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PLANT ANTIMICROBIAL PEPTIDES: A NOVEL APPROACH AGAINST DRUG RESISTANT MICROORGANISMS

Mr. Priyadarshini Karre[1], Ms. Yerravalli Priyanka[2], Mrs. Sayeeda Tabassum[3]

Assistant professor[1,2,3]

Department of Pharmacology,

Global College of Pharmacy, Hyderabad. Chilkur (V), Moinabad (M), Telangana- 501504.

ABSTRACT: Plant antimicrobial peptides, or antimicrobial peptides (AMPs), are critical components of host defense systems. AMPs are present in a wide variety of species, from microbes to humans, and they include thionins, defensins, haveins, and others. Glucose transporter, shrimp for animals.An invaluable source of natural antimicrobials, plants.

INTRODUCTION:

Antibiotics have saved millions of lives over the years, but their efficacy is under jeopardy due to the fast rise of resistant bacteria. The emergence of superbugs due to antibiotic resistance has been linked to many factors, including the following: the abuse and misuse of pharmaceuticals; the continued use of the same chemical moieties as antibiotics; and the pharmaceutical industry's failure to create new treatments.

Multiple drug-resistant TB was responsible for the deaths of around 170,000 persons worldwide in 2012, according to a study by the World Health Organization. 4. A strong method is required to counter the increasing number of bacteria that are resistant to antibiotics, since the emergence of resistant human pathogenic fungal species is making treatment strategy planning more difficult. Some have compared antimicrobial peptides (AMPs) to long-lost evolutionary weapons that fought against infections caused by microbes. Innate immunity in bacteria6, insects7, plants8, and mammals 9 relies heavily on antimicrobial peptides. Natural products derived from plants have long been an important part of human health maintenance, particularly in the area of infection management. 10. There are several therapeutic plants that contain antimicrobial chemicals; one of these molecules is plant AMP, which is known as PAMPs. Multi Drug Resistant (MDR) infections are presently challenging to cure; however, PAMPs have the potential to be a game-changer in the creation of novel antibiotics by blocking the MDR pump. Although there is a small risk that bacteria may acquire resistance to these peptides 11, PAMPs are a promising option because of the wide range of effects they have on germs. PAMPs are tiny, positively charged peptides that have antibacterial properties; they consist of 10–50 amino acids and have a mass of around 2–9 kDa. The primary building blocks of these peptides are hydrophobic amino acids arranged in a helical fashion. 12. While most of the time PAMPs target microbes, certain AMPs have the ability to attack herbivorous insects. 13. Different families of PAMPs are defined by similarities and differences in amino acid sequences, identities, cysteine residue numbers, and spacing between them. twelve, fourteen. The electrical charges of these peptides allow them to be generally categorized as either anionic (AAMPs) or cationic (CAMPs). 15. Among PAMPs, there are six main families. Thionins, defensins, cyclotides, snakins, and lipid transfer proteins are all part of this class. and hevein- like peptides [12].

Mode of Action: We still don't know how AMPs work exactly. A number of researchers have put out competing theories on the action mechanism of antimicrobial peptides. Their antibacterial effect is attributed to certain properties of these peptides, such as their size, cationic charge, and amphipathicity. One possible explanation for how AMP works is that it interacts with lipid molecules on the surface of bacteria and/or other targets in the cytoplasm, leading to membrane breakdown (16–19).Additionally, Yeaman and Yount postulated that AMPs' charge affinity for cations is probably a significant component, giving AMPs their selectivity (n= 19). The "barrel-stave" (membrane pore production), "carpet-like" (membrane destruction/solubilization), and toroidal (wormhole) models were among the several that were suggested based on the mechanism of action of AMPs (Fig. 1).A peptide attaches to the acidic surface of the bacterial membrane, causing membrane breakdown in the "carpet-like" process. 20. Alignment of the peptide inside the phospholipid membrane of bacteria determines the antibacterial specificity of the peptide. According to Matsuzaki et al., the magainin-2 peptide forms channels (pores) by binding laterally to the surface of the membrane, which triggers the translocation of phospholipid bilayers (21, 22). influenced by both the physicochemical characteristics of the peptides and the phospholipids found in bacterial membranes (23).The "toroidal (or wormhole) model" suggests that AMPs might collect on the membrane, creating "bends" where the bilayers of phospholipids can connect to create pores 25, 26.

