AGRICULTURAL BIOLOGY,
2023, V. 58, Iss. 5, pp. 789-820
[SEL'SKOKHOZYAISTVENNAYA BIOLOGIYA]

ISSN 2412-0324 (English ed. Online)

ISSN 0131-6397 (Russian ed. Print)
ISSN 2313-4836 (Russian ed. Online)

Reviews, challenges

UDC 632: 571.27

doi: 10.15389/agrobiology.2023.5.789eng
doi: 10.15389/agrobiology.2023.5.789rus

MICROBIAL PROTEINS AS ELICITORS OF PLANT RESISTANCE TO PATHOGENS AND THEIR POTENTIAL FOR ECO-FRIENDLY CROP PROTECTION IN SUSTAINABLE AGRICULTURE

(review)

L.A. SHCHERBAKOVA[1] ✉, V.G. DZHAVAKHIYA[1], Y. DUAN[2], J. ZHANG[2]

[1]*All-Russian Research Institute of Phytopathology,* 5, ul. Institute, pos. Bol'shie Vyazemy, Odintsovskii Region, Moscow Province, 143050 Russia, e-mail larisavniif@yahoo.com (✉ corresponding author), dzhavakhiya@yahoo.com;
[2]*Nanjing Agricultural University,* No. 1 Weigang, Nanjing, Jiangsu province, China, e-mail dyb@njau.edu.cn, 2018202061@njau.edu.cn
ORCID:
Shcherbakova L.A. orcid.org/0000-0003-0254-379X
Duan Y. orcid.org/0000-0002-4183-8729
Dzhavakhiya V.G. orcid.org/0000-0001-8704-0512
Zhang J. orcid.org/0000-0003-2541-4923
The authors declare no conflict of interests
Acknowledgements:
Supported financially by the Russian Scientific Foundation (project No. 22-16-00154)
Final revision received May 29, 2023
Accepted July 20, 2023

Abstract

To combat plant diseases, modern agriculture has a large arsenal of xenobiotic pesticides that are toxic to microorganisms. However, the hazardous effects of such pesticides or their degradation products on the environment and human health urgently require the search for new harmless and environmentally safe means of plant pathogen control. In this regard, the attention of researchers is attracted by the phenomenon of natural plant resistance, including the active plant immunity and natural substances that can induce plant defense mechanisms (J.D. Jones et al., 2006; M. Albert, 2013; L. Wiesel et al., 2014; E.J. Andersen et al., 2018; D.F. Klessig et al., 2018). Various microorganisms, pathogenic or nonpathogenic to plants can serve as sources of such substances, including proteins and peptides. When interacting with them, microbial proteins play the role of nonspecific resistance elicitors recognized by plants as conserved microbial patterns (MAMPs or PAMPs) that induce the first line of active plant defense (basic resistance, or PTI) (C. Zipfel, 2009; M.A. Newman, 2013; J. Guo, Y. Cheng, 2022).. Other microbial proteins play the role of effectors involved in the development of the disease, and, if recognized by host plants, can also activate defense responses as elicitors of race-specific resistance (B.P. Thomma et al., 2011; W. Zhang et al., 2022; B.C. Remick et al, 2023). The perception of microbial protein elicitors by plant receptors causes rapid responses and can lead to the development of prolong systemic resistance in plants (T. Boller, G. Felix, 2009; J.B. Joshi et al., 2022; S. Wang et al., 2023). Studying the properties and mechanisms of action of microbial proteins is new and fast-paced research cluster, which results create the basis for one of the most eco-friendly avenue in the field of plant protection and can lead to the development of novel effective biocontrol agents for sustainable agriculture. Over the past few decades, in nonpathogenic and plant pathogenic fungi, oomycetes, bacteria, and viruses, including those affecting agricultural crops, a number of elicitor proteins have been identified that belong to the MAMP/PAMP type, as well as effectors that induce specific immunity (ETI). The review below summarizes and analyzes information on the most important advances in the identification and studying of elicitor proteins produced by various bacteria, fungi, oomycetes, and viruses (D. Qutob et al., 2003; M. Tarallo et al., 2022; Q. Xu et al., 2022). If the corresponding information is known, the peculiarities of the elicitor structure and their mechanisms of action, namely, defense responses of various plants induced by the corresponding elicitors, are briefly described. We also tried to illustrate the diversity of microorganism species able to produce elicitor proteins, which trigger the mechanisms of both specific and nonspecific resistance. The examples of protein and peptide elicitors, for which both basic and novel data are presented, are described in more details. Such bacterial elicitors include flagellin, harpins (similar and differing effects of harpins and flagellins are described), elongation factor Tu, cold shock proteins; elicitors produced by mycelial fungi include effectors of *Cladosporium fulvum*, elicitors of pathogenic and nonpathogenic Fusarium fungi, and recently discovered MAMPs/PAMPs and ETI-inducing proteins. The review also includes

information on the oomycetal elicitors, microbial enzymes possessing eliciting properties, glycoproteins and peptidoglycans, and vector proteins of viruses (Y. Jin et al., 2021; L. Cai et al., 2023). In addition, the prospects for practical application of microbial elicitor proteins are described in a separate section by the example of commercial preparations based on bacterial and fungal protein elicitors, which have been developed in Russia and China and have proved their protective efficiency under field conditions (Dzhavakhiya et al., 2003; W.P. Liu et al., 2007; J. Mao et al., 2010; Q. Dewen et al., 2017).

Keywords: biogenic elisitors, microbial proteins and peptides, vicrobial patterns, effectors, PTI, ETI, plant defence responces, biocontrol, eco-friendly remedies

In intensive crop production, high crop yields cannot be achieved without combating plant diseases. Currently, there are various approaches to crop protection (creation of resistant varieties, treatment with chemical pesticides, biocontrol of plant pathogens using antagonistic microbes, crop rotation and other agrotechnical measures), among which breeding for resistance and the chemical method are leading [1, 2]. However, environmental pollution combined with food safety problems that arise from excessive or inappropriate use of pesticides [3, 4] and the loss of crop productivity due to overcoming resistance by plant pathogens [5] are of concern worldwide and stimulate for replenishing the arsenal of reliable and safe means for harvest preservation. One such approach may be induced resistance (IR) based on the activation of natural plant defense mechanisms. The most promising for practice are nonspecific systemic resistance (SR) and priming [6-8] when local treatment with a certain inducer promotes the entire plant resistance to several pathogens and accelerated defense response to a pathogen invasion [9, 10].

Studies of the molecular mechanisms of interaction between plants and microorganisms have led to the identification of a number of metabolites, currently known as biogenic elicitors, which, by activating plant signaling systems [11], trigger certain protective responses leading to the formation of SR [12].

Noel T. Keen in 1975 was the first to propse the definishion “elicitor” to refer to molecules capable of inducing phytoalexins. But now biogenic and non-biogenic compounds that stimulate any type of plant defense response involved in the formation systemic acquired resistance (SAR) or induced systemic resistance (ISR) to pathogens and pests [13] are commonly called elicitors. Biogenic elicitors do not have a general chemical structure and belong to a wide range of different classes of compounds, including oligosaccharides, peptides, proteins and lipids.

