L−1. The acclimatization protocol was found to promote faster and healthier growth to *K. alvarezii* seedlings with DGR of 3.91 ± 0.16% day−1 and 83.33 ± 5.77% of survival after they have been out-planted to the open sea.

Furthermore, acclimatization of G. dura in outdoor tank culture with continuous aeration and daily replenishment of 2/3 seawater without supplementation of nutrients has been suggested to improve the growth performance of the species prior to their transplantation to open sea. The growth rate of G. dura seedlings during their acclimatization phase was reported to achieve 2.25 ± 0.14% day−1 as comparable to other outdoor cultures of *Gracilaria* species. Apart from that, the used of perforated polythene bags covered in nylon net bags has been recommended for the out-planting of acclimatized seedlings to avoid loss of biomass, which may result from grazing and drifting. The application of floating rafts accompanied by net convening has also been discovered to ease the seeding, maintenance, and harvesting processes of seaweed farming in the open sea.

18. Quality assessment of tissue-cultured seaweeds

Comparison of the quality between tissue-cultured and conventional cultivated *K. alvarezii* has been reported especially on their growth rate and carrageenan properties. After 60 days of post-cultivation from their first introduction as seedlings in open sea, tissue cultured *K. alvarezii* has been reported to achieve higher growth rate (6.3 ± 0.1% day−1) as compared to conventional cultivated seaweeds (3.4 ± 0.3% day−1). No epiphytes have been discovered on the tissue-cultured K. alvarezii, while the presence of epiphytes and symptoms of “ice-ice” disease were observed on the conventionally cultivated seaweeds. From the analyses of their semi-refined carrageenan properties, tissue-cultured *K. alvarezii* was found to produce higher carrageenan yield with significantly better quality in viscosity, gel strength, and sulfate content in contrast to conventional cultivated seaweeds.

In terms of other chemical composition, tissue-cultured *K. alvarezii* has been revealed to produce significantly higher total lipids and mineral elements including calcium, magnesium, beryllium, cobalt, copper, lithium, manganese, and zinc against the conventional cultivated seaweeds. Research finding has suggested tissue-cultured seaweeds to be a better food source for consumption and other seaweed-related industries. Bioprocess technology for the production of high-value chemicals such as food additives and biomedicinals from cell and tissue cultures of different macroalgae has been proposed and developed using specially designed photobioreactor. Additionally, new approaches in understanding seaweed physiology, biochemistry, and molecular biology has been anticipated to contribute new insights into human nutrition and enable genetic engineering of favorable agronomic traits to improve the quality and the overall productivity of commercially important seaweeds.

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Article ID: 285

GUARDIANS OF THE COAST: THE VITAL ROLE OF MANGROVES IN COASTAL ECOSYSTEMS

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Abstract

Mangroves are unique intertidal ecosystems found in tropical and subtropical areas of the world, that they provide habitat for a wide variety of aquatic and land animals. These ecosystems are extremely productive everywhere in the world while being delicate and rarely distributed (Thatoi and Biswal 2008). Mangroves have extremely developed morphological and physiological adaptations to adverse conditions to deal with such a hostile habitat. They maintain and protect coastal areas while nourishing the coastal water with nutrients. With rapid industrialization and urbanization, heavy metal pollution has become one of the most prominent problems in the ecological environment of mangrove ecosystems. The relatively high concentration of nutrients and metals suggests that water is in very dreadful condition, which will ultimately affect flora and fauna of this ecosystem

Keyword: Mangroves, Ecosystems, Estuary, Biodiversity

Introduction

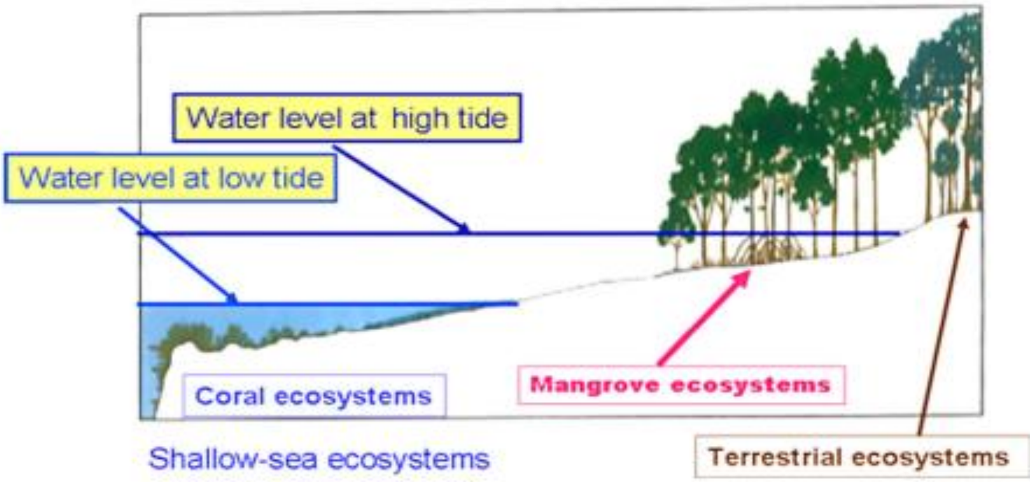
Mangroves are unique intertidal ecosystems found in tropical and subtropical areas of the world, that they provide habitat for a wide variety of aquatic and land animals. Mangroves, which are recognized as highly productive ecosystems of tremendous ecological significance, it refer to approximately 60 to 70% of the world's tropical and subtropical coastlines. The mosaic of mangrove habitats offers a range of biodiversity elements that are crucial to the functioning and environmental quality of tropical estuary ecosystems. Mangroves also play a significant role in maintaining water quality and shoreline stability by regulating the distribution of nutrients and sediment in estuary waters. With rapid industrialization and urbanization, heavy metal pollution has become one of the most prominent problems in the ecological environment of Mangrove ecosystems, which are found along the estuarine shores in tropical and subtropical regions.

Habitat Adaptations

Mangrove plants grow in harsh conditions such as high salinity, hypoxic (oxygen-deficient) wet soil strata, tidal pressures, strong winds, and sea waves. Mangroves have extremely developed morphological and physiological adaptations to adverse conditions to deal with such a hostile habitat. The mangrove root system absorbs oxygen from the atmosphere. Mangroves have special roots for this purpose called breathing roots or **pneumatophores**.

Mangroves, like desert plants, store fresh water in their thick succulent leaves. A waxy covering on the leaves seals in water and reduces evaporation. They are found in the intertidal zone, which is where the terrestrial limit for other plant communities ends and the influence of the marine ecosystem begins.

Occurrence of mangroves in the intertidal zones



There are types of Mangroves:

Red – Found along the coastlines

Black – Major feature of such mangrove trees is their dark bark. They have access to more oxygen.

White – Compared to Red and Black mangroves; they grow at the highest elevation.

Species Diversity

Mangroves are a diversified group of mostly tropical plants and shrubs that are functionally diverse and complicated. They also provide structure and habitat for a variety of marine and intertidal organisms. Mangrove vegetation takes several forms, including trees, shrubs, palms, and ground ferns. Avicenniaceae and Sonneratiaceae are the only plant families that are entirely composed of mangrove taxa, though this is still contested. True mangrove species have been described as 69 species in 27 genera belonging to 20 families (Duke, 1992; Kathiresan and Bingham, 2001; Selvam et al., 2004). Asia is the highest diversity of mangrove species, with 44 species recorded. According to FAO (2003), there are 77 species of mangrove plants worldwide. There are 21 flowering plant genera found in mangroves, indicating a relatively high degree of specialisation for the tidal wetland ecosystem. Twelve angiosperm genera are exclusively mangrove, while ten others comprise non-mangrove species.

Mangrove ecosystems possess four unique characteristics of high productivity, high return rate, high decomposition rate and high resistance to extreme weather events and anthropogenic activities as one of the unique marine ecosystems in the world (Wang, 2019; Liu and Wang, 2020; Liu *et al.,* 2020). Mangrove systems serve as habitat and nursery area for many juvenile fish and crustaceans, which have both direct and indirect socio-economic importance. They also provide erosion mitigation and stabilisation for adjacent coastal landforms (Harty, 1997). Mangroves are also one of the world's richest repertoire for biological and genetic diversity of fauna and flora along roughly 60–75% of the world's tropical coastal zones. There is also an amazing richness of microorganisms and microbial diversity in such ecosystems. Furthermore, 90% of the marine organisms spend part of their life cycles in this ecosystem and 80% about the global fish catches are dependent on mangrove wetlands. The net primary productivity of mangrove ecosystem is up to 2000 g $C\cdot m^{-2}\cdot a^{-1}$, with high strength material cycles, energy flow, as well as maintaining biodiversity (Lin, 1997; Wang, 2019).