FIG.1:DIFFERENTMODELSSHOWINGACTIONOF ANTIMICROBIAL PEPTIDES (AMPs) CAUSINGLIPID MEMBRANE PERMEABILIZATION

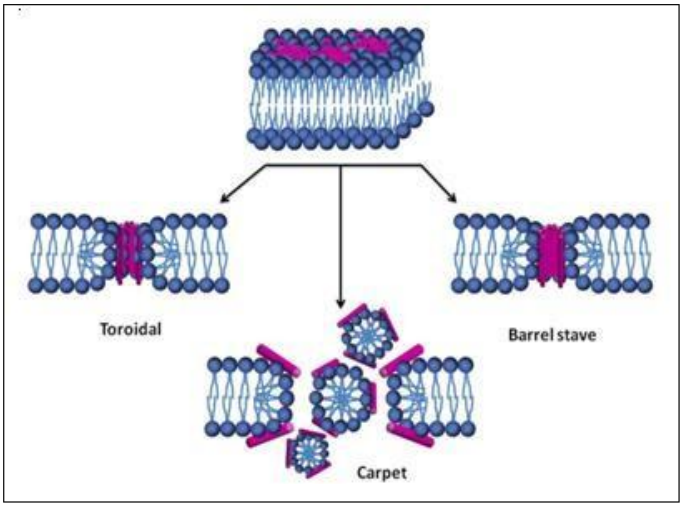
Initial steps include peptide structural changes brought on by assimilation at the membrane surface. Once a peptide concentration reaches a certain threshold, one of these three techniques will be used to break the membrane. Molecules of AMP insert themselves perpendicularly into the membrane in the Barrel-Stave model. An opening is left behind in the membrane when AMP molecules attach to a section of the membrane that has hydrophobic sides pointing inward, according to the Carpet model. Molecules of AMP constantly come into contact with the head groups of phospholipids in the toroidal pore model 27, 28.

These theories propose that membrane pores are formed when peptide concentrations reach certain critical thresholds. When holes form in the membrane, ions and metabolites seep out, the respiratory process is disrupted, and cell wall production is interrupted, which eventually leads to the membrane collapsing. 19.

In microbial cell membranes, cationic peptides engage in electrostatic interactions with negatively charged molecules such phospholipids, lipopolysaccharides (LPS), and teichoic acid. In addition, some receptors on the cell surface allow the cationic residues to bind with lipids in the membrane 15, 17, 19. The fact that certain bacteria are able to persist in the presence of permeable membranes for long periods of time suggests that cell death occurs via processes other than membrane lysis. One of these non-membranolytic mechanisms is the binding of polymerase chain reaction (PAMP) molecules to certain proteins, such as replicating enzymes, transcription factors, cofactors, etc., which in turn causes a halt in DNA synthesis, transcription, and translation, and finally cell death.

SixMainFamiliesofPAMPS:

Thionins: With a molecular weight of around 5 kDa and an abundance of arginine, lysine, and



cysteine residues, thionins are positively charged proteins. Their structure is made up of two α-helices that are not parallel to one other and a β-sheet that is not parallel to each other either, with three or four disulfide connections in between. Cell breakdown and eventual cell lysis may occur when the Tyr 13 residue, located in the groove between the α-helices and β-sheets, interacts with lipid molecules of the membrane (12, 29). Type I, Type II, Type III, Type IV, and Type V thionins are the five main categories into which thionins fall. Bacteria (30), fungus (31), mammal cells (32), and insect larvae (33), among other organisms, are poisonous to thionins. Eight or six cystine residues, which will be called 8C or 6C thionins 34 from now on, make up α/β thionins.

The preproteins consist of these secreted peptides, and the prothionin domain has conserved sequences at both the N- and C-termini that act as signaling peptides and acidic domains, respectively, 35–37. Thionins are molecules containing eight cystine residues that are held together by four disulfide bonds. These connections connect β1 to the C-terminal coil, the end of β1 to the beginning of β2, α1 and the loop following α2, and α1 and α2 are linked by Cys IV-Cys V. But three disulfide bridges keep six-cysteine thionins stable. The disulfide link between Cys II and Cys VII is not present in these peptides (34, 38–40). Most of the amphibian-like phytoestrogens (AMPs) found in plants are members of the thionin and defensin families. Purothionin, one of the earliest basic polypeptides, was found in the endosperm of cereals including wheat (Triticum aestivum) and others. It inhibited the development of some phytopathogens like Xanthomonas, Corynebacterium, and Pseudomonas.

The α and β purothionins were isolated from a crude extract of purothionin 30, which had been partly purified, using carboxy methylcellulose column chromatography. Thionin, a compound produced by Pyrularia pubera 32, was extracted from the plant's nuts. The 47 amino acids that made up this peptide had a basic character and included 4 disulfide linkages in addition to 2 tyrosine residues. Notable hemolytic, cytotoxic, and neurotoxic effects were associated with this peptide. They demonstrated that pyrularia thionin loses its hemolytic activity, cytotoxic activity, and lethality when iodinated in mice. Cell death is the end outcome of this peptide's chain reaction, which begins with membrane depolarization and continues with a channel-mediated influx of Ca2+ ions that activate phospholipase A2. This research found that iodination inhibits all three of these cellular reactions, which might mean that tyrosine is involved in either preserving the structure of the peptide or facilitating its connection with the cell membrane.