Interacting with a variety of naturally occurring microorganisms, plants recognize conservative patterns of molecular structures and metabolites common to entire microbial taxa. These are MAMPs (microbe associated molecular patterns) necessary for non-pathogenic, including beneficial, microorganisms, to exist, and PAMPs (pathogen associated molecular patterns). Additionaly, DAMPs, the damage-associated molecular patterns (or danger-associated molecular patterns) of some own plant metabolites are produced during pathogen invasion to signal the danger of damage [7].

In essence, PAMPs, which combine metabolite molecules from phytopathogenic fungi, bacteria, and oomycetes, are a subgroup of MAMPs [14]. MAMPs, PAMPs, and compounds that, like these biogenic patterns, cause resistance are exogenous elicitors, while DAMPs are endogenous. Upon contact of such microbial or endogenous patterns with pattern recognition receptors (PRRs) [14-17], plants generate a cascade of signals [14, 18-22] that activate various mechanism of plant innate immunity (base resistance) or patogen-triggered immunity (PTI) [23-25].

Protecting themselves from pathogens that have overcome PTI, plants, using intracellular R-receptors, recognize their effectors, the proteins produced by plant pathogens to disrupt the PAMPs and DAMPs reception or suppress the defense reactions that PAMPs and DAMPs induce, thereby ensuring the host

plant colonization [7].

Effectors recognized by R proteins initiate the second stage of active phytoimmunity, the effector triggered immunity (ETI) [26]. In general, PTI and ETI are shaped by similar plant defense responses [27], including the hypersensitivity response (HSR) [28, 29], although in the case of ETI the HSR develops more frequently and more intensely, leading to rapid localization of pathogens [30, 31].

In microorganisms non-pathogenic for plants, phytopathogenic fungi, oomycetes, bacteria and viruses that infect agricultural crops, a number of elicitor proteins have been found that belong to the MAMP/PAMP type [7, 31-33], as well as effectors, including race-specific, which, after recognition by the plant, can act as specific elicitors [34]. To denote elicitors of the first type, the term general elicitors is also used, which in the domestic scientific literature corresponds to the term nonspecific elicitors [35].

The study of the properties and mechanisms of action of these elicitors shape a new research cluster to proceed one of the most environmentally friendly areas in plant protection.

The most promising are biogenic elicitors that cause SR and are biodegradable in nature without the formation of toxic products. Another promising agents are those for which there are accessible sources and relatively cheap production technologies can be developed. Many protein elicitors found in microorganisms have these properties. For protein elicitors, in addition to being used through seed or foliar treatments, constitutive or inducible expression of their transgenes in plants is possible. Finally, the absence of a direct biocidal effect of SR protein elicitors on plant pathogens minimizes the likelihood of developing resistance to these protective agents.

Over the past few decades, significant progress has been made in identifying microbial proteins that are involved in interaction with crop plants as inducers of protective responses or as virulence factors. Now, there is a better understanding how this interaction occurs at the molecular level, which signaling systems are triggered by MAMP-type protein elicitors and how recognition of pathogen effector proteins involves ETI.

The data obtained indicate the promise of using resistance-inducing proteins as new biocontrol agents. In this regard, publications on induced plant immunity to diseases and abiotic stress are increasingly appearing in Russian scientific journals [36-38]. Non-protein compounds, mainly chitosan, synthetic analogues and derivatives of plant signaling molecules salicylate (SA) and jasmonate (JA) and substances reproducing their effect, are considered as inducers to be the basis of created protective drugs. A number of reviews have examined in detail the protective plant proteins and their role in immunity [39], and some publications indicate the protective role of antimicrobial peptides of ribosomal and non-ribosomal synthesis from biocontrol fungi and bacteria [40]. However, microbial proteins that induce disease resistance are not fully described.

This review analyzes the most important achievements in identification and study of the properties of microbial elicitor proteins. Here, we consider both conservative and specific elicitors. The former are bacterial, fungal and viral proteins and peptides recognized by plants as MAMP/PAMP which provide plants nonspecific resistance (PTI) and general defense reactions; the latter are effector proteins of fungi and bacteria [41, 42]. The structural features of elicitors and the mechanisms of their action, that is, the induced protective responses, if known, are briefly described. We also tried to show the diversity of microorganism species that are capable of producing elicitor proteins. A separate section is devoted to protein-based elicitor drugs, including those developed in Russia and China which have already proven their effectiveness in fields and have found practical application.

Endogenous elicitor proteins of the DAMP type, which play an important role in IR, and mediator proteins involved in signal recognition and transduction, are the own active molecules of plants, but not microorganisms, and therefore will remain outside the scope of the review. In the section on glycoproteins and peptidoglycans, only those are mentioned the elicitor activity of which is associated with protein fragment.

Elisitor proteins and peptides of bacteria. *Flagellin.* Flagellin of pathogenic and non-pathogenic bacteria is the most studied MAMP/PAMP protein elicitor [43]. It is a globular acidic protein that forms the outer helical filament of the bacterial flagellum. Flagellin is characterized by highly conserved N- and C-terminal sequences, a very low content of tyrosine and phenylalanine, the absence of tryptophan and cysteine, and therefore double bonds, and a very high content of alanine, glutamic and aspartic amino acids [44]. This evolutionarily conserved protein, vital for mobile bacteria, can be recognized by plants. Plant flagellin-sensitive receptors (FLS) belong to the leucine-rich repeat receptor kinase XII protein family (LRR-RLK XII) [45-47].

In most cases, plants recognize the flg22 epitope as an elistor capable of inducing both local and systemic immune responses. The flg22 is a fragment of N-terminal sequence consisting of 22 amino acids, the main motif of which contains the 15 most conserved amino acid residues. Most plants have a receptor FLS2 (flagellin-sensitive kinase) that recognizes this particular region of the protein [45, 48]). Interestingly, in the flagellin molecules that form the protofilaments of the bacterial flagella, the flg22 oligopeptide is hidden inside the protein globule and is not accessible to FLS2. However, in flagellin monomers, during flagellum self-assembly or after their disintegration upon death, it becomes available to this receptor [34] and acts as a potent elicitor at subnanomolecular concentrations [33].

Other plants, in particular rice (*Oryza sativa* L.), are able to recognize a fragment of the C-terminal region of flagellin (CD2-1) consisting of approximately 35 amino acid residues [49]. Tomato (*Solanum licopersicy* L.) and potato (*S. tuberosum* L.) plants recognize in the N-terminal region of flagellin the flgII-28 fragment differing from flg22 and having its own special receptor FLS3 [50, 51].

Upon recognition of flg22, the FLS2 receptor interacts with the serine-threonine kinase BAK1 (brassinosteroid receptor), which activates the intracellular kinase domains of these receptors [50], initiates elicitor signal transmission, and induces systemic plant defense responses, leading to the formation of PTI [44, 51] to all bacteria with flagella.