Importance of Mangrove Forest

The mangrove forests are of great environmental significance and socioeconomic value:

- Protect coastlines from wind, waves, and sea currents
- Reducing soil erosion and siltation
- Protecting coral reefs, seagrass beds and shipping lanes
- Providing wood and other forest products, renewable fuel source
- Providing habitat and nutrition for a variety of creatures
- Supporting coastal fisheries and livelihoods
- Essential nursery areas for finfish and shellfish
- Mangrove foliage as feed for domestic animals
- Provide opportunities for tourism, education, and scientific research

Threats:

Large scale clearing: To accommodate the human population, agriculture, and aquaculture. This has resulted in forest fragmentation, loss of biodiversity, and a decline of mangrove dominant shorelines.

Small scale harvesting and grazing: For timber, fuel wood, fodder, and other products of persons and their livestock who venture into the forests?

Industrial threats: Effluent pollution, mining, industrial growth, and oil spills are all causes of pollution.

Conservation of the Mangrove ecosystem:

- ✓ Afforestation
- ✓ Legislation (including laws and policies)
- ✓ Monitoring and Surveys (land and aerial, etc.)
- ✓ Protection (including conservation, parks and reserves development, etc.)

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Article ID: 286

INNOVATIONS IN INTELLIGENT PACKAGING: ENHANCING FOOD SAFETY, QUALITY, AND CONVENIENCE

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Abstract:

Intelligent packaging stands at the forefront of food packaging innovation, integrating state-of-the-art technologies to elevate protection, communication, convenience, and containment functions. By incorporating smart devices such as barcodes, RFID tags, indicators, and vital biosensors, this technology ensures real-time monitoring of essential parameters like temperature, pH, and microbial content. It enables precise data collection and robust traceability throughout the supply chain, aligning with Hazard Analysis and Critical Control Points (HACCP) principles to proactively enhance food safety. Beyond this, intelligent packaging is revolutionizing kitchens with its adaptive microwave oven systems, tailoring cooking instructions to specific ovens and user preferences. Advanced data processing and algorithm integration refine these systems, providing personalized and user-friendly cooking solutions. Driven by consumer demands for freshness, safety, and convenience, the future of intelligent packaging promises safer, tailored food experiences, meeting the intricate needs of an evolving global market.

Keywords: Intelligent Packaging, Convenience, Smart Package Devices, Data Carriers, Package Indicators, Biosensors, HACCP, Traceability Systems, Freshness Monitoring, Consumer Demands

Introduction:

Food packaging innovations concentrate on improving the core functions of protection, communication, convenience, and containment. Intelligent packaging, a recent trend, utilizes technologies enabling interaction with the product or its surroundings. This type of packaging offers valuable insights into the product's condition, including factors like temperature, humidity, and freshness. The article delves into the advantages and uses of intelligent packaging, alongside the technologies driving these innovative solutions.

Definition of Intelligent Packaging:

Intelligent packaging, refers to a packaging system with intelligent capabilities, including detection, sensing, recording, tracing, communication, and scientific analysis. Its primary purpose is to enhance decision-making processes by extending product shelf life, ensuring safety, improving quality, providing information, and alerting about potential issues. The key distinction lies in its communicative ability. As the package and food travel together in the supply chain, the package becomes the ideal conduit for communicating the food's condition.

Traditionally, packaging functions as a means to contain and protect products during transportation. However, its potential for information exchange, often overlooked, is crucial. Intelligent packaging not only carries tangible information along with the product but can also transmit data visually (through indicators) or electronically (via barcodes or the Internet) throughout the entire supply chain cycle.

Smart Package Devices: Smart package devices, typically affixed to primary or secondary packaging, enable communication within the supply chain to enhance food quality, safety, and efficiency.

Two Basic Types of Smart Package Devices:

1. Data Carriers (e.g., Barcodes and RFID Tags):

Barcodes: Cost-effective and popular for data storage.

UPC Barcode: Represents 12-digit data, often limited to manufacturer ID and item number.

Reduced Space Symbology (RSS): A family of barcode symbologies for encoding more data in a smaller space.

RSS-14 Stacked Omni-directional Barcode: Encodes the full 14-digit Global Trade Item Number (GTIN) for items with space constraints.

Radio Frequency Identification (RFID) Tags:

Advanced Data Carriers: Used for automatic product identification and traceability.

Reader Emission: Emits radio waves to capture data from RFID tags.

Passive Tags: Powered by reader's energy; reading range up to 15 feet.

Active Tags: Have their own battery; reading range over 100 feet.

Advantages Over Barcodes: No line-of-sight required, larger data storage capacity (up to 1 MB), supports real-time updates, can read multiple tags simultaneously.

2. Package Indicators:

1. Time-Temperature Indicators (External) monitors freshness and safety based on temperature and time. Tracks storage conditions, ensuring proper temperature control. TTIs serve a vital function by tracking temperature variations throughout the distribution and storage phases. They achieve this by displaying visual changes, such as increasing color intensity and dye diffusion, with these alterations being directly influenced by temperature fluctuations. This visual feedback serves as a clear and immediate signal, allowing for timely interventions and ensuring the preservation of food quality and safety.

2. Oxygen Indicators (Internal) ensures modified or controlled atmosphere in packaging, especially for perishable foods. Its proficient in detecting oxidative rancidity, color changes, and microbial spoilage, offering crucial insights into the product's freshness.

3. Carbon Dioxide Indicator (Internal) measures carbon dioxide levels inside the package. Monitors foods stored in reduced oxygen concentration, ensuring quality and safety.

4. Microbial Growth Indicators (Internal/External) detects spoilage microorganisms, indicating microbial quality. Assesses the freshness and safety of perishable foods, especially meat, fish, and poultry.

5. Pathogen Indicators (Internal) identifies specific harmful bacteria like *Escherichia coli* O157. Ensures detection of pathogens in food, enhancing food safety protocols.

6. Mechanical Indicators checks physical integrity of packaging. Detects package leaks, ensuring product containment and preventing contamination.

7. Chemical Indicators indicates specific chemical reactions, verifying storage conditions. Ensures proper storage conditions and detects leaks, maintaining food safety standards.

8. Enzymatic Indicators monitors enzyme reactions related to food quality and freshness. Tracks changes in food components, ensuring quality and detecting spoilage.

9. Redox Dyes reveals changes in food quality through redox reactions. Indicates alterations in food composition and freshness, guiding quality control measures.

10. pH Dyes (Chemical and Enzymatic) monitors pH level variations in packaged products. Detects shifts in acidity or alkalinity, ensuring products meet specified quality standards.

11. Dyes Reacting with Specific Metabolites detects specific components, ensuring freshness and quality of the packaged product. Reacts with volatiles or non-volatiles indicating freshness.

12. Chemical and Immunochemical Methods reacts with toxins or specific molecules, identifying contaminants. Ensures identification of toxins and contaminants, maintaining food safety and quality assurance.

Biosensors: Biosensors play a crucial role in detecting, recording, and transmitting biochemical reactions and information. Comprising a bioreceptor that recognizes specific target analytes and a transducer converting biochemical signals into electrical responses, they possess distinct characteristics such as high specificity, sensitivity, reliability, portability, and simplicity. These attributes make them invaluable for rapid on-site analysis, encompassing tasks like assessing pollutants, detecting pathogens, and monitoring various parameters related to food quality.

Application of Intelligent Packaging: Enhancing Food Safety, Quality, and Convenience

1. **Tracking and Monitoring (Food Safety and Biosecurity)**, it enables real-time tracking using bar codes, RFID tags, and sensors measuring parameters like pH and microbial content. This integration ensures swift data exchange, aiding rapid decision-making for implementing food safety strategies, thereby maintaining product integrity.
2. **Improving Traceability Systems**, intelligent packaging technologies like bar codes, RFID tags, and sensors are integrated, guaranteeing precise data collection across the supply chain. By strengthening communication links, this integration provides comprehensive information, ensuring effective traceability and robust quality assurance.

3. **Implementing HACCP Principles**, it facilitates the application of HACCP principles for food safety. By identifying critical control points and encoding limits within bar codes/RFID tags, coupled with Time-Temperature Indicators (TTIs) for continuous monitoring, this system promotes data sharing among various devices. This collaboration enables rapid detection and correction of safety concerns, ensuring a proactive approach in maintaining food safety standards.
4. **Intelligent Microwave Oven System (Enhancing Food Quality and Convenience)** enhances microwave cooking experiences. Utilizing bar codes for precise oven matching and incorporating real-time temperature and moisture feedback sensors, this technology provides tailored heating instructions for different oven types. It offers accessibility features for visually impaired users, language translation, and online resources like recipes and allergen information. Furthermore, it tracks dietary intake and provides allergen warnings, enhancing user convenience and food safety.
5. **Data Processing and Algorithm Integration**, scientific expertise shapes data layers and processing systems. By integrating data from various sources and employing algorithms rooted in heat transfer principles, the system generates precise instructions for controlling microwave settings. This approach accommodates user preferences and dietary requirements, ensuring a customized and user-friendly cooking experience, ultimately guaranteeing optimal cooking outcomes.