From maize kernels, Duvick and colleagues were able to extract the MBP-1 antimicrobial peptide, which is a member of the thionin family of AMPs (41). Packed inside this peptide are 33 amino acid residues, totaling 4127.08 Da. Fusarium moniliforme Sheld. and Fusarium graminearum are both inhibited in their spore germination and hyphal elongation processes by MBP-1. Clauibacter michiganense sp. Nebraskense was another bacterium that these peptides shown antibacterial action against. Chitin affinity chromatography and reverse-phase high performance liquid chromatography (HPLC) 42 were used to purify novel thionin antimicrobial peptides Tu-AMP1 and Tu-AMP2, with molecular weights of 4,988 Da and 5,006 Da, respectively, from bulbs of Tulipa gesneriana L.

The antimicrobial peptides Pp-AMP 1 and Pp-AMP 2, which were isolated from Japanese bamboo shoots (Phyllostachys pubescens) by means of a straightforward chitin affinity chromatography process, were shown to have action against harmful bacteria and fungi. The homology between Pp-AMP1 and Pp-AMP2 and mistletoe toxins was observed. 43. During the crystallization process, the cytotoxic thionin peptides viscotoxin A1 and B1 from mistletoe both formed dimers. While Viscotoxin A1 (net charge +6) 44 exhibited no connection, Viscotoxin B2 (net charge +4) coordinated with sulfate or phosphate anions during dimer formation. European mistletoe (Viscum album) has viscotoxins that inhibit the growth of cancer cells and phytopathogenic fungi. 45.The viscotoxin peptides were fused with thirteen different proteins, including versions of GB1 with His6 tags, ZZ tags, Z tags, maltose binding protein, NusA, glutathione S-transferase,

thioredoxin, green fluorescent protein, and periplasmic and cytosolic versions of Disulfide bond C and Disulfide-bond A oxidoreductase-like protein, in order to be expressed in Escherichia coli cells. The viscotoxin-thioredoxin protein fusion produced the most soluble viscotoxin. It was discovered that HeLa cells were poisonous to recombinant viscotoxins made utilizing the aforementioned procedure. Here are a few more thionins: in **Table 1**.

TABLE 1: LIST OF THIONIN PEPTIDES SUBMITTED TO PHYTAMP DATABASE HAVING ANTIMICROBIAL ACTIVITY AGAINST TARGET ORGANISM

S.no.	Name	Organism	Activity	References
1	Viscotoxin-B (Viscotoxin-B2(VtB)Viscotoxin-a3(Vta3)	*Viscum album*	Antifungal	46 -48
2	Pp-AMP1 Pp-AMP2	*Phyllostachys pubescens*	Antibacterial Antifungal	43
3	Tu-AMP1	*Tulipa gesneriana*	Antibacterial Antifungal	42

Defensins: Defensins are tiny cationic peptides of 45–54 amino acids, which are ubiquitous and have a molecular weight of around 5 kDa. You may find these peptides expressed in many sorts of plant parts, but seeds have the highest concentration. The presence of defense peptides in seeds enhances their resilience during germination and protects them against fungal infections (49, 50). Defensins were formerly categorized as ϒ-thionins and ϒ1/ ϒ2 purothionins due to a 32-36% sequence similarity with ű/α thionins 51, 52. Plant defensins are structurally identical to insect defensins and scorpion poisons, which number fourteen.

From a variety of plant sources, many AMPs belonging to the defensin family have been isolated. Isolated from Solanum tuberosum is a 5kDa polypeptide called Pseudothionin-St1 (Pth-Stl). Results showed that Clavibacter michiganensis, Pseudomonas solanacerum, and Fusarium solani were all susceptible to the Pth-St1 peptide. None of the properties shown in genuine thionins were present in pseudothionin. The reclassification of pseudothionin under defensins 53 has taken place. From radish and dahlia seeds, Thevissen and colleagues extracted the antifungal defensin Rs-AFP2 in 1996 and Dm-AMP1 in 1997. Fungal cell rupture occurred as a result of fast K+ ion efflux, Ca2+ ion inflow, alteration of fungal cell membrane potential, and alkalization of medium brought about by treatment of Neurospora cassa hyphae with Rs AFP2 and Dm-AMP1. In the N. crassa membrane, Rs-AFP2 produces hyperpolarization while Dm-AMP1 causes depolarization. The IC50 value against Saccharomyces cerevisiae 55 was found to be about 2 µM when an antifungal peptide Dm-AMP1 was extracted from Dahlia merckii seeds. One way to create [35S] Dm-AMP1 was by radioactively labeling antifungal peptides using t-butoxyczrbonyl-[35S]- L-methionine N-hydroxy-succinimidylester (Boc-Meth-NHS). Section 35: The antifungal activity of Dm-AMP1 peptide was comparable to that of Dm-AMP1 peptide.

It was shown that the addition of 35 S to Dm-AMP1 had no effect on the peptide binding sites. Fungal cell death is caused by Dm-AMP1 binding to particular locations on their plasma membrane. This study paves the way for the monitoring and, more importantly, the mechanism of action of defensin peptides in different species without interfering with their function.