However, in the course of evolution, some bacteria, such as *Pseudomonas syringae* pv. *tomato*, *Xanthomonas campestris* pv. *campestris* or *Ralstonia solanacearum* have acquired modified flg22 sequences. This allows them to evade full recognition by the FLS2 receptor when bacteria with this modification attack Arabidopsis (*Arabidopsis thaliana* L.) Heynh or nightshade crops [33, 52]. Studies of the molecular mechanisms of resistance of the *Solanaceae* family plants to pathogens with modified flg22 led to the identification of the above-mentioned FLS3 receptor which recognizes the flgII-28 sequence [53]. In particular, in potato, under the influence of flgII-28, there is an active release of Ca^{2+} ions into the cytosol, a shift to the alkaline extracellular pH, generation of reactive oxygen species (ROS), phosphorylation of mitogen-activated protein kinase and activation of plant defense genes [54]. Interestingly, all protective reactions in the case of flgII-28 are much more pronounced than in the case of flg22. It should be noted that the ability of bacteria to modify flagellin elicitor peptides appears to be useful in symbiotic interaction. For example, altered flg22, which legumes do not respond to, is found in the nitrogen-fixing nodule bacterium *Rhizobium meliloti*. This helps

avoid triggering plant defense responses and provids an interaction beneficial to both the bacterium and the host plant [33].

Thus, different plant species recognize different regions of flagellin, and its peptides flg22, CD2-1 and flgII-28 act as active MAMP/PAMP elicitors of PTI. Flagellin is also recognized by the Toll-like receptor TLR5 of macrophages and dendritic cells and activates innate immunity in higher animals. However, TLR5 does not interact with flg22, CD2-1, or flgII-28, but with another flagellin domain formed by the N- and C-terminal regions of the peptide chain [42, 55].

Elongation factor Tu. Another conserved bacterial protein identified as a MAMP/PAMP-type elicitor is elongation factor Tu (EF-Tu). Its discovery was led to by studying the ability of extracts from *R. solanacearum*, *Sinorhizobium meliloti* with a modified flg22, which is not available for FLS2 recognition, and from a mutant strain of *Escherichia coli* with a defective flagellin gene to cause a reversible change in ion exchange in a suspension culture of Arabidopsis cells. It turned out that the studied extracts, despite the absence of flg22 or its receptor, induce this protective response typical of flagellin [56]. As a result, EF-Tu was isolated from an *E. coli* extract, the elicitor activity of which is responsible for the elf18 peptide, consisting of 18 amino acid residues and localized in the N-terminal sequence of the protein. The shorter N-terminal fragment of EF-Tu, the elf12 has no inducing activity and acts as an elf18 antagonist.

The elf18 elicitor fragment is recognized by the EFR receptor. Like the flagellin recognition receptors FLS, EFR belongs to subgroup XII of the LRR-RLK family [47]. Genes encoding EFR-like proteins with high sequence similarity in kinase domains are found in many plants, including *Brassicaceae* species and rice, with different ectodomains determining elf18 recognition [33].

Subsequently, the contribution of individual amino acids to the elicitor activity of elf18 was evaluated by the alanine scanning method. Peptides with amino acid residues at positions 1, 3, 6, 9, 10, 11, 12, or 13 replaced with alanine had the same activity as elf18. In contrast, substitutions at positions 2, 4, 5, and 7 resulted in a 10- to 400-fold decrease in peptide activity. When residues at positions 2 and 5 were simultaneously replaced, activity reduced 50,000-fold [56].

N-terminal sequences with high homology to elf18 were found in EF-Tu of a number of phytopathogenic bacteria, for example, *Erwinia amylovora*, *E. chrysanthemi*, *P. syringae*, *Xylella fastidiosa*, *S. meliloti* and *Agrobacterium tumefaciens*. Peptides of the latter two species, consisting of 18 amino acid residues, were as active as elf18, and similar N-terminal peptides from *P. syringae* (a pathogen of tomato) and *X. fastidiosa* (a pathogen of grapes, citrus, olives and other crops) even at concentrations 4 and 7.5 times higher than the concentration of elf18 from *E. coli* were less active [56, 57].

A detailed study of the structure of elf18 and its functional relationship with elicitor activity made it possible to use this peptide for in-depth investigtions of the PTI development mechanisms. In particular, several years ago, through translatome profiling of *Arabidopsis* leaves infiltrated with elf18, the molecular mechanism of global translational reprogramming that occurs in plants during PTI formation was elucidated [56].

Harpins. The protective properties of bacterial harpin proteins were first discovered in the early 1990s by Z.-M. Wei and a group of researchers from the laboratory of Steven V. Beer at Cornell University, USA. They found that one of the proteins of the phytopathogenic bacterium *Erwinia amylovora*, which causes fruit blight of the *Rosacea* family (harpin), can induce a hypersensitivity reaction (HSR) and rapid death of the leaf blade in tobacco (*Nicotiana tabacum*), which is not a host plant for this bacterium, and also cause reversible increase in pH in a suspension of its cultured cells [58].

Almost at the same time (in 1993), a group of researchers headed by S.Y. He [59] showed that an extracellular protein from *P. syringae* pv. *syringae* cause a HSR response in plants that are not hosts for this bacterium. The harpinPss protein (a product of the *hrp* gene) caused necrosis on the leaves of tobacco and other plants [59]. It was later determined that the harpin Hpa1 from *X. axonopodis* pv. *glycines*, the causative agent of soybean pustular spot, induces microwave cell death in tobacco [60].

All currently known harpins belong to thermostable cysteine-deficient acidic proteins with a relatively high content of glycine and serine residues [61, 62]. Harpins are introduced into plants by the type three secretion system (T3SS) of gram-negative phytopathogenic bacteria during their interaction with the host plant and can play the role of both effectors (T3 effector, T3E), necessary for pathogenicity that disrupt the integrity of the host membrane, and elicitor microbial patterns that induce protection against pathogens. In the latter case, treating plants with harpins increases resistance to diseases and stimulates crop growth, leading to an increase in yield and improvement in its quality. Moreover, the harpin HrpN induces drought tolerance in *Arabidopsis* by activating the jasmonate-dependent signaling pathway [63].

The discovery of the harpin *hrp/hrc* gene cluster in the mid-1980s and the subsequent demonstration that these genes encode a family of TTSS proteins in Gram-negative bacteria pathogenic to plants and animals, as well as plant symbionts, was a major milestone in molecular phytopathology [64]. In particular, it was found that in *E. amylovora*, the genes of the *hrp* cluster responsible for the production of harpins also control the pathogenicity of the bacterium and its ability to cause microwave death of plant cells [65]. In the *X. oryzae* genome, there were discovered both *hrp* cluster containing the conserved *hrp* and *hrc* genes which products can be recognized by plants and induce resistance, and the pathogenesis-associated genes *hpa* [65, 66]. It was then established that activation of the expression of genes encoding harpins and the subsequent synthesis of these proteins occurs during the interaction of the pathogen with the host plant [67, 68].