Conclusion:

The future of intelligent packaging is bright, driven by the increasing emphasis on freshness and safety, heightened consumer demands, expanding global markets necessitating enhanced traceability in longer logistic chains, and the capability to facilitate comprehensive in-house control throughout the food supply chain. It ensures real-time monitoring, enhances traceability and HACCP procedures, and elevates overall food quality and convenience by integrating sensors and advanced data processing systems, ultimately ensuring safer and more convenient food experiences.

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Article ID: 287

NANOPARTICLES AS ANTIMICROBIAL AGENTS

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Abstract:

Plant diseases caused by pests and pathogens result in significant annual crop losses, urging the need for innovative and sustainable disease management strategies. Nanotechnology offers a promising solution by harnessing the unique properties of nanoparticles. This chapter explores the multifaceted roles of nanoparticles in plant disease management, serving both as protectants and carriers of active molecules. Nanoparticles, with their ability to penetrate microbial cells, disrupt vital processes, making them efficient protectants. Furthermore, nanoparticles act as carriers for insecticides, fungicides, herbicides, and RNAi agents. Silica, chitosan, solid lipid, and layered double hydroxide nanoparticles serve as carriers, providing controlled release and targeted delivery. Despite their potential, the commercialization of nanoparticle-based products in agriculture faces challenges, including limited field trials and underutilization of host systems.

Keywords: Nanoparticles, antimicrobial properties, protectants, nanocarriers, RNAi

Introduction

Each year, 20% - 40% of crops are lost due to plant pests and pathogens. Existing plant disease management relies predominantly on toxic pesticides that are potentially harmful to humans and the environment. Nanotechnology can offer advantages to pesticides, like reducing toxicity, improving the shelf-life, and increasing the solubility of poorly water-soluble pesticides, all of which could have positive environmental impacts. In this contemporary landscape of agricultural science, the integration of nanotechnology into disease management strategies has emerged as a focal point of research and innovation due to their profound antimicrobial properties, paving the way for ground breaking advancements in agricultural practices.

Nanoparticles, owing to their diminutive size and unique physicochemical attributes, possess exceptional reactivity and surface area-to-volume ratios. These factors endow them with unparalleled capabilities to interact with microbial entities at the nanoscale level. Unlike traditional antimicrobial agents, nanoparticles can penetrate microbial cell walls and membranes, disrupting vital cellular processes and inhibiting pathogenic growth. Such intricate mechanisms of action are deeply rooted in the principles of nanoscience and material physics, driving the scientific community's curiosity to explore their applications in agriculture comprehensively. Nanoparticles can be used in two directions in which nanoparticles can be utilized for plant disease management: either as nanoparticles alone, acting as protectants or as nanocarriers for insecticides, fungicides, herbicides, and RNA-interference molecules.

Definition: Nanoparticles are particles with dimensions ranging from 1 to 100 nanometres. Silver nanoparticles, for example, have gained attention due to their potent antimicrobial properties. These particles, at the nanoscale, have an increased surface area, allowing more contact with microbial cells, enhancing their efficacy.

Rationale for Use: Nanoparticles, such as copper nanoparticles, are used in agriculture due to their ability to release copper ions, which are toxic to bacteria and fungi. In plant disease management, the small size of nanoparticles allows them to penetrate microbial cells more effectively, disrupting their vital processes.

Types of Nanoparticles and Their Antimicrobial Activities

- **Silver Nanoparticles:** In a study conducted on powdery mildew in grapes, silver nanoparticles were found to significantly reduce the disease severity. These nanoparticles adhered to the fungal cells, disrupting their structure and inhibiting their growth.
- **Copper Nanoparticles:** A study on tomato plants infected with bacterial wilt demonstrated the effectiveness of copper nanoparticles. These nanoparticles penetrated the bacterial cells, releasing copper ions. This disrupted the bacterial metabolism, inhibiting the progression of the disease.
- **Zinc Nanoparticles:** Research on wheat plants infected with rust diseases highlighted the inhibitory effect of zinc nanoparticles. By disrupting the spore germination process of the rust fungi, zinc nanoparticles reduced the infection rate and minimized crop damage.

Mechanisms of Antimicrobial Action

- **Physical Mechanisms:** Silver nanoparticles, for instance, have been observed to physically attach to the cell membrane of pathogens. Once attached, they create pits and cause membrane damage, leading to cell death. This mechanism prevents the pathogens from reproducing and causing further infection.
- **Chemical Mechanisms:** Copper nanoparticles release copper ions, interfering with the enzymes and proteins within the microbial cells. This disruption of cellular processes weakens the pathogens, making them less harmful to plants. The selectivity of nanoparticles ensures minimal impact on beneficial microbes.

Nanoparticles in Plant Disease Management: Protectants and Carriers

- **Nanoparticles as Protectants**

Nanoparticles, particularly silver, copper, zinc oxide, and titanium dioxide, show promise in shielding plants against pests and pathogens. Silver nanoparticles, produced through eco-friendly methods involving plants, bacteria, fungi, or yeast, exhibit potent antifungal properties against various harmful fungi. They have demonstrated effectiveness against pathogens like *Alternaria alternata*, *Sclerotinia sclerotiorum* and *Botrytis cinerea*. However, challenges like production complexity and potential toxicity hinder their widespread use.

Other metals like copper, titanium dioxide, and gold are also explored for plant disease management. Chitosan nanoparticles, due to their biodegradability and antimicrobial properties, provide viral resistance and combat diseases like *Fusarium* crown rot in tomatoes and *Botrytis* bunch rot in grapes. Their mechanisms include cell membrane disruption and inhibition of toxin production.

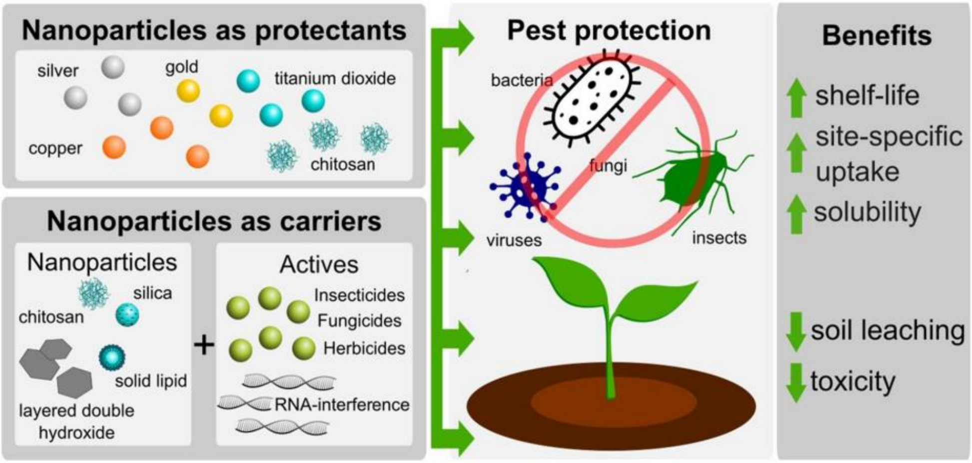

Figure 1. Nanomaterials as protectants or carriers to provide crop protection.

- **Nanoparticles as Carriers:**

Nanoparticles serve as carriers for active molecules like insecticides, fungicides, herbicides, and RNAi agents. Silica, chitosan, SLN, and LDH nanoparticles are common carriers explored for plant disease management, each offering unique advantages in agricultural applications.

Silica nanoparticles, with their customizable structure, act as efficient delivery vehicles. Porous hollow silica nanoparticles (PHSNs) and mesoporous silica nanoparticles (MSNs) protect active molecules from degradation and offer sustained release. Chitosan nanoparticles, often modified for better solubility, adhere well to plant surfaces, enhancing bioactive molecule uptake. Solid lipid nanoparticles (SLNs) entrap lipophilic molecules without organic solvents, ensuring controlled release.

Layered double hydroxides (LDHs) trap active substances between their layers, aiding in safe transport across plant cell walls.

Applications in Plant Disease Management

- **Seed Treatment:** In a study with soybean seeds, zinc oxide nanoparticles were applied as seed coatings. These nanoparticles protected the seeds from soil-borne fungi, enhancing germination rates and ensuring healthier seedlings.
- **Foliar Sprays:** Copper nanoparticles, when formulated into a foliar spray, have been successfully used to manage downy mildew in vineyards. The nanoparticles adhered to the leaf surface, creating a protective layer that prevented the entry of the mildew-causing pathogen.

- **Soil Application:** Silver nanoparticles, when incorporated into the soil, reduced the population of root-knot nematodes in a tomato field. The nanoparticles interfered with the nematodes' reproduction, decreasing their numbers and subsequently reducing plant damage.