According to research by Thevissen et al., glucosylceramides found in the cell membranes of fungus and yeast cause Rs-AFP2 to interact, leading to cell death by membrane permeabilization. Defensin 1 (PhD1), which is derived from Petunia hybrida flowers, is an amino acid sequence with 47 cystine-rich residues and five disulfides bonds for stability. According to 1H NMR spectroscopy, the structure of PhD1 is an α-helix, a β-sheet with three strands that are anti-parallel to each other, and 57 cystine-stabilized α β motif folds. The first plant defensin to demonstrate insecticidal action was 59 the Vigna radiata (mung bean) defensin 1 (VrD1). The α-helix, triple-stranded antiparallel β-sheet, and 310 helix structure of VrD1 are held together by four disulfide bridges that form a distinctive cysteine-stabilized α β

motif. In Tenebrio molitor, VrD1 inhibits the activity of α-amylase. Complete resistance to bruchids was also shown by the production of α-amylase inhibitor 1 in transgenic pea.

The antifungal peptides RsAFP2, HsAFP1, and PvD1 were found to suppress the growth of S. cerevisiae and Candida albicans, respectively, and were extracted from the plants Raphanus sativus, Heuchera sanguinea, and Phaseolus vulgaris. The peptide-glucosylceramide (GlcCer) bonding process in fungal and yeast cell membranes

oxidative stress on cells, associated with the generation of nitric oxide (NO) and reactive oxygen species (ROS) 59–61.

Nicotiana alata NaD1 defensin inhibited Candida albicans growth 62. The permeabilization of fungal cell walls is caused by NaD1's interaction with these walls. The entry of NaD1 into the cytoplasm triggers an overabundance of ROS, which cause oxidative damage, in the cell. An essential function in protecting fungal cells against NaD1 is the high osmolarity glycerol (HOG) pathway. This suggested that the HOG pathway would be an appropriate place to aim the antimicrobial peptides in order to enhance their efficacy against Candida albicans.

The cowpea, or Vigna uniguiculata, is the source of the putative defensin gene. The mature putative defensin peptide exhibited similarities to a normal defensin peptide in terms of sequence, amino acid arrangement, splicing analysis, secondary and tertiary structures. Using real-time polymerase chain reaction (RT-PCR) 63, researchers compared pathogen-treated and untreated plants in response to biotic stimuli by measuring gene expression levels for a defensin. There is a catalog of defensin peptides that details where they came from and how they work against various microbes. **Table 2**.

TABLE 2: LIST OF DEFENSIN PEPTIDES SUBMITTED TO PHYTAMP DATABASE HAVING ANTIMICROBIAL ACTIVITY AGAINST TARGET ORGANISM

S.no.	Name	Organism	Activity	Targetorganism	References
1	Floraldefensin-likeprotein 1 (PhD1) Floraldefensin-likeprotein 2 (PhD2)	*Petunia hybrida*	Antifungal	*Fusarium Oxysporum, Botryris cinerea*	64
2	VrD1	*Vigna radiata*	Antibacterial Antifungal	*Escherichiacoli* *Rhizoctoniasolani*	65
3	DefensinJ1-1 DefensinJ1-2	*Capsicum annuum*	Antifungal	*Fusariumoxysporim, Botrytiscinera*	66,67
4	AntifungalproteinAX1 AntifungalproteinAX2	*Beta vulgaris*	Antifungal	*Cercosporabeticola* andotherfilamentousfungi	68
5	Fabatin-1 Fabatin-2	*Vicia faba*	Antibacterial	*Bacillus subtilis, Enterococcushirae, Escherichia coli Pseudomonasaeruginosa*	69
6	Rs-AFP1 Rs-AFP2	*Raphnus sativus*	Antifungal	*Alternariabrassicola, Botrytis cinerea, Fusarium culmorum, Fusariumoxysporum, Pyriculariaoryzae, Verticiliiumdahlia,*	51,70,71
7	Flowerspecificgamma-thionin(Nad1)	*Nicotiana tabacum*	Antifungal	*Fusariumoxysporum Botrytiscinerea*	72,73

Cyclotides: With 28–37 amino acid residues, cyclotides make up the biggest family of circular proteins. The six conserved cysteine residues that make up the cyclic cystine knot (CCK) motif are present in these peptides. Besides their antibacterial effects, cyclotides have been shown to have insecticidal and anti-cancer properties 74. Isolated from the African shrub Oldenlandia affinis 75 was the first water-soluble oxytocic polypeptide, kalata B1, which has about 30 amino acids. Since kalata B1 was found to be the primary uterotonic component of the plant, it was employed by the locals to induce labor. Another unique cyclotide called "kalata B8" was also

found in Oldenlandia affinis 76. While Kalata B8 does show anti-HIV efficacy, it does not have any hemolytic effects. The peptide's hydrophilic properties might explain this. The seeds of the Mirabilis jalapa L. plant yielded two antimicrobial peptides, Mj-AMP1 and Mj-AMP2. There are 37 residues in Mj-AMP1 and 36 in Mj-AMP2, for a total of 77.