To date, *hrp* genes have been found in a number of phytopathogenic bacteria, including those of the genera *Erwinia*, *Pantoea*, *Pseudomonas*, *Xanthomonas*, and *Ralstonia* [63], and several harpins have been identified, which, based on their structure, are divided into four main groups HrpN, HrpZ1, HrpW1 and Hpa1 [61, 66, 69-72]. Some authors consider the HpaG harpins as a separate group [61, 69]. For each of these groups, several proteins with slight structural differences are known to be produced by pathogens that attack various plants, e.g., citrus fruits [73], soybeans [74], rice [75, 76], peppers [77], and cotton [70]. For example, many *Xanthomonas* species secrete harpin effectors of the Hpa1 group. In particular, in the rice blight pathogen *X. oryzae* pv. *oryzae*, the harpin Hpa1Xoo was found [78]. Harpin Hpa1Xag is isolated from *X. axonopodis* pv. *glycines* [73]. Strain *P. syringae* pv. *tomato* DC3000 produces two types of harpins, HrpZ1 and HrpW1 [79]. The harpins HrpNEa, HrpNEch, HrpZPss and HrpZPsph are synthesized by *E. amylovora*, *E. chrysanthemi*, *P. syringae* pv., *syringae* and pv. *phaseolicola*, respectively [80-82]. Orthologues of the HrpW group were found in *E. amylovora* (HrpW), *P. syringae* pv. *syringae* (HrpW1, HopAK1) and *Ralstonia solanacearum* (PopW, PopA1), and HpaXm in *X. citri* subsp. *malvacearum* [64]. Howeber, harpin-like proteins have not yet been found in bacteria pathogenic for cassava.

It has been established that harpins are not only capable of triggering a local hypersensitivity response but also act as SAR elicitors [82]. Thus, the harpin HrpZ1, which evokes a hypersensitivity response in various plant species, can activate several of their signaling systems and trigger both early and later defense responses [83]. It turned out that HrpZ1 harpins have high affinity for plant cell

membranes and specifically bind to them. This suggested that the development of protective reactions in response to HrpZ1 occurs after its binding to the receptor. Indeed, membranes of microsomes obtained from parsley cell culture contain a protease-resistant thermostable binding site for the C-terminal fragment of HrpZ1 [84]. Interaction with plant receptors has also been shown for other harpins, in particular, for the harpin HrpN from *E. amylovora* which in vitro binds to the small (6.5 kDa) HIPM protein associated with the plasma membrane of a recombinant yeast clone expressing this apple protein. Orthologs of HIPM were later discovered in rice (OsHIPM) and Arabidopsis (AtHIPM) [85].

As for the protective reactions that harpins induce, along with microwaves they often cause the ROS formation. Thus, harpins from *P. syringae* pv. *glycinea* and *X. oryzae* pv. *oryzae* are capable of causing the generation of ROS in tobacco cells, and one of the harpins from *E. amylovora* stimulates the formation of ROS not only in tobacco cells, but also in *A. thaliana*. Treatment of a suspension culture of grape cells with the commercial harpin-containing drug Messenger (EDEN Bioscience Corp., USA) causes an intense oxidative burst and activation of the stilbene synthase gene responsible for the synthesis of phytoalexin resveratrol [86]. S. Sang et al. [86] showed that harpins activate defense reactions through the transmission of a signal generated in the apoplast with the participation of the plant NADPH oxidase system. In transgenic *Arabidopsis* plants expressing the gene for harpin Hpa1Xoo, hydrogen peroxide is formed and accumulated in the cytoplasm and apoplasts. Suppression of apoplastic H_2O_2 production reduces both the accumulation of ROS in the cytoplasm and the resistance of plants to bacterial pathogens. These data suggest that apoplastic H_2O_2 undergoes cytoplasmic translocation to participate in plant defense against pathogens. Both harpin and H_2O_2 induce the expression of plant genes encoding enzymes involved in SR, such as phenylalanine ammonia lyase (PAL), glutathione S-transferases (GST) and anthranilate synthase 1 (ASA1). In *Arabidopsis*, H_2O_2 enhances the expression of PAL1 and GST, but not ASA1. It is assumed that harpins activate two different signaling pathways, one to increase the formation of ROS and the expression of PAL1 and GST mRNA, the other to increase the expression of GST and ASA1 independent of H_2O_2 [87].

Translation of genes encoding bacterial harpins into plants leads to increased resistance to various pathogens. Thus, in transgenic cotton plants expressing the *hpaXoo* gene (one of the genes encoding harpins), when infected with the pathogenic fungus *Verticillium dahliae*, increased ROS production was noted. Recombinant HpaG protein from *X. oryzae* pv. *oryzicola* effectively induced resistance in rice [88], and in transgenic tobacco and beet plants expressing the HrpZ protein gene from *P. syringae* pv. *phaseolicola*, resistance to rhizomania increased [89].

As mentioned above, the structure of harpins isolated from different bacterial species varies, but the common ability of these proteins to induce resistance to various pathogens in plants that do not serve as their hosts remains. In this regard, studies have been carried out on the functions of individual fragments in harpin molecules [69, 71, 89]. It was established that certain fragments of Hpa1-like harpins of *X. citri* subsp. *malvacearum* (fragment including amino acid residues 35-51) and *X. oryzae* pv. *oryzae* (fragment of residues 36-52) can induce a microwave response [82]. In addition, it was shown that this reaction is caused by the infiltration into tobacco leaves of a 24 amino acid fragment present in the C-terminal region of the harpin HrpZ isolated from *P. syringae* [71]. It was also found that the C-terminal fragment of the HrpZ1 protein from *P. syringae* pv. *phaseolicola* has the properties of a PAMP-type elicitor and is capable of inducing early plant defense responses, in particular, activating two protein kinases (MPK3

and MPK6) from the MAP kinase signaling cascade. Insertional mutagenesis of the gene responsible for the synthesis of HrpZ1 confirmed the importance of the the C-end sequences for the elicitor activity of the entire protein [86]. In addition, when studying the functions of individual domains of harpin proteins, it was shown that the fragment HpaG10-42 of the harpin HpaGXooc from *X. oryzae* that contains amino acid residues from 10 to 42, serves as a more active inducer than the full-length HpaGXooc protein. Under field conditions, the fragment HpaG10-42 increases resistance of rice plants to bacterial rot (*X. oryzae* pv. *oryzae*), blast (*Magnaporthe oryzae*) and rhizoctonia (*Rhizoctonia solani*) [90]. The same HpaG10-42 fragment can trigger defense reactions in wheat plants expressing it [91].

In general, studies of the elicitor properties of harpins indicate that these bacterial proteins have all the important features of PAMPs: they are widely distributed in various bacterial species, bind to PRR receptors, and trigger primary plant defense responses. The key determinants responsible for elicitor activity, at least in some harpins, may be their fragments. In addition, the interaction of harpins with plant receptors can lead not only to the development of SR due to the activation of genes involved in the implementation of protective reactions in plants, but also to the stimulation of their growth, improvement of the physiological state and increase in yield [72, 73].

Comparison of the elicitor effects of harpins and flagellin. Flagellin and harpins are evolutionarily distant bacterial MAMPs/PAMPs. A comparison of the responses of plant cells to the effects of flg22 and harpins shows a number of differences in mechanisms of their elicitor action. Thus, when treating cell cultures of two grape varieties, it was found that the PTI-activating elicitor flg22, in contrast to harpins, causes HSR cell death much less frequently. In addition, treatment with harpin leads to a more rapid and intense development of the oxidative burst compared to flg22 [84]. Recently L.B. Sands et al. [91] showed that harpin was able to effectively induce hemp resistance to *P. aphanidermatum* and stimulate the growth of infected plants. Treatment with flg22 did not cause protective reactions and did not stimulate seedling growth. In this regard, the authors suggested that the protective response induced by flg22 are not active enough to effectively suppress *P. aphanidermatum* infection in seedlings of plants of this species.