Future Prospects and Research Directions

- **Nanotechnology in Precision Agriculture:** Researchers are exploring the integration of Nano sensors with nanoparticles. These sensors can detect disease signals in real-time and trigger the release of nanoparticles precisely when and where they are needed, enhancing the efficiency of disease management.
- **Nanoparticles for New Disease Targets:** Ongoing studies are focusing on emerging threats, such as sudden oak death in trees. By tailoring nanoparticles to combat specific pathogens, scientists are developing targeted solutions to protect vulnerable plant species from devastating diseases.
- **Multidisciplinary Research:** Collaborative efforts between plant pathologists, nanotechnologists, and agronomists are essential. By combining expertise, researchers can develop comprehensive solutions that not only combat diseases effectively but also ensure the sustainability of agricultural practices.

Conclusion

Despite the several potential advantages associated with the use of nanoparticles, not many nanoparticle-based products have been commercialized for agricultural application. The scarcity of commercial applications could be explained by several factors, such as an insufficient number of field trials and underutilization of pest-crop host systems. In other industries, nanotechnology has progressed rapidly, and the only way to keep up with this advancement for agricultural applications is by understanding the fundamental questions of the research and addressing the scientific gaps to provide a rational and facilitate the development of commercial nanoproducts.

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Article ID: 288

NEUTRACEUTICALS USED AS FEED ADDITIVES IN AQUACULTURE

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Abstract

Nutraceuticals (often called phytochemicals or functional foods) are natural bioactive, chemical compounds with health-promoting, disease-prevention, or medicinal properties. Herbal extracts are natural plant product that acts as anti-stressors, immunostimulants, growth promoters, and antimicrobial agent and stimulates appetite due to the active components such as alkaloids, flavonoids, pigments, phenolics, terpenoids, steroids, and essential oils present in them. In the present article, an overview of what is neutraceuticals and how it is the beneficial role in fish health has been discussed.

Introduction

The Global Population will be 8 billion in 2022; it is expected to reach 9.7 billion in 2050. So the Demand for food and quality food will be increased, and the Global apparent consumption of aquatic food rate is almost twice (3%) than annual world population growth (1.6%) (FAO, 2022). So we need to focus on increasing fish production to increase the supply of quality feed for the future. In India, the total output is 16.25 MMT in which, 12.12 MMT is Inland, and 4.13 MMT is Marine production(Ministry of Fisheries, Animal Husbandry and Dairying Annual report 2021-22). Also, due to escalation, an aquaculture system is a significant pressure on the aquafeed industry to prepare cost-effective feed. For economic preparation of feed, we can use the economically available neutraceuticals such as natural herb products used as feed additives instead of the costly feed additive, which will cause side effects to the fish.

The term "nutraceutical"-was coined from "nutrition" and "pharmaceutical" in 1979 by Stephen DeFelice, MD- founder and chairman of the Foundation for Innovation in Medicine. He states, "a nutraceutical is any substance that is a food or a part of food and provides medical or health benefits, including the prevention and treatment of disease" (Biesalski, 2001).

Such products may range from isolated nutrients, dietary supplements, and specific diets to genetically engineered designer foods and herbal products(De Felice Stephen, 1995; Rishi, 2006). Doubtlessly, many of these products possess pertinent physiological functions and valuable biological activities (Andlauer and Furst, 2002)

Needs for neutraceuticals as Feed Additives

Due to the escalation of aquaculture production, aquafeed demand also increased. If demand increases, the cost of aquafeed increase and become low availability of fish meal. Need to search for cheaper protein sources such as unconventional plant-based feed ingredients, which show lower nutrient digestibility which is attributed to a high level of carbohydrate, especially non-starch polysaccharides and various anti-nutritional factors like saponin, protease inhibitors, tannin, etc.,. The fish Early stage had high metabolic activity, due to this reason nutrient dense diet needed but digestive system not well developed. So improper digestion and malabsorption of nutrients causes reduced growth, impaired immunity, allergic reaction, poor wound healing etc., due to this problem nutraceuticals added in feed and tackled all health issue as a feed additive.

Functional foods Versus nutraceuticals

"Functional food" is described as food prepared or cooked with "scientific intelligence," whether or not the cook is aware of the method's usage or purpose. Thus, available food provides the body with the required amount of vitamins, fats, proteins, carbohydrates, etc., for healthy survival.

Nutraceuticals are a type of functional foods that assist in preventing and treating diseases and disorders. Fortified dairy products like milk and citrus fruits like orange juice are examples of nutraceuticals (Kalra EK 2003).

Classification

- Acidifiers: Organic acids such as citric acid and benzoic acid
- Enzymes: Exogenous enzymes such as amylase, phytase, etc.
- Peptides: Hormones etc
- Prebiotics: Metal ions
- Probiotics: Fish gut bacteria
- Plant extracts
- Essential oils: Fish oils and linseed oils
- Antioxidants: 1. Tocopherols (Vit. C, E) and Tocotrienols 2. Carotenoids (carrot, red pepper) 3. Flavonoids (onion, grapeseeds, citrus) 4. Polyphenols (Green tea, Tannins)
- Antimicrobials-1. Sulfur compounds: Onion, Garlic family, Sulphides, and Thioles 2.Terpene compounds: Thyme, Oregano, Turmeric, Ginger. 3. Phenols: Cloves, Nutmegs, Cinnamons, Eugenol, Cinnamic acid, Thymol. 4. Glycosides: Sugar,

non-sugar, Glycon, Aldehydes like Citral, Citronellol 5. Ester: Eugenyl acetate and Linalyl acetate. 6. Alcohols: Limalool and Terpinol.

- ❖ Antivirals
- ❖ Anti-inflammatories
- ❖ Anti-allergenic
- ❖ Anti-helminths
- ❖ Immuno- supportive- 1. Fungal ß –Glucans 2. Microbial Levan 3. Chitin and chitosan 4. Bovine lactoferrin 5.Nucleotide 6.Fucoidan 7. Propolis extract 8. Carotenoids
- ❖ Mucosal-supportive
- ❖ Capillary protective
- ❖ Polysaccharides- 1. Sodium alginate 2. Chitin 3.Chitosan 4.Carageenan 5.Beta-glucan 6.Levan 7.Fucoidan

Benefits

- Bridging the gap between food and medicine
- Increase the health value of the diet to feed the fishes
- May help fish live longer
- May help to avoid particular medical conditions
- May have a psychological benefit from doing something for oneself
- Perceived to be more "natural" than traditional medicine and less likely to produce unpleasant side-effects
- May present food for populations with special needs (e.g., nutrient-dense foods for the elderly) (Consumer Association of Canada 2009)

Problems tackled by Neutraceuticals:

- Fish lives in an aquatic environment where they are prone to various stressors
- Recently, fish and shellfish farmers faced mortality due to bacterial and viral diseases, so there is a need to stimulate the immune system
- Vaccination impractical due to the unavailability of well-practiced personnel
- Nutraceuticals are an alternate option through diet
- Shift from maximum growth to health of the fish
- Antibiotic ill effects are bioaccumulation and resistant strain generation
- Reducing the investment cost through natural herb inclusion in feed

Conclusion

Nutraceutical conglomerates can be used in aqua feed in proper combinations to boost immunity and remove the anti-antinutritional factor of plant-based ingredients. It provides a new thrust area for future research, production of low-cost feed for sustainable global aquaculture.

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Article ID: 289

OPTIMIZING FORTIFIED FOODS: A COMPREHENSIVE EXPLORATION OF MINERALS, VITAMINS, AND FOOD PROCESSING

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Abstract:

Food fortification, a vital strategy in global nutrition, involves enriching food with essential minerals and vitamins to combat nutritional deficiencies. This article meticulously explores the selection criteria for these nutrients, focusing on their stability, solubility, and bioavailability. A detailed examination of various minerals (iron, zinc, calcium, and selenium) and vitamins (A, E, D, K, and B-vitamins) is presented, outlining their specific applications in fortification. Additionally, the study delves into the intricate world of food processing, evaluating methods such as baking, extrusion, microwave and pressure cooking, and soaking. These techniques significantly impact mineral bioavailability in fortified foods, with nuanced outcomes observed in selenium retention and iron and zinc loss. The findings underscore the complexity of maintaining nutritional integrity during processing, emphasizing the need for meticulous cooking practices and innovative technologies. Awareness of these alterations empowers consumers, chefs, and nutritionists to contribute significantly to public health. Continuous research, awareness, and innovative approaches are imperative to optimize fortification programs, ensuring fortified foods retain their intended nutritional benefits, thus positively impacting communities worldwide.

Introduction:

The fortification of food with essential minerals and vitamins is a pivotal strategy in combating global nutritional deficiencies. The meticulous selection of minerals and vitamins for fortification involves a careful consideration of their stability, solubility, and bioavailability. This article delves into the intricate criteria that guide the selection of various minerals and vitamins commonly utilized in food fortification. Understanding the properties of these nutrients is crucial in developing effective fortification programs that can make a substantial difference in public health on a global scale. Additionally, this study explores the intricate realm of food processing, where techniques such as baking, extrusion, microwave cooking, pressure cooking, and soaking significantly impact the bioavailability of essential minerals in fortified foods. By dissecting these effects, we aim to shed light on how these methods influence the nutritional quality of fortified foods, providing essential insights for both the scientific community and consumers alike.