There are three disulfide bridges in this fundamental molecule. Although these peptides were inactive against gram-negative bacteria and human cells in vitro, they exhibited broad-spectrum antifungal and anti-gram-positive bacterial action. Even after increasing the dosage several times, the Mj-AMPs did not impact the transmission of pulses in insect neurons, despite the fact that their sequence was identical to ¬-agatoxins, an insecticidal neurotoxic peptide derived from spider venom. The coffee plant was used to extract four macrocyclic cystine-knot peptides: kalata, circulin A and B (CirA and CirB), and cyclopsychotride. The minimum inhibitory concentration (MIC) for Staphylococcus aureus against Kalata and CirA was determined to be 0.2 µM.

Kalata and Cir A, on the other hand, had no effect on E. coli and Pseudomonas aeruginosa. The good news is that both Gram-positive and Gram-negative bacteria were discovered to be susceptible to CirB and cyclopsychotride. Although none of the four cyclic peptides worked against albicans, they fared somewhat well against kefyr and tropicalis Candida.

In a study conducted by Viola hederaceae (vhl) 79, four unique cyclotides, which are proteins with macrocyclic knots, were discovered. vhl-1 was a 31-residue cyclotide that was unique to leaves. The HIV-virus was shown to have an EC50 value of 0.87M for vhl-1. Isolated from Luffa cylindrical 80 seeds was the ribosome-inactivating peptide known as Luffin P1. The C8166T cell lines, which are infected with HIV-1, were discovered to be susceptible to Luffin P1. The structural unit of Luffin P1 is a helix-loop-helix motif with alpha helices connected by a two-didulfide link. Luffin P1 used N-glycosidase activity to destroy cells that were infected with HIV. The seeds of the buckwheat plant, Fagopyrum esculentum, were used to isolate a trypsin inhibitor (BWI-2c).

BWI-2c peptide comprises of 41 amino acid residues with two disulfide bonds and shows structural similarity with VhT1 peptide isolated from *Veronica hederifolia*. Both these represents new family of protease inhibitors.**Table 3** depicts cyclotides peptides submitted to PhytAMP Database having antimicrobial activity against target organism.

TABLE3: LISTOF CYCLOTIDESPEPTIDESSUBMITTEDTO PHYTAMPDATABASEHAVING ANTIMICROBIAL ACTIVITY AGAINST TARGET ORGANISM

S.no.	Name	Organism	Activity	Targetorganism	References
1	CycloviolacinH-2 CycloviolacinH-3	*Viola hederacea*	Antiviral	HIVvirus	79
2	Kalata- B8	*Oldenlandiaaffinis*	Antiviral	HIVvirus	76
3	Circulin–F(CIRF) Circulin–E(CIRE) Circulin–D(CIRD) Circulin–C(CIRC)	*Chassalia parviflora*	Antiviral	HIVvirus	82
4	Cycloviolin-A Cycloviolin-B Cycloviolin-C Cycloviolin-D	*Leoniscymosa*	Antiviral	HIV-1 virus	83
5	Kalata- B2	*Oldenlandiaaffinis*	Insecticidal	*Hellicoverpaarmigera*	84, 85
6	Circulin-A Circulin-B	*Chassalia parviflora*	Antibacterial Antifungal	*Micrococcusluteus,Escherichiacoli, Proteus vulgaris,Klebsiellaoxytoca* *Candidaalbicans,Candidakefyr, Candidatropicalis*	86-89

Snakin: Both monocotyledonous and dicotyledonous plant species include Snakins Protein (SN), a distinct class of antimicrobial peptides rich in cysteine. In contrast to the other identified families, which only include two or four disulfide bridges, peptides belonging to this family possess six possible disulfide bridges. Snake venoms that are comparable to hemotoxic desintegrin have an amino acid sequence with the snakin-1 peptide. The 9-kDa radish seed peptide, which has sequence similarities with other plant species' nonspecific lipid transfer proteins, has antifungal properties. It was later determined that this peptide belonged to the snakins family, namely 91.

Against many fungi, the peptide demonstrated antifungal efficacy. Isolated from potato tubers 90 was Snakin-1 (StSN1), which has 63 amino acid residues. This featured a small core hydrophobic region of 25 to 30 amino acid residues, was quite basic, and was encircled by lengthy, very polar domains at the N- and C-termini. Research has shown that this peptide may inhibit the growth of plant-harming bacteria and fungi. 'Kistrin' snake venom, which is hemotoxic, shared sequence similarities with StSN1.

In a similar vein, 66 amino acid residues made up the snakin-2 (StSN2) peptide that was isolated from potato (Solanum tuberosum cv Jaerla) tubers; it had a molecular weight of 7,025 Da. The cDNA of the mature StSN2 peptide encodes for a signal sequence, which is preceded by a sequence of fifteen acidic residues. It was found that StSN2 was effective against all of the picked fungus and gram-positive bacterial pathogens for the potato plant, but it was found to be ineffective against a few Gram-negative bacteria. When combined with StSN2 snakin peptide, St-PTHl defensin and StSN1 snakin peptides accumulated to toxic levels, protecting potato plants against bacterial and fungal diseases (53, 90, 92).