X. Chang et al. [92] found that flg22 induces the accumulation of jasmoate in rock grape plants (*Vitis rupestris* Scheele), while harpins do not cause such an effect, although activation of JA-dependent signaling, as noted above, can occur under their influence, increasing resistance to abiotic stress [64]. Moreover, signal transduction associated with JA-dependent signal transmission involving ethylene (ET) was observed after treatment of hemp seedlings with both harpin and flg22. Both of these bacterial elicitors are capable of stimulating the expression of similar defense genes and inducing similar plant defense responses [93, 94]. In addition, harpins, like flg22, can activate the salicylate-dependent signaling pathway that leads to SAR [95]. However, in contrast to flg22, which causes typical PTI, the defense responses that are characteristic of ETI are more often developed in response to harpins [96], but sometimes the type of defense response depends on the way the plants are treated. For example, charipne from *E. amylovora*, when infiltrated into intercellular spaces, causes a microwave reaction, and when sprayed on plants, it induces SAR [58]. It is possible that the combined use of these elicitors can lead to synergism due to the simultaneous activation of PTI and ETI, and significantly enhance the plant defense response [96].

Cold shock proteins. Cold shock proteins (CSP) are widespread in pro- and eukaryotes. All of them contain conserved CSD domains (cold shock domain), have the ability to bind to nucleic acids and activate or suppress the expression of a large number of genes involved in cell division, differentiation and encoding a

variety of protective proteins [97-99].

Bacterial CSPs comprise a family of proteins with a molecular mass of about 7 kDa, consisting of a single amino acid sequence of about 70 amino acid residues in length [98]. The synthesis of CSP is activated at low temperatures and represents one of the mechanisms of adaptation of organisms to unfavorable environmental conditions. CSPs, originally discovered in cold-stressed bacteria, were later shown to be synthesized constitutively as well as in response to other stresses [99]. As in bacteria, in plants CSPs promote adaptation to cold, and alien CSPs are recognized as components of MAMPs/PAMPs and induce resistance to pathogens. The elicitor activity of bacterial CSPs of the CspB family was first demonstrated by classical experiments in which a reversible change in ion exchange on the plasmalemma of the plant cell was recorded by a reversible increase in the extracellular pH of a plant cell suspension culture as one of the earliest protective responses [99, 100].

It was discovered that one of the surface domains of CSP, including an RNA-binding domain known as RNP-1 (or NPCS), is responsible for the ability to induce a rapid response in cultured tomato, tobacco and potato cells. Further research showed that many other species of the family *Solanaceae* recognize a highly conserved RNP-1 domain motif of a peptide containing 22 aromatic and basic amino acid residues called csp22as as MAMP/PAMP. Its receptors are the LRR-RL kinase CORE discovered in the cultivated tomato species *Solanum lycopersicum* L. and the receptor-like protein NbCSPR from *Nicotiana benthamiana* Domin [101]. Homologs of NbCSPR are found only in some *Solanaceae* species and are absent in *S. lycopersicum*, while LRR-RLKs homologous to CORE are found in a number of *Solanaceae* species, including *Solanum* and *Nicotiana* species. Transgenic Arabidopsis plants expressing CORE become sensitive to csp22, which makes them more resistant to the pathogenic strain *P. syringae* pv. *tomato*. The minimum amino acid sequence of csp22 which activates defense responses in plant cells has 15 amino acid residues. This highly conserved csp15 epitope induces plant defense responses. In particular, at a concentration of about 0.1 nM it induces an oxidative burst in tobacco and potato cells, but is not recognized as an elicitor by rice and cucumber (Cucumis sativus L.) cells [100].

CspD is another cold shock protein isolated from *Bacillus thuringiensis* and is also known as microbial factor 2 (MF2). The *B. thuringiensis* gene encoding this 7.2 kDa thermostable low molecular weight protein was cloned and sequenced (AY272058, GenBank, https://www.ncbi.nlm.nih.gov/genbank/). It turned out that its nucleotide sequence and the amino acid sequence of CspD have high homology with other bacterial CSPs. Like csp22 or csp15, CspD causes a reversible change in ion exchange in cultured tobacco cells, but, unlike these elicitors, it is able to activate a protective response in tomato and rice cells and induce nonspecific systemic resistance in various monocots (rice, wheat) and dicotyledons (tomato, tobacco, potatoes) plants to pathogenic fungi, oomycetes and viruses [101]. Thus, after applying CspD to wheat leaves or spraying potato and rice seedlings with this elicitor, an increase in resistance to the pathogens of septoria (*Parastagonospora nodorum*), late blight (*Phytophthora infestans*) and blast (*M. oryzae*) was observed [102].

Exposure of tobacco plants to CspD reduced its infection by tobacco mosaic virus (TMV) and potato virus X. The csp15 peptide of the CspD protein (VKWFNAEKGFGFITP) also had elicitor activity. Its addition to a suspension of cultured tobacco cells activated the H^+ pump and caused a reversible change in extracellular pH. This peptide, like CspD itself, exhibited inducing activity in model plant—pathogen systems. Treatment of tobacco leaf halves with 1 to 10 mM csp15 1 day before TMV inoculation led to a sharp decrease in the number of

necrosis on the treated halves compared to control halves or untreated leaves of the same plant [102]. Application of csp15 to the surface of discs cut from potato tubers enhanced the HSR of cells to *P. infestans* and caused the accumulation of salicylic acid in the tissues of tubers treated with this peptide [103]. Transgenic tobacco lines of the necrotic cultivar Xanthi NN and the TMV-susceptible cultivar Samsun nn exhibited increased resistance to *Alternaria longipes* and TMV, and CspD expression positively correlated with plant resistance to both pathogens [103].

Eliminator proteins and peptides of filamentous fungi. Like bacteria, micromycete fungi synthesize a number of proteins and peptides with the functions of nonspecific or specific elicitors.

Resistance-inducing effectors of Cladosporium fulvum. Specific protein elicitors of the phytopathogenic fungus *C. fulvum* cysteine-containing peptides Avr2, Avr4, Avr4E, Avr9 Ecp1, Ecp2, Ecp4 and Ecp5 were discovered and studied in detail by a group of researchers from Wageningen University, the Netherlands, led by P.J. de Wit [104, 105]. These effectors function as virulence factors. By binding chitin, they prevent its detection by pattern recognition receptors when tomato is infected by compatible races of the fungus and prevent the degradation of chitin by plant chitinases [106]. However, tomato (the host plant for *C. fulvum*) has an efficient system for recognizing these effectors, after which plant defense responses, including HSR, are induced [107].

The Avr4 elicitor consists of 101 amino acids, including 6 cysteine residues, and Avr9 is a 28 amino acid peptide. The sequences of Avr4 and Avr9 have low homology. The mature Avr2 peptide contains 58 amino acids 8 of which are cysteine. After processing of larger precursor proteins by fungal and/or plant proteases, mature peptides are found in the tomato apoplast and induce HSR in plants with complementary resistance genes *cf4*, *cf9*, or *cf2*, followed by other defense responses [31, 106]. Below, as examples, three (Avr4, Avr9, Ecp6) of the currently identified Avr and Ecp elicitors will be discussed in more detail.