Minerals for Fortification:

1. Iron:

Ferric pyrophosphate: Minimal color change, resistant to rancid fat, suitable for preserving Vitamin A.

Ferrous sulfate: Limited use in coating/dusting, recommended by WHO for wheat fortification.

Ferric ortho phosphate: Lower bioavailability, utilized in the US.

Iron ethylene diamine tetraacetic acid sodium salt (NaFeEDTA): High Fe bioavailability, used in cereals like wheat and maize.

2. Zinc:

Zinc oxide: Highly water-soluble, cost-effective, suitable for wheat and maize fortification.

Zinc sulfate: More expensive, maintains Vitamin A stability in rice.

3. Calcium:

Calcium carbonate: Whitening effect in rice, used in hot extrusion.

Calcium lactate gluconate: Recommended for highly soluble cases.

4. Selenium:

Sodium selenite: Employed in rice fortification, notable in countries like Costa Rica.

Vitamins for Fortification:

1. Vitamin A:

Beta carotene: Provitamin A, colorant, stable source of Provitamin A and an antioxidant.

Vitamin A Palmitate: Stabilized with BHT/BHA, widely used in grain fortification.

Vitamin A acetate: Less effective, used in spray-dried forms in rice.

2. B-Vitamins:

Thiamine mononitrate: Common in rice and maize fortification.

Riboflavin: Water-soluble, used in rice, wheat, and maize.

Nicotinic acid: Used in wheat fortification.

Pyridoxine hydrochloride: Water-soluble, used in various foods.

Folic acid: Stable, used in rice, maize, and wheat fortification.

Cyanocobalamin: Used in rice, wheat, and maize fortification, often combined with folic acid.

3. Vitamin C:

Ascorbic acid: Suitable for rice fortification, often combined with Beta carotene.

4. Vitamin E:

Vitamin E acetate: Very stable, utilized in dry preparations or pure oily forms.

5. Vitamin D:

D3 Cholecalciferol: Not commonly used in rice, prevalent in dairy and flour fortification.

6. Vitamin K:

Not typically used in rice and other cereals.

7. Amino Acid:

Lysine hydrochloride: Highly water-soluble, suitable for rice fortification, especially in extrusion applications.

Effect of Various Food Processes on Mineral Bioavailability in Fortified Foods:

Baking:

1. Fortified Whole Wheat Flour: Reduction in Mn, Fe, Cu, and Zn contents observed after baking.

2. Bread from Se-Enriched Whole Meal/White Flour: No significant reduction in Seleno-methionine content after processing, indicating retention of selenium during bread production.

Extrusion:

Extruded Foods: Phytase enzyme inactivation/deactivation occurs, affecting mineral availability.

Microwave Cooking:

Common Cereals: Significant decrease in the bioaccessibility of Se, SeMet, and seCys2 observed, impacting selenium absorption.

Pressure Cooking:

Common Cereals: Decrease in Se content, with a better effect on retaining Se forms compared to microwave cooking, suggesting pressure cooking preserves certain selenium forms.

Soaking:

Brown Rice Cultivar: High loss of Fe (5%) and Zn (>64%) noted during soaking, indicating a significant reduction in iron and zinc content.

Advantages of Fortification:

1. Improved Nutritional Intake: Fortified foods provide essential nutrients that might be lacking in people's diets. This is particularly important in regions where certain micronutrient deficiencies are prevalent, such as vitamin A, iron, iodine, and folic acid.

2. Prevention of Micronutrient Deficiencies: Fortification helps prevent and alleviate micronutrient deficiencies, which can lead to various health problems, especially in vulnerable populations like pregnant women, infants, and children. For example, iodized salt prevents iodine deficiency disorders, including goitre and intellectual disabilities.

3. Enhanced Public Health: By addressing specific nutrient deficiencies, food fortification contributes to improved overall health in communities. It can reduce the prevalence of diseases associated with nutrient deficiencies, leading to a healthier population.

4. Cost-Effectiveness: Food fortification is often a cost-effective way to deliver essential nutrients to a large population. Compared to individual supplementation or clinical interventions, fortifying the commonly consumed foods is generally more affordable and sustainable in the long term.

5. Widespread Reach: Fortified foods, especially staple foods like rice, wheat flour, salt, and cooking oils, are widely consumed across various socioeconomic groups. This widespread consumption ensures that the fortified nutrients reach a broad segment of the population, including those with limited access to healthcare.

6. Reduction in Birth Defects: Fortifying certain foods, such as fortification of flour with folic acid, has been shown to reduce the incidence of neural tube defects (serious birth defects of

the brain and spine) in newborns. Adequate folic acid intake before and during pregnancy is crucial for preventing these defects.

7. Supports Cognitive Development: Fortification with nutrients like iron and certain vitamins supports proper cognitive development in children. Iron, for instance, is essential for brain development, and iron-fortified foods help prevent iron-deficiency anaemia in children.

8. Combats Hidden Hunger: Hidden hunger refers to micronutrient deficiencies that occur even when caloric intake is sufficient. Fortified foods address hidden hunger by providing necessary micronutrients without significantly increasing caloric intake.

9. Addressing Specific Health Concerns: Fortification can be tailored to address specific health concerns within populations. For instance, calcium-fortified foods are beneficial for bone health, especially in aging populations at risk of osteoporosis.

10. Complements Diverse Diets: In regions where diets lack diversity or where certain food groups are not readily available, fortification ensures that essential nutrients are still accessible, regardless of the local food supply.

Disadvantages of Fortification:

1. Overconsumption of Nutrients: Fortified foods can lead to excessive intake of certain nutrients if people consume multiple fortified products. This overconsumption can be harmful, especially for fat-soluble vitamins like Vitamin A and D, and minerals like iron.

2. Imbalance in Nutrient Intake: Fortification may create an imbalance in nutrient intake if not carefully regulated. For instance, excessive fortification of certain nutrients may lead to a disproportionate intake of nutrients, causing health issues.

3. Interaction with Medications: Fortified foods might interact with medications. For example, calcium-fortified foods may interfere with the absorption of certain antibiotics or thyroid medications.

4. Allergic Reactions: Some individuals may be allergic to certain fortification ingredients. For instance, fortifying foods with nuts or soy can pose a risk to people with allergies to these ingredients.

5. Masking Poor Diet Quality: Fortified foods might give a false sense of security to individuals who rely on them, leading them to neglect a balanced and varied diet. Fortified foods should not replace whole foods, fruits, and vegetables in a healthy diet.

6. Impact on Natural Food Sources: Fortifying specific foods might reduce the demand for natural food sources of essential nutrients. This could affect farmers and communities relying on these natural sources for their livelihoods and nutrition.

7. Cost: The process of fortification can increase the cost of production, which might be passed on to consumers, making fortified foods more expensive for low-income populations.

8.Regulatory Challenges: Ensuring the right levels of fortification and avoiding excessive nutrient intake requires strict regulations and monitoring. Inadequate regulations or lack of enforcement can lead to issues related to the quality and safety of fortified foods.

9. Cultural and Ethical Concerns: Fortifying certain foods might not align with cultural or ethical dietary practices. For instance, fortifying foods with animal-derived nutrients might not be suitable for vegetarian or vegan populations.

10. Environmental Impact: The production and processing of fortified foods, including the production of synthetic nutrients, can have environmental impacts, such as pollution and energy consumption.

Conclusion:

In conclusion, the multifaceted impact of food processing techniques on mineral bioavailability in fortified foods highlights the complexity of ensuring their nutritional integrity. While baking, extrusion, and cooking methods can lead to substantial reductions in vital minerals, there are nuanced ways in which certain nutrients are retained. The stability observed in selenium retention during bread production and the preservation of specific selenium forms in pressure cooking underscore the intricate nature of these processes. This exploration emphasizes the need for meticulous attention to cooking practices, dietary choices, and innovative food processing technologies. By being aware of these alterations, consumers, chefs, and nutritionists can contribute significantly to public health. The optimization of mineral and vitamin fortification programs demands continuous research, awareness, and innovative approaches, ensuring that fortified foods maintain their intended nutritional benefits, thus positively impacting the well-being of communities worldwide.