The 31-amino acid Snakin-Z peptide, isolated from Zizyphus jujube fruit, had antibacterial efficacy against bacteria and fungus but had no impact on human red blood cells, according to a study 93. The results suggested that Snakin-Z could be an effective antibacterial peptide for medical use. Several other snakin peptides may be found in

Table 4.

TABLE4:LISTOFSNAKINPEPTIDESSUBMITTEDTOPHYTAMPDATABASEHAVINGANTIMICROBIAL ACTIVITY AGAINST TARGET ORGANISM

S.no.	Name	Organism	Activity	Targetorganism	References
1	Snakin-1 (StSN1)	*Solanum tuberosum*	Antibacterial	*Clavibactermichiganensis, Ralsotonia solanacearum*	90,92
	Snakin-2 (StSN2)		Antifungal	*Botrytis cinerea, Fusarium solani, Fusariumculmorum,Fusariumoxysporumf.*sp. *Lycopersici, Plectosphaerella cucumeria, Collectotrichumgraminicola,Aspergillusflavus*	

Hevein Like Peptides: Miniature chitin-binding peptides with a molecular weight of 4.7 kDa and 43 amino acid residues are known as hevein-like peptides. It was from the latex of the rubber tree (Hevea brasiliensis) that the hevein protein was first isolated and studied. Amino acid residues containing cystine make up 19.2% of these peptides. Through its binding to chitin 95, this protein hinders the hyphal development of fungus. Avesin An oat seed peptide contains a chitin binding domain and a conserved domain with 37 amino acids sharing a similar structural pattern, including many glycine and cysteine residues. When tested on a small number of fungal strains, Avesin A had a modest antifungal effect. Isolated from the leaves of Wasabia japonica L. 97 is the hevein-like peptide Wj AMP-1. As an added bonus, Wj AMP-1 had antifungal and antibacterial properties. Sequence comparisons with hevein-like proteins from Hevea brasiliansis and Arabidopsis thaliana revealed that Wj AMP-1 had 60% and 70% sequence similarity, respectively. Two hevein homologues, Pn-AMP1 and Pn-AMP2, were isolated from Pharbitis nil and had antifungal properties against fungi that contain and do not contain chitin.

Tomato plants transgenic with Pn-AMP2 cDNA produced under the control of the CaMV35S promoter exhibited enhanced resistance to both non-chitinous and chitin-containing fungi 98. This provided further evidence that Pn-AMP's antifungal action may involve more than just chitin. Deoxyribonucleic acid from Eucommia

(EAFP2) extracted from the olive tree of Eucommia ulmoides has a hydrophobic face and a distinctive disulfide connection between its N- and C-terminal 99 residues, making it a chitin binding domain. Isolated from Triticum kiharae 100 seeds were two novel antimicrobial peptides, WAMP-1a and WAMP-1b. A single amino acid residue at the C-terminus is the only difference between WAMP-1a and WAMP-1b. The recombinant WAMP-1a, which was engineered in Escherichia coli, has a wide range of inhibitory effects against both chitin-containing and chitin-free bacteria and viruses. The chitin-binding polypeptide WAMP-1a, which exhibited similarities to hevein-like peptides 101, was isolated from Triticum kiharae. The WAMP-1a peptide demonstrated antimicrobial and antifungal properties. The carbohydrate binding effectiveness of WAMP-1a is reduced owing to a replacement of a conserved serine residue with a glycine residue. Alternanthera sessilis 102 was used to identify and describe six hevein-like peptides rich in cysteine, aSG1-G3 and aSR1-R3. There are six cysteine residues, seven glycine residues, four proline residues, and a conserved chitin-binding domain, according to proteomic research. Three disulfide linkages were detected in a cysteine knot motif according to nuclear magnetic resonance (NMR) research. Here are a few hevein-like peptides with antimicrobial action that were submitted to the PhytAMP Database: (Table 5).

TABLE5:LISTOFHEVEINLIKEPEPTIDESSUBMITTEDTOPHYTAMPDATABASEHAVING ANTIMICROBIAL ACTIVITY AGAINST TARGET ORGANISM

S.no.	Name	Organism	Activity	Targetorganism	References
1	AC-AMP1 AC-AMP2	*Amaranthus caudatus*	Antibacterial	Grampositivebacteria:*Bacillusmegaterium, Sarcina lutea*	103
			Antifungal	*Alternaria brassicola,Ascophyta pisi, Botrytis cinera,Fusarium culmorum*	
2	Ee-CBP (Bark) Ee-CBP(leaves)	*Euonymus europaeus*	Antifungal	*Fusarium culmorum, Fusarium oxysporum, Mycosphaerellaeumusae,Neurosporacrassa*	104
3	Ar-AMP	*Amaranthus retroflexus*	Antifungal	*B.cinerrea,F. Culmorum, H. satium, A. consortiale*	105
4	EAFP2 EAFP1	*Eucommis ulmoides*	Antifungal	*Phytophthorainfestans,Ascochytalycopersici, Alternaria nicotianae, Fusarium moniliforme, Fusariumoxysporum*	99,106,107
5	PN-AMP-1 PN-AMP-2	*Ipomoea nil*	Antibacterial Antifungal Anti yeast	*Bacillus subtilis* *Botrytiscinerea,Fusariumoxysporum, Phytophthora parasitica, Pythium sp.* *Saccharomyces cerevisae*	108
6	Fa-AMP1 Fa-AMP2	*Fagopyrum esculentum*	Antibacterial Antifungal	*Clavibactermichiganesis,Erwiniacarotovora, Agrobacteriumradiobacter,Agrobacterium rhizogenes* *Fusariumoxysporum,Geotrichumcandidum*	109