Like most fungal effectors, Avr4 and Avr9 enter plants through the secretory pathway of the endoplasmic reticulum of *C. fulvum* and are recognized by R gene products as race-specific elicitors. Avr4 recognizes and binds the receptor-like protein Cf4 without kinase activity. In addition, the interaction of Cf4 with the receptor-like kinase OBIR1 may be necessary for the HSR [108, 109]. The HSR induction in response to Avr9 can apparently occur as a result of its recognition in one of two ways, i.e., after the formation of Avr9 complex with a high-affinity binding site and subsequent interaction with the membrane-anchored extracellular receptor glycoprotein protein Cf-9, or after a direct low-affinity binding of Avr9 to Cf-9 [99, 100]. Infiltration of Avr4 and Avr9 into tomato tissue leads to the specific activation of an entire arsenal of plant defense responses, including electrolyte leakage, stimulation of plasma membrane H-ATPase, ROS generation with oxidative burst, increased lipoxygenase activity and induction of PR proteins β-glucanase and chitinase [31].

Ecp6 is another chitin-binding effector protein that *C. fulvum* secretes into the apoplast when colonizing tomato plants. It contains three lysine motifs LysM1, LysM2 and LysM3. LysM2 and LysM3 are proposed to form domains required for interaction with chitin, and LysM1 may be recognized by the plant because it is able to trigger a response in tomato cell suspension culture [110]. It is assumed that in tomato plants that develop HSR when Ecp6 is infiltrated a hypothetical receptor called Cf-Ecp6 is localized on the cell surface [111]. Regarding the distribution of specific elicitors similar to those described here for *C. fulvum*, homologs of Avr4 are found in fungi from the class *Dothideomycetes*, and Ecp6-like genes are widespread in the fungal kingdom [112, 113].

C. fulvum is also known to produce the protein CfHNNI1 homologous to the translation factor bZIP. This non-specific elicitor induces the expression of *lehsr203*, a marker gene for HSR and causes resistance in non-host plants belonging to three different families. Transgenic tobacco plants with the gene encoding CfHNNI1 acqured resistant to *P. parasitica* var. *nicotianae*, *P. syringae* pv. *tabaci* and TMV. The ability of CfHNNI1 to induce HSR is due to amino acid residues conserved for bZIP factors. It has also been shown that its mechanism of action is not associated with activation of the JA/ET-dependent signaling pathway and that the elicitor activity of CfHNNI1 is reduced under the influence of inducers of SA-dependent signaling [31].

Eliminators of phytopathogenic and non-pathogenic Fusarium fungi. Like *C. fulvum*, the pathogen causative agent of tomato Fusarium wilt *Fusarium oxysporum* f. sp. *lycopersici* (FOL) produces cysteine-rich effector proteins that are recognized by plants with complementary resistance genes. One such effector, called the SIX1 protein, was the first avirulence factor discovered in FOL and other root-invading plant pathogens [113]. After maturation of the 32 kDa precursor, a 12 kDa polypeptide from the central part of SIX1, containing 6 cysteine residues, is secreted into the xylem sap of infected plants and induces protective response in tomato lines with a gene for resistance to this effector. In *F. oxysporum*, at least 11 small cysteine-rich proteins of the SIX1 type have been identified and it has been shown that at least three of them (SIX1, SIX3/SIX4) play the role of avirulence factors, which in tomato varieties and lines with gene *I* (*I-1*, *I-2*, *I-3*) induce resistance to FOL forms specific for different host plants. A homolog of SIX1 was also found in *F. graminearum* [113].

From the mycelium of the non-pathogenic tomato strain *F. oxysporum* CS-20 which protects plants from FOL strains that cause wilt, the elicitor CS20EP was isolated. CS20EP is the main polypeptide of the protein fraction which reduces the damage to tomato by this pathogen by weakening symptoms and slowing down the development of the disease. CS20EP is a small cysteine-rich basic protein (pI 9.87) with a molecular weight of approximately 10 kDa, containing 23% hydrophobic amino acid residues. Its cysteine motif is composed of 11 cysteine residues of which 6 are located in the N-termus. The fraction containing CS20EP induces a rapid reversible change in extracellular pH in tomato cell culture, increased chitinase activity in the root system of its seedlings, and provides a systemic increase in the expression of genes encoding the mediator protein PR-1, a SAR marker, in leaves. It is suggested that this elicitor may be secreted by strain CS-20 and contribute to its biocontrol effect [114].

In another biocontrol agent, the non-pathogenic wheat isolate *F. sambucinum* FS-94, protein elicitors were also discovered and partially identified which, unlike CS20EP, induce systemic and local resistance of plants not only to FOL [115], but also to pathogens of root rot of wheat (*Fusarium* spp., *Bipolaris sorokiniana*) [116] and gray rot of cruciferous crops (*Phoma lingam*). These nonspecific elicitors combine a fraction of proteins ranging from 40 kDa to 67 kDa in size the main polypeptide component of which has a molecular mass of ~ 50 kDa, and activate both early and later defense responses of dicotyledonous and monocotyledonous plants. In particular, in a suspension culture of wheat cells of *Triticum aestivum* L. and *T. kiharae* Dorof. et Migush. these proteins cause a reversible change in ion exchange and increase cell tolerance to lysis caused by *F. culmorum*. In seedlings from treated *T. kiharae* seeds, they upregulate the expression of genes for defensin-like antimicrobial peptides (AMPs) highly homologous to the Tk-AMP-D and Ec-AMP-D1 defensins found in wheat and wild cereals, respectively. These AMPs in micromolar concentrations are active against several phytopathogenic fungi, including *Fusarium* spp. and *H. sativum* (syn. *B. sorokiniana*).

From culture filtrates of *F. oxysporum* f. sp. *erythroxyli* (FOE) which causes vascular wilt of coca plants (*Erythroxylum coca* Lam.), the Nep1 protein (24 kDa) with a conserved GHRHDWE domain was isolated. It induces necrosis and ET production in many dicotyledonous plant species. In tobacco leaves, Nep1 causes the accumulation of 1-aminocyclopropane-1-carboxylic acid (ACC) synthase and ACC oxidase involved in the synthesis of ET, and in plant cell culture it causes a reversible change in ion exchange and the ROS generation [32]. Genetically unrelated but having similar activity proteins of the same size were found in *F. accuminatum* and *F. avenaceum*, and a protein homologous to the elicitor PaNie213 from *Pythium aphanidermatum* described hereinbelow was identified in FOE. Although Nep1 from FOE activates many of the same processes as typical elicitors in model tests, it has pronounced phytotoxicity towards dicotyledons and does not induce plant disease resistance. Moreover, it may act as a contact herbicide that promotes plant pathogen rather than plant protection [117]. Currently, Nep1-like proteins (NLPs) have been found in many plant-associated microorganisms, and some of them produce NLPs that are not phytotoxic. Thus, the biotroph *Hyaloperonospora arabidopsidis*, the causative agent of Arabidopsis downy mildew secretes 10 noncytotoxic NLPs which do not cause necrosis, but trigger PTI mechanisms and induce resistance to *H. arabidopsidis*. One of these NLPs, HaNLP3, activates the expression of a large set of Arabidopsis defense genes, acting as a MAMP-type elicitor. Interestingly, a 24 kDa fragment from the central part of the NLP of some other fungi, as well as bacteria, is also capable of activating PTI [118].