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Article ID: 290

THE ROLE OF BIOMANIPULATION IN FISHERIES OF LACUSTRINE ECOSYSTEMS

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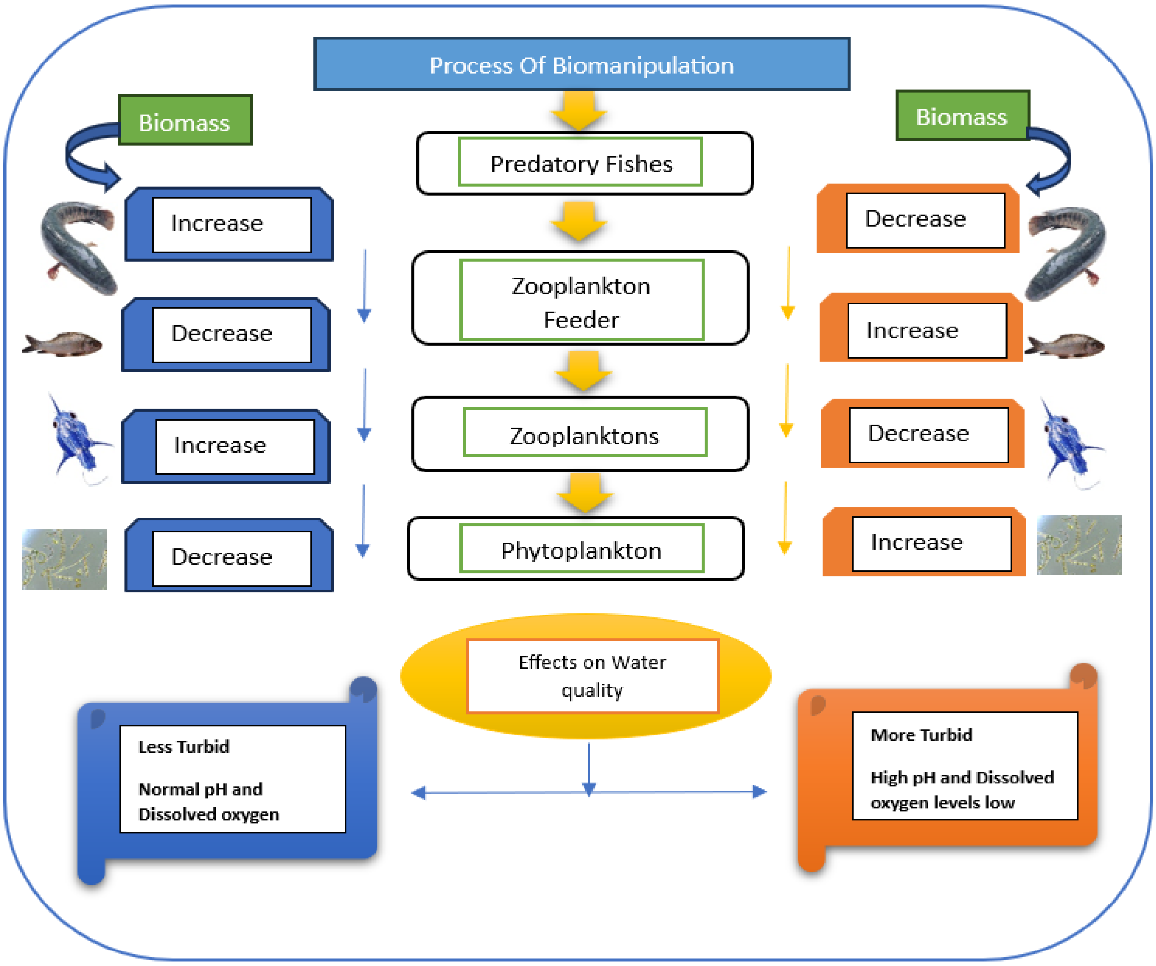
Abstract

Eutrophication resulting from high nutrient loading has been the paramount environmental problem for lakes world-wide for the past four decades. Efforts are being made in many parts of the world to reduce external nutrient loading via improved wastewater treatment or diversion of nutrient-rich inflows. In that case, biomanipulation technique which is cost-effective technique can be considered as one of the most suitable solution to overcome the problem of eutrophication and to restore the quality of water in small and shallow eutrophied lakes. Biomanipulation was originally based on the idea that when the number of planktivorous fish are reduced, the density of large cladoceran zooplankton increases, and their grazing during the summer period can reduce certain species of planktonic algae and reduce the algal turbidity of the water. There is a need of more scientific research with respect to the potential bio-manipulation of lake ecosystems with the long-term positive effects.

Keyword: Eutrophication, Lake Ecosystem, Biomanipulation

Introduction

The problem of eutrophication nowadays is greatly affecting the water quality and aesthetic values of the lake ecosystems. In that case, bio-manipulation technique which is cost-effective technique can be considered as one of the most suitable solution to overcome the problem of chances for success of bio-manipulation technique depends on the approach adopted i.e. whether the removal of fish was partial or complete, if only predatory fish were stocked, size (small lakes are easy to manipulate, Meijer 2000) and depth of the lake but the complete success through bio-manipulation is still debatable. Bio manipulation is generally described as engineering technology." Biomanipulation has become a routine technique for improving water quality of lakes and reservoirs (Hansson *et al.,* 1998; Drenner & Hambright, 1999). Bio manipulation biology is further explained as a management practice adopted by humans to improve degraded water bodies. Wysujack & Mehner (2002) suggested Best bio-manipulation strategy for the fisheries management is the combination of piscivore introduction, manual removal of fish species and its integration with the nutrient management. There is a need of more scientific research with respect to the potential bio-manipulation of lake ecosystems with the long-term positive effects.



Major components behind Bio-manipulation mechanisms

Fish:

Fish is an important component of the bio manipulation process. There are two types of approaches we can follow in order to control the population of planktivorous fishes and to increase the zooplankton biomass. First approach to reduce the biomass of planktivorous fish is through kill (using some chemicals like rotenone by following the recommended dose) and removal (capturing the fish directly by netting) whereas secondly, we can go for the stocking of predatory (piscivorous) fishes. A reduction in the biomass of benthivorous fish is also suggested to have a favourable impact on the bio-manipulation process because the benthivorous fish while feeding will stir-up the bottom causing more turbidity, impairing the colonization & growth of macrophytes and their stability.

Whereas the role of Phyto-planktivorous fish in bio-manipulation process is important as they improve the water quality by actively grazing upon the phytoplankton blooms. Radke and Kahl (2002) carried out an experiment of fish bio-manipulation and concluded that silver carp (Hypophthalmichthys molitrix), a Phyto-planktivorous fish species can more strongly impact the planktonic Cladocera's than phytoplankton, therefore, this fish cannot be considered as a candidate fish species for bio-manipulation in mesotrophic lakes. Starling *et al*. (2002) carried out a study and suggested that introduction of some small-scale commercial fish species targeted against these fishes would not only improve the water quality of the reservoir but also provide the local population with good quality protein. As an alternative to the fish , Roy *et al.* (2010) and Gulati *et al.* (2008), suggested that the introduction of mussels can also create clear water through the filtration of water as they are the filter-feeders and by reducing the nutrient load. In this case, Zebra mussel (Dreissena polymorpha) could be considered as a suitable candidate for bio-manipulation technique as it can reduce the phytoplankton biomass through filter feeding (Caraco *et al.* 1997 and Reeders *et al*. 1993)

Zooplankton:

Being a key component of lake ecosystems, zooplanktons have a very important role in the process of bio-manipulation. Cooke (1986) stated that larger zooplanktons can consume a variety of algal blooms more efficiently as compared to smaller ones. Daphnia is recognized as the most significant genus to impact upon algae blooms and major contributor to the success of bio-manipulation. When large Daphnia are absent, zooplankton cannot reduce phytoplankton biomass. Mehner *et al.* (2002) suggested that intensive grazing by Daphnia on phytoplankton leads to the better water quality and also macrophytes becomes the dominant primary producers which would ultimately suppress the phytoplankton biomass. So, the main goal of the bio-manipulation technique is to lower the mortality of Daphnia to achieve the desired results.

Macrophytes:

Aquatic macrophytes have been identified as a key component for the long-term success of biomanipulation management. Macrophytes stabilize the sediment preventing re-suspension of nutrients as well as utilizing nutrients for their own growth. The main role of the macrophytic communities of the lake is that they provide refuge to the zooplanktons and create the zone of low-oxygen levels where planktivorous fishes cannot survive well which would result in the prohibited entry of planktivorous fishes to zooplankton refuge (Shapiro 1990). Increased macrophytic population will reduce the algal blooms ((Hosper 1990). Fugl and Myssen (2007) attempted the restoration of lake L. Rogbolle in Denmark with the natural establishment of submerged flora whereas Moore *et al.* (2010) attempted the restoration of Freshwater tidal area in USA by following the artificial or man-made introduction of shoots, seeds and seed pods of Vallisneria americana and protection against grazing.

Purpose of bio manipulation

The key purpose of this technique is to decrease the high concentration of toxic phytoplankton in the water bodies that cause eutrophication. The growth of phytoplankton is controlled by introducing zooplanktons that eventually improve water quality and nutrient cycling.

Importance of bio manipulation

Bio manipulation has significant importance for fisheries. This method is being used by people for decades. Eutrophication has an impact on the diversity of the fish population, this could cause fishermen to lose their source of livelihood. Predator species have been purposefully introduced into the aquatic ecosystem by humans in an effort to control and enhance water quality through a biological process. It is important for the transparency of water in lakes. The habitat is restored for the fish population and aquatic biodiversity.

Advantages of Biomanipulation

1. Natural Process Introduced by Humans, It is a kind of natural/ biological process induced by humans to improve the water quality of the aquatic ecosystem.