Lipid transfer proteins (LTPs): The nonspecific lipid transfer proteins (ns-LTPs) are small proteins (approximately 8.7 KDa) composed of 90 amino acids preserved by four disulfide bonds accompanying central hydrophobic shaft. Due to small size of LTPs, they are able to penetrate through the fungal membrane and create pores that lead to efflux of intracellular ions and finally cell death [110].

Tassin*etal*.,isolatedantifungal*Ace*-AMP1peptide from the seeds of onion [111]. NMR spectroscopy revealed that *Ace*-AMP1 peptide comprises of 93 amino acid residues and four disulfide bonds. Structure of *Ace*-AMP1 peptide consist of four

helices connected by three loops and a C-terminal tail without secondary structures, except for 3_{10}- helix turn and a β turn.

LTPs of 9 kDa cysteine-rich cationic peptide was isolated from *Vigna unguiculata* seeds [112]. These peptides play an important role in plant

defense mechanism against microbial infection. Lin *et al*., isolated a novel antifungal lipid transfer peptide from the seeds of *Brassica campestris* [113]. This peptide hinders the mycelial growth in *Fusarium oxysporum* and *Mycosphaerella arachidicola.* Similarly, lipid like protein of approx. 9 kDa was isolated from the seeds of chilli pepper [114].

This peptide showed antifungal activity against *Fusariumoxysporum,Colletotriumlindemunthianum , Saccharomyces cerevisiae, Pichia membranifaciens, Candidatropicali*and*Candidaalbicans.*Thispeptide causes multiple changes in the morphology of *P. membranifaciens*. Some of the other lipid transfer peptides are listed in **Table 6**.

TABLE6:LISTOFLIPIDTRANSFERPEPTIDESSUBMITTEDTOPHYTAMPDATABASEHAVINGANTIMICROBIAL ACTIVITY AGAINST TARGET ORGANISM

S.no.	Name	Organism	Activity	Targetorganism	References
1	La-LTP(LJAFP)	*Leonurus artemisia*	Antibacterial	*Bacillus subtilis Pseudomonassolanacearum Ralstoniasolanacearum*	115
			Antifungal	*Alternariaalternate,Alternariabrassicae, Aspergillus niger, Bipolaris maydis, Botrytis cinerea, Cerospora personata, Colletotrichumgloeosporiodes*	
2	Hv-LTPCw-18 (PKG2316) Hv-LTP-1 (LTP4.1)(CW21)	*Hordeum vulgare*	Antifungal	*Fusarium solani, Pseudomonassolanacearum, Clavibacter michiganensis*	116
3	IWF1(Bv-LTP), JWF2(Bv-LTP2)	*Beta vulgaris*	Antifungal	*Cercospora beticola*	117
4	Ace-AMP1	*Allium cepa*	Antibacterial	*Bacillusmegaterium Sarcinalutea*	118
			Antifungal	*Alternaria brassicola, Ascockyta pisi Botrytis cinerea, Colletotrickum lindemutkianum,Fusariumculmorum, Pyriculariaoryzae*	
5	Pa-LTP1	*Phaseolus aureus*	Antibacterial Antifungal	*Staphylococcusaureus Fusarium oxysporum, Pythium aphanidermatum,Sclerotiumrolfsii*	119,120
6	Lc-LTP4, Lc-LTP8, Lc-LTP6, Lc-LTP5,Lc-LTP2	*Lens culinaris*	Antibacterial	*A.tumefaciens*	121

Some Other Classes of PAMPS: Several scientific organizations have isolated and studied some of the unclassified AMPs. The guava plant's storage glycine-rich peptide (Pg-AMP1) has a molecular weight of 6029.24 Da. Klebsiella and Proteus sp. were both inhibited by PgAMPs1. On the other hand, against fungus, it showed no action. Homology to enterotoxin, an antimicrobial protein from Escherichia coli 122, is seen in the three-dimensional structure of PgAMP1.

A cytotoxic peptide that targets stomach cancer was largely isolated from the hydrolysate of the Euphorbia hirta protein. Two to ten amino acid residues make up these bioactive peptides. Low molecular weight (3KDa) peptides were obtained by ultrafiltration of Euphorbia hirta protein hydrolysate. The little size of these peptides increases their biological effects and their likelihood of passing the intestinal barrier 123. A glycine-rich peptide 124 known as Shepherin I (Shep I) is the chemically produced AMP. It was produced utilizing a normal heating system and the solid-phase technique at 600C. Bacteria and mycelia fungus were unaffected by this peptide, while it worked wonders against Candida species. Shep I's antifungal activity is diminished when its N- or C-terminal portions are truncated. Despite an increase in activity against Saccharomyces cerevisiae, carboxy-amidation of Shep I had no effect on its antifungal action.