Elicitors from other micromycetes. Various proteins and peptides with elicitor properties have been identified in other phytopathogenic filamentous fungi belonging to necrotrophs or hemibiotrophs, as well as in saprotrophic antagonists of these phytopathogens.

The race-specific elicitor NIPI, a product of the *AvrRrs1* gene, was isolated from the cultural filtrate of *Rhynchosporium secalis,* the causative agent of blight of rye and barley. This gene encodes an 82 amino acid protein, which, after cleavage of the signal peptide, is converted into a mature 60 amino acid protein containing 10 cysteine residues in five intramolecular disulfide bonds. In barley varieties with the *Rrs1* gene that are resistant to incompatible races of *R. secalis*, NIPI induces necrosis and accumulation of pathogenesis-related proteins PR-1, PR-5, PR-9 and PR-10. It is also capable of causing necrosis in barley plants, regardless of their genotype, by stimulating the H^+-ATPase activity in the plasma membrane of plant cells. Along with this, in virulent races of the pathogen it works as a toxic effector. Variations in the biological properties of NIPI depend on its primary structure. At least four isoforms of NIP1 are known which differ significantly in bioactivity [119].

Many antagonist strains in some species of the genus *Trichoderma* are widely used in agricultural practice as biocontrol agents. Hydrophobins, the low molecular weight (less than 20 kDa) secreted hydrophobic fungal proteins with a conserved motif of eight cysteine residues forming four disulfide bonds, have elicitor activity. Thus, the elicitor Sm1 was detected in the culture filtrates of *Trichoderma virens* Gv29-8 [120]. The authors report that this hydrophobin-like protein has a high percentage of hydrophobic residues (40%) and includes four cysteines and three tryptophans. Mature Sm1, with a calculated molecular mass of 12.55 kDa and a pI 5.78, consists of 120 amino acids and appears to undergo post-translational modification as it contains sulfation, phosphorylation, and N-glycosylation sites. Sm1 does not have phyto- and fungitoxicity. In rice plants and cotton cotyledons treated with Sm1, it triggers local and systemic responses, namely, ROS generation, superoxide dismutase expression, production of oxylipins,

phytoalexins, synthesis of PR proteins, an increase in their activity, and prevents plant colonization by the anthracnose pathogen. Elicitor activity was also detected in two other hydrophobins from the group of low molecular weight cysteine-containing fungal proteins, the EPL1 (12.6 kDa, pI 5.5-5.7) and EPLT4 (EPL1 homolog) produced by *T. atroviride* and *T. asperellum*, respectively [121, 122]. Another hydrophobin with elicitor activity, named HYTLO1, was isolated from *T. longibrachiatum* MK1 [123]. It is completely similar to the HYTRA1 protein from *T. harzianum* T22 and is a 7.2 kDa protein with eight cysteine residues located, like in other hydrophobins, in a conserved motif. Along with the ability to inhibit the growth of certain microbes and to stimulate plant growth, HYTLO1 serves as a strong stimulator of plant defense responses. Its infiltration into tomato leaves leads to the development of local and systemic resistance to the gray mold pathogen (*B. cinerea*) due to HSR, generation of an oxidative burst in plant cells and increased transcription or activity of PR proteins [124].

The SCFE1 and PebC1 proteins are nonspecific elistors produced by the phytoptogenic necrotrophic fungi Sclerotinia sclerotiorum and B. cinerea, respectively [125, 127]. SCFE1 is a secreted protein found in vitro in culture fluid, and PebC1 is isolated from mycelium. It has been shown that in Arabidopsis plants, SCFE1 is recognized by the receptor-like protein RLP30 and, interacting with the BAK1 kinase, induces defense responses characteristic of PTI [125]. Treatment of tomato seedlings with the PebC1 protein (36 kDa, pI 4.85) increased their resistance to the pathogen and led to an increase in the content of enzymes associated with its development (PAL and polyphenoloxidase) and ROS generation in plant tissues, and also stimulated the development of the root system of wheat seedlings and their drought tolerance [29].

Elicitor proteins PemG1 (36 kDa, Ip 4.7) and PevD1 (~ 16 kDa) were found in the phytopathogenic hemibiotrophs *M. oryzae* and *Verticillium dahliae*, respectively [126, 127]. PemG1 is thermostable and activates the rice control system against *M. oryzae*, as well as the resistance of rice and Arabidopsis to bacterial infections. In these plants, it induces overexpression of genes that control the SA-dependent pathway of elicitor signal transmission, transient expression of PR protein genes, accumulation of ROS and a rice SAR marker OsPR-1a, and increases the activity of cellulase and alcohol dehydrogenase. Calcium channel blockers prevent PemG1-induced accumulation of OsPR-1a. Arabidopsis mutants defective in JA/ET-dependent signaling, after treatment with PemG1 exhibit increased resistance to bacterial infection, while mutants with impaired signal transduction via the salicylate pathway, on the contrary, do not respond to exposure to the elicitor. This suggests that PemG1 functions as an activator of salicylate- and Ca^{2+}-dependent signaling and a SAR elicitor [127]. In order to obtain the predicted PevD1 protein and confirm its elicitor properties, the gene encoding PevD1 in *V. dahliae* was introduced into *E. coli*. Plants treated with recombinant PevD1 acquired SR to TMV. In response to treatment of cultured tobacco cells with the elicitor, a change in ion exchange occurred, which was recorded by the alkalinization of the suspension. In treated tobacco plants, the production of hydrogen peroxide increased, the deposition of callose occurred, and the synthesis of phenols and lignin was enhanced [128]. *V. dahliae* also secretes 65 kDa and 28 kDa glycoproteins which induce the synthesis of phytoalexin gossypol in cultured cotton cells. Using enzymatic proteolysis and periodate oxidation of carbohydrates, the protein component was shown to be responsible for the elicitor activity of these glycoproteins [7, 31].

Alternaria tenuissima is a source of several elicitor proteins, such as the SAR inducer of Hrip1 which induces HSR cell death in tobacco and the expression of genes associated with SR to TMV [129]. The same species produces the

elicitors PeaT1 and PeaT2. PeaT2 is highly similar to the multifunctional structural membrane proteins prohibitins. In addition to inducing resistance, it can stimulate seed germination and root growth in wheat [130]. PeaT1 is a thermostable acidic protein (Ip 4.22) isolated from fungal mycelium, which, like Hrip1, induces tobacco SR but does not cause HSR. By expression in *E. coli*, recombinant PeaT1 (~ 22.6 kDa) was produced. Bioinformatic analysis of its structure showed that PeaT1 is a conserved protein of plant pathogens that contains a UBA domain associated with ubiquitin. The UBA motif, consisting of approximately 45 amino acid residues, is present in various proteins involved in cell signaling through receptor-like protein kinases. PeaT1, lacking the UBA domain, is unable to induce SR. Data on the structure of the signal peptide of this elicitor suggest that it enters the endoplasmic reticulum and not the plant apoplast. There is also evidence for the existence of protein binding sites for PeaT1 on the plasma membrane of tobacco plant cells which may transmit signals into the cytoplasm of cells, causing SAR [131].