2. Reduces Turbidity

3. No Requirement of Chemicals

4. Improves Fisheries

5. Maintain Nutrient Cycling

6. Supports Biodiversity, the decreasing biodiversity due to algal blooms starts increasing as water quality gets better.

Disadvantages of bio manipulation

- ✓ **Management of Lakes:** When using the bio manipulation technology, lakes need to be managed continuously.
- ✓ **Poisoning of Water Bodies:** The water bodies are poisoned by Rotenone application and water is not suitable for human consumption.
- ✓ **Lack of Awareness:** The fishermen are often not aware of the side effects of dominant species. Awareness about the importance of predator species is important for Fishermen.
- ✓ **Expensive Treatment:** The cost of the bio manipulation method is high and it is totally dependent on the method which is being used.

Challenges for bio-manipulation

- ✓ Bio-manipulation can only be used for small, shallow and closed system which means lake system needs to be totally closed (no connection with other water bodies)
- ✓ It is must to remove some fish fauna prior to the introductions of new piscivorous fishes to the lakes to reduce the risk of competition for food, shelter and breeding ground
- ✓ The eutrophication of lakes is being caused by increased worldwide development activities and climate change, which are doing so through increased nutrient influx.
- ✓ Modifications to the lake ecosystems' structure
- ✓ Limited long-term effectiveness

Conclusion

In summary, biomanipulation plays a crucial role in fisheries management in lacustrine ecosystems by influencing fish populations and trophic interactions to restore and maintain ecological balance. It's an approach that requires careful planning, monitoring, and adaptive management to ensure its effectiveness and minimize potential negative impacts on the ecosystem.

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Article ID: 291

ZEBRAFISH AS A MODEL ORGANISM FOR TOXICITY STUDIES

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ABSTRACT

One of the key challenges in toxicology studies is the need for reliable and efficient model organisms that can help researchers assess the effects of chemicals, drugs, or pollutants precisely. Zebrafish can provide a solution to this challenge as they have genetic similarities with humans, short generation time, as well as high fecundity. Zebrafish Embryo toxicity tests (ZFET) are widely employed because of transparent embryos that help in observing each stage of embryonic development and OECD has recommended the use of zebrafish fish embryos as an alternative for acute toxicity tests. In recent years, zebrafish adults and embryos have been widely employed as a versatile vertebrate model for toxicity evaluation, and several endpoints such as hatching Success, organ abnormalities, skin damage, aberrant behavior, respiratory behavior, immunotoxicity, neurotoxicity, endocrine disruption, and reproductive toxicity were investigated (Chakraborty *et al.*, 2016). Zebrafish is also used in various fields such as drug development, cancer studies, toxicity studies, molecular and genetic studies for its advantages such as accessibility, operability, and economy.

Introduction

Using fish as model organisms for toxicology is becoming more popular because they absorb as well as metabolize pollutants directly from contaminated water or indirectly through the food chain and exhibit toxicological responses to chemicals even at low concentrations (Hariri *et al.,* 2017) and also respond to toxicants like higher vertebrates (Mir *et al.,* 2014). Zebrafish possess a remarkable feature during their early development such as transparent embryos which gives enormous advantages for research purposes such as direct monitoring of developmental stages, aids in identifying phenotypic traits during mutagenesis, allows for endpoint assessment in toxicity testing, and permits direct observation of gene expression using light microscopy. In addition to their optical clarity, other advantages of zebrafish are short life cycles, high fecundity, and easy maintenance. Zebrafish embryos are recommended as an ideal alternative to traditional animal models since they reduce required workspace, laboratory solution costs, expenses associated with test chemicals, and manpower required. Throughout their life stages, starting from embryos to adults, zebrafish are employed in a diverse array of research domains, with a primary focus on exploring various facets of toxicology concerning chemicals and pharmaceuticals.

Moreover, the accessibility of the zebrafish's complete genome sequence adds to its attractiveness for molecular and genetic research pursuits.

Biology of zebrafish

Kingdom:Animalia
Phylum:Chordata
Class:Actinopterygii
Order: Cypriniformes
Family: Cyprinidae
Genus: Danio
Species:D. rerio

The zebrafish, scientifically known as *Danio rerio* (Hamilton-Buchanan, 1842), is a small tropical cyprinid fish that predominantly inhabits rivers connected to the Ganges River system, including regions such as Burma, the Malakka Peninsula, and Sumatra (Talwar & Jhingran, 1991). The ideal temperature for zebrafish is about 28-29°C. This species of zebrafish typically attains a size ranging from 3 to 5 cm and carries a weight of 150 to 300 mg. With a relatively short lifespan of approximately three months, the zebrafish is renowned for being one of the most readily available and accessible fish species for research purposes.

Zebrafish embryos

Zebrafish are highly prolific breeders and have a fecundity of 200- 300 eggs on a weekly basis. Zebrafish embryos are naturally transparent, allowing researchers to easily observe and record their development. The embryos have all major organs developed within 5 to 6 days (Basu and Sachidanandan, 2013). Zebrafish undergo faster development, originating from a single-cell zygote. Gastrulation initiates around 6 hours after fertilization, leading to hatching as free-swimming larvae within 2 days. Embryos will be dependent on the yolk sac up to 96 hours after fertilization and then the independent feeding starts. These zebrafish typically attain sexual maturity at approximately 3 months of age and exhibit a lifespan of up to 5 years.

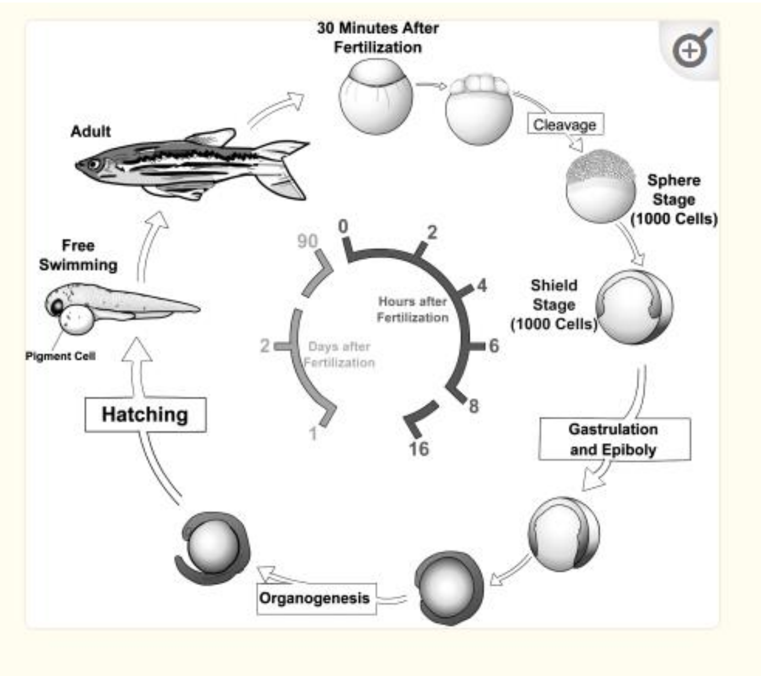

Fig 1. Life cycle of zebrafish. Source: 10.1089/zeb.2008.0562

Zebrafish Embryo Toxicity test (ZFET)

Biomarkers serve as valuable tools for assessing the impact resulting from exposure to xenobiotics or the consequences of anthropogenic activities (Celander, 2011). An effective, non-invasive biomarker that proves particularly useful for screening chemicals during the early developmental stages of zebrafish is the Zebrafish Embryo Toxicity Test (ZFET). This approach provides valuable insights into potential toxicity during later stages of a zebrafish's life, (Braunbeck *et al.,* 2015). Additionally, the developmental process of zebrafish is highly susceptible to external substances, further underscoring the significance of this methodology. It can be used as a powerful tool for assessing developmental abnormalities and signaling potential teratogenic effects (OECD, 2013, OECD, 2019).The zebrafish embryo toxicity test (ZFET) is a 96-hour cell culture-based assay that exposes zebrafish embryos to test solutions during their early developmental stages. It is a highly effective method for identifying both acute and sub-lethal toxicity. ZFET must be set within 4 hours post fertilization embryos and only fertilized should be selected for the exposure.

Fig 2. Use of zebrafish as a model in various toxicity studies

Zebrafish as a model for teratogenicity

Any external agent that affects cellular differentiation, cellular proliferation, or apoptosis might cause the embryo toxic or teratogenic consequences (Gilbert, 2010). Simple teratogenicity tests that are generally reliable in determining teratogenic potential are gaining popularity. Zebrafish embryos are widely used as recognized models to screen various chemicals for teratogenic effects using a variety of endpoints. Lethal endpoints include egg coagulation, non-tail detachment, lack of heartbeat, and lack of somite formation. Sub-lethal and teratogenic endpoints are malformations of the heart, facial structure, eyes, brain, somite, otoliths, spinal cord, heart, yolk, tail, fin, mouth, and pharyngeal arch, or stomach. It is valuable in determining the sub-lethal endpoints, since it provides information on teratogenic effects and long-term impacts on offspring, in addition to acute exposures and mortalities (Arellano-Aguilar et al., 2015).