Using ion exchange and reverse-phase high performance liquid chromatography (HPLC), a cysteine peptidase (Cg24-I) with a molecular weight of 24,118 Da was extracted from the latex

of Cryptostegia grandiflora. Cg24-I inhibited Fusarium solani 125 growth at a dose of 28.1µg/ml. Using reversed phase-high performance liquid chromatography (HPLC), Mandal et al. isolated Cn-AMP1, Cn-AMP2, and Cn-AMP3 peptides from green coconut (Cocos nucifera L.) water with molecular weights less than 3kDa (858 Da, 1249 Da, and 950 Da, respectively). In comparison to Cn-AMP2 and Cn-AMP3, the Cn-AMP1 peptide shown superior efficacy against Gram-positive and Gram-negative bacteria. Six medicinal plants found in India—Foeniculum vulgare, Cucumis sativus, Ammi majus, Allium ascolinicum, Cichorium intybus, and Rumex vesicarius—were used to extract antimicrobial peptides. A sodium phosphate citrate buffer with pH values of 5.2, 5.8, 6.8, 7.4, and 7.8 and a sodium acetate buffer with a pH of 6.5 were used to create the protein extracts. At a pH of 5.8, seed extracts of Allium ascolinicum showed strong antibacterial activity against 3 different types of bacteria: Proteus vulgaris (17 mm), Escherichia coli (17 mm), and Staphylococcus aureus (15 mm). Rumex vesicarius seed extracts (7.6), Ammi majus (6.8), Cichorium intybus (7.4), Cucumis sativus (7.8), and Foeniculum vulgare (6.5) were also shown to have antibacterial activity. 127.

Fifty seeds were tested for the presence of short peptides with antimicrobial properties by Golla et al. 128. The seeds included soya, barley, maize, jowar, paddy, millets, foxtail millets, red gram, green gram, black gram, groundnut, pea, field bean, and wheat. Liquid nitrogen and phosphate buffer (PBS) treatments were used to extract protein from seeds. The four clinically significant bacteria that were tested for antimicrobial activity were Pseudomonas aeruginosa, Staphylococcus aureus, Escherichia coli, and Klebsiella pneumoniae. Germinated barley, soya, jowar, wheat, and maize seed protein extracts inhibited the growth of gram-positive and gram-negative bacteria.

The "Hispidalin" cationic bioactive peptide was isolated from Benincasa hispida seeds. With 129 amphipathic amino acid residues and a molecular weight of 5.7 kDa, this peptide was rather unusual. Its lack of sulfur-containing amino acids indicates that it is structurally undefined. A gram-negative bacterium called Aeromonas veronii produced a peptide that was 66% identical to hispidalin, which was distinct from plant defensin. The antibacterial activity of hispidalin was shown against a range of bacterial and fungal infections. According to the PhytAMP database 130, a total of 273 antimicrobial peptides have been described.

Information regarding plant antimicrobial peptides, including taxonomic, microbiological, and physicochemical details, may be found in the PhytAMP database. The 271 items in this database are from a variety of plant families, with the majority belonging to the Violaceae and Brassicaceae. Of all the entries, only 102 peptides (36.63%) were found to have a three-dimensional structure. The percentage of plant AMPs that were approved for biological activity was just 39.5%. When tested, 51% of the AMPs were effective against fungal infections, compared to 33% against bacteria and 10% against viruses. These results highlight the need to isolate and characterize new AMPs from plants that are effective against microbes.

Plants in India account for 11.7% of the global flora, with 28% of those plants being indigenous. The Indian System of Medicine recognizes the medicinal use of over 6,000 plant species, both food and non-edible. Despite the abundance of plant and animal life in the Indian subcontinent, there is a dearth of literature detailing the isolation and description of PAMPs.

CONCLUSION: Research done thus far opens the door to the prospect of extracting antimicrobial peptides from both edible and non-edible plant components. These peptides might be effective treatments for a range of human ailments. To combat certain pathogenic microbes, insects, and pests, one may choose, employ, or alter a PAMP from a variety of families, each with its own unique mode of action and target molecules in the organism. When compared to conventional antibiotics, AMPs provide a more effective and broad-spectrum option for infection treatment. Amplification of genes and transgenesis are two practical approaches of increasing synthesis and improving targeted action of AMPs encoded by small genes with conserved sequences.

The study of AMPs has dual purposes: first, in

the medical field, by creating analogs and by-products of natural antimicrobial peptides for use in medication development; and second, in plant genetic engineering, by enhancing resistance to diseases. The need for huge amounts of pesticides used in agriculture is reduced as disease resistance increases. So, AMPs protect plants from pests and diseases while also providing a new, environmentally friendly model for natural antibiotics that may be used therapeutically in healthcare.

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