Advantages of using Zebrafish as model organisms

(1) It is a small-sized (1–1.5 inches long) freshwater species that is simple to maintain in different environments (Giannaccini et al., 2014).

(2) It has a short generation time, reaches adulthood within three months (Kimmel et al., 1995), and breeds almost all year round.

(3) It has a high fecundity of 200–300 eggs in one morning, and if appropriately maintained, they can provide this yield every 5–7 days (Hill et al., 2005).

(4) Small and transparent eggs (1.0-1.2 mm diameter) can be easily observed using light microscopy (Giannaccini et al., 2014).

(5) Faster embryonic development, since embryos hatch within 48-72 hours post-fertilization (hpf) and their main organs are well developed within 120 hpf (Kimmel et al., 1995).

(6) Much has already been written about the development (Westerfield, 1998) and ecotoxicology of this species (Hallare et al., 2005).

(7) 70% of zebrafish genes have similarity with human orthologues (Hill et al., 2005; Pereira et al., 2020).

Conclusion

In recent years, the zebrafish (Danio rerio, Hamilton–Buchanan, 1822) model has proven to be a simple and successful model for toxicity evaluation. The preliminary step in environmental monitoring and restoration is to assess the degree of pollution in the ecosystem and zebrafish can be effectively used for eco-toxicological studies of the environmental pollutants.

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Article ID: 292

CONTRACT FARMING: A CRITICAL ASPECT IN AGRICULTURE

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Introduction

Contract farming is an agreement between farmers and processing and/or marketing firms for the production and supply of agricultural products under forward agreements, frequently at predetermined prices. The arrangement also invariably involves the purchaser in providing a degree of production support through, for example, the supply of inputs and the provision of technical advice. The basis of such arrangements is a commitment on the part of the farmer to provide a specific commodity in quantities and at quality standards determined by the purchaser and a commitment on the part of the company to support the farmer's production and to purchase the commodity. The intensity of the contractual arrangement varies according to the depth and complexity of the provisions in each of the following three areas:

- *Market provision:* The grower and buyer agree to terms and conditions for the future sale and purchase of a crop or livestock product;
- *Resource provision:* In conjunction with the marketing arrangements the buyer agrees to supply selected inputs, including on occasions land preparation and technical advice;
- *Management specifications:* The grower agrees to follow recommended production methods, inputs regimes, and cultivation and harvesting specifications.

With effective management, contract farming can be a means to develop markets and to bring about the transfer of technical skills in a way that is profitable for both the sponsors and farmers. The approach is widely used, not only for tree and other cash crops but, increasingly, for fruits and vegetables, poultry, pigs, dairy produce and even prawns and fish. Indeed, contract farming is characterized by its "enormous diversity" not only with regard to the products contracted but also in relation to the many different ways in which it can be carried out.

The contract farming system should be seen as a partnership between agribusiness and farmers. To be successful it requires a long-term commitment from both parties. Exploitative arrangements by managers are likely to have only a limited duration and can jeopardize agribusiness investments. Similarly, farmers need to consider that honouring contractual arrangements is likely to be to their long-term benefit.

Contract farming is becoming an increasingly important aspect of agribusiness, whether the products are purchased by multinationals, smaller companies, government agencies, farmer cooperatives or individual entrepreneurs. As noted above, the approach would appear to have considerable potential in countries where small-scale agriculture continues to be widespread, as in many cases small-scale farmers can no longer be competitive without access to the services provided by contract farming companies. It must be stressed, however, that the decision to use the contract farming modality must be a commercial one. It is not a development model to be tried by aid donors, governments or non-governmental organizations (NGOs) because other rural development approaches have failed. Projects that are primarily motivated by political and social concerns rather than economic and technical realities will inevitably fail. Tamil Nadu has become the first State in India to enact a law on contract farming.

Advantages and problems of contract farming

Advantages for farmers

- Inputs and production services are often supplied by the sponsor.
- This is usually done on credit through advances from the sponsor.Contract farming often introduces new technology and also enables farmers to learn new skills.
- Farmers' price risk is often reduced as many contracts specify prices in advance.
- Contract farming can open up new markets which would otherwise be unavailable to small farmers.

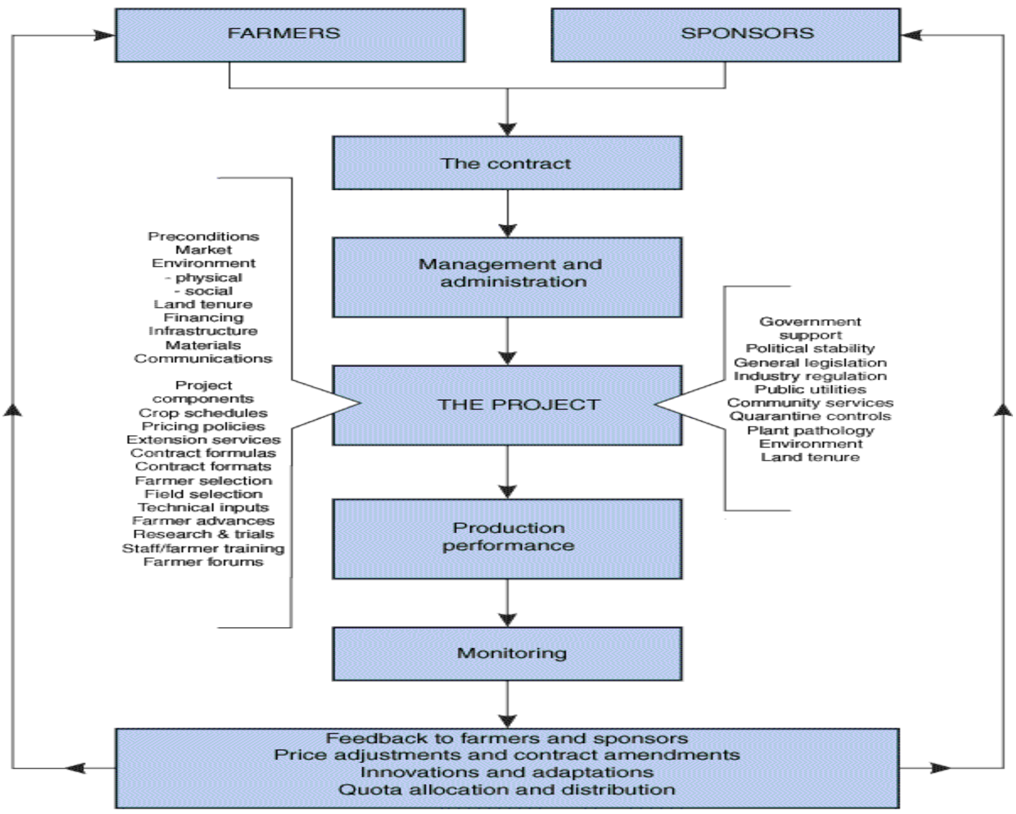

Fig. a framework of contract farming

Problems faced by farmers

- Particularly when growing new crops, farmers face the risks of both market failure and production problems.
- Inefficient management or marketing problems can mean that quotas are manipulated so that not all contracted production is purchased.
- Sponsoring companies may be unreliable or exploit a monopoly position.
- The staff of sponsoring organizations may be corrupt, particularly in the allocation of quotas.

SPONSOR

Advantages for sponsors

- Contract farming with small farmers is more politically acceptable than, for example, production on estates.
- Working with small farmers overcomes land constraints.
- Production is more reliable than open-market purchases and the sponsoring company faces less risk by not being responsible for production.
- More consistent quality can be obtained than if purchases were made on the open market

Problems faced by sponsors

- Contracted farmers may face land constraints due to a lack of security of tenure, thus jeopardizing sustainable long-term operations
- Social and cultural constraints may affect farmers' ability to produce to managers' specifications
- Poor management and lack of consultation with farmers may lead to farmer discontent
- Farmers may divert inputs supplied on credit to other purposes, thereby reducing yields

CONCLUSION

One way to create an agrarian economy that provides a billion people with food and nutrition security is through contract farming. It is a practical alternative farming strategy in India that may offer the farmers guaranteed and trustworthy input services and desired farm products to the contracting firms. Additionally, it facilitates the backward and forward market linkages that form the basis of commercial agriculture, particularly to address many of the issues small farmers have with access to markets. Physical infrastructure, telecommunications infrastructure, land availability and tenure, input availability, and social concerns are all factors that contracting companies must take into account. However, contract farming is currently a win-win situation for businesses and farmers alike. The prospects for contract farming in India are generally favourable as a result of expanding middle-class awareness of food safety and quality, as well as the quality demands of the export market in the developed countries. In addition to the already-existing strategies for connecting farmers with consumers, this endeavour is crucial for lowering transaction costs by developing connections between farmers and processors. Therefore, the government should set up a monitoring system and a dispute resolution agency to make sure that both parties follow the contract's provisions.

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