Recently discovered fungal MAMPs/PAMPs and ETI elistors. Recently, a gene encoding a novel effector CsSP1 (a small protein with a predicted molecular mass of 25.9 kDa and Ip 8.84) which is secreted by this pathogen during infection was discovered in the hemibiotroph *B. sorokiniana*. The amino acid sequence of CsSP1 is similar to sequences inducing necrosis and production of ET specific Nep elicitors in other fungi [32, 119]. This effector is required for the pathogen to infect, overcome PTI, and successfully develop in wheat plants. It acts as an elicitor that triggers ETI in the host plant. New elicitors also include the small protein PeSy1 (11 kDa) which was found in the actinomycete *Saccharothrix yanglingensis* Hhs.015, an endophyte that increases plant resistance to pathogens. It has been established that PeSy1 actively induces HSR, and the recombinant protein PeSy1 causes early protective responses (e.g., ROS generation, callose deposition) and transduction of elicitor signal. This leads to increased resistance to *S. sclerotiorum* and *P. capsici* in tobacco (*N. benthamiana*) and resistance to *P. syringae* pv. *tomato* in tomato. PeSy1 interacts with a specific, previously unknown receptor-like cytoplasmic kinase RSy1 (Response to PeSy1). These results and the ability of PeSy1 to enhance the expression of PTI marker genes suggest that it acts as a MAMP-type elicitor [132]. New effectors, the orthologues of Ecp (*C. fulvum*) were also found in the fungus *Dothistroma septosporum* that attacks pine trees. Some of the proteins induce HSR and contribute to the development of resistance in plants that are not hosts of the pathogen [133]. Finally, very recently, two new proteins, Fg02685 and Fg62 found in a causative agent of wheat head blight *Fusarium graminearum*, are required by the fungus for colonization and are secreted into the apoplast during plant infection by the pathogen [134, 135].

The small protein Fg02685 has been identified as a new fungal PAMP. In tobacco (*N. benthamiana*), it causes protective responses typical of PTI, namely HSR cell death, expression of plant genes associated with nonspecific resistance, activation of the MAP kinase signaling cascade, accumulation of ROS and callose. The receptor-like kinases BAK1 and SOBIR1 are not involved in the recognition of Fg02685, and the conserved α-helical motif of the N-terminal region is responsible for the ability to induce HSR. The elicitor activity of Fg02685 is due to the 32-amino acid N-terminal motif, the FgNP32 peptide, which increases plant resistance to *Fusarium* and *Phytophthora* species. Interestingly, homologues of Fg02685 are frequently found in rust fungi (*Puccinia* spp.) and other micromycetes, as well as among *Phytophthora* species and other oomycetes [136]. Fg62 causes resistance of *N. benthamiana* to *P. capsici*. A signal peptide is responsible for its HSR-inducing activity. This protein does not have conserved cysteine domains, but contains 7 cysteine residues. In *N. benthamiana* it induces the

expression of two PTI marker genes. Apparently, Fg62 is an orphan protein; sequences homologous to Fg62 have so far been found only in *F. culmorum* and *F. pseudograminearum* [136].

Elicitors of oomycetes. *Elisitins.* Elicitins are small (~ 10 kDa) hydrophilic proteins of *Phytophthora*, *Pythium* and some other oomycetes, highly conserved within the family *Pythiaceae*. They have been convincingly characterized as PAMPs. All elicitins can bind sterols. Therefore, it is generally accepted that oomycetes, which are not capable of synthesizing these metabolites, use elicitins as transporters of plant sterols into their cells, including for embedding sterols into zoospore membranes. However, these proteins are not oomycete effectors. Moreover, *Pythiaceae* species produce a number of effectors to neutralize the activity of elicitins. For this activity, the sterol-binding ability is not decisive, but, by increasing the fluidity of the plasmalemma of infected plant cells, it can probably contribute to both signal transduction and induced ROS generation [137-139]. Some authors propose to consider elicitins as avirulence factors associated with resistance at the species level which represent an intermediate link between PAMPs and specific elicitors [140].

Elicitins were discovered back in the late 1980s as elicitors of the HSR in tobacco infected with *P. cryptogea* or *P. capsici*, but continue to be intensively studied to this day. They have been studied in most detail in *P. cryptogea*, *P. capsici*, *P. parasitica*, *P. megasperma* and *P. infestans* and are known under the names cryptogein (CRY), capsicin (CAP), parasiticin (PAR), megaspermine and INF1, respectively. A total of 100 sequences of these proteins have now been analyzed [139]. Based on the primary structure, they are divided into several classes and into acidic (α) and alkaline (β) proteins, and according to their coding genes, into true elicitins (ELI) and elicitin-like proteins (ELL) that form phylogenetic clades different in their HSR-inducing activity among different species [138, 140]. Comprehensive studies of ELI and ELL proteins disclosed their structure and provided information about their reception and role in the interactions of oomycetes with plants [138, 140]. It has been established that all elicitins contain a signal peptide, a highly conserved domain consisting of 98 amino acids with 6 non-variing cysteine residuesthat form 3 disulfide bridges, and variable C-terminal sequences. Typically, the C-terminal domains of ELI and ELL proteins (with the exception of the ELI-1 clade) are enriched in threonine, serine, and proline [138]. A study of the three-dimensional structure of β-CRY and other elicitins from the ELI-1 clade showed that their conserved domains are formed by five α-helices, one β-antiparallel sheet, and one ω-loop [137]. ELIs are thought to be secreted into the extracellular space, while some ELL proteins are likely anchored in the plasma membrane (including the membrane of motile zoospores) and the cell wall of oomycetes [141].

Like many other MAMPs/PAMPs, elicitins are recognized by PRRs, including receptor-like kinases such as SERK3/BAK1 which are among the major PTI receptors for microbial elicitors [139] and can directly or indirectly interact with specific R elicitor receptors [140]. Elicitin receptors likely recognize their conserved domain. It was established that a specific region of the ω-loop contains a highly conserved leucine residue at position 41 which is important for the perception of elicitins by different plant species [141].

Plants that respond to elicitin exposure by developing a local HSR or SAR belong to different families. Thus, in addition to tobacco, the protective effect of elicitins was demonstrated in tomatoes, peppers, potatoes, turnips, radishes, peas, grapevines, citrus fruits and oak seedlings, and the response varied among different taxa and among different plants within the same taxon [139]. The lysine or valine residues at position 13 and lysine at position 39 plays a key role in inducing the

HSR and necrosis formation, and the SAR-inducing activity of elicitins results from a combination of several features of the primary structure, including overall surface charge and the presence of specific lysine residues [138].

Treatment of plants with elicitins before inoculation increases resistance to pathogenic oomycete species and to other pathogens, including causative agents of bacterial diseases [138-142].

Like many other resistance inducers, elicitins cause early plant defense responses, namely, a reversible shift in extracellular pH [139] and explosive ROS generation associated with activation of plant NADPH oxidase and MAP kinase cascade. However, except for *Nicotiana* spp. and some *Solanum* species, the oxidative burst induced by elicitins does not always lead to HSR cell death and necrosis, sometimes the HSR develops under the influence of a later burst of ROS that is characteristic of elicitins [138, 139] with phosphorylation of MAP kinases and a long-term increase in their activity [140]. Initially, SA-mediated development of SARs and accumulation of PR proteins were considered the main effects of elicitin exposure in plants [142]. However, it was later discovered that treatment of tomato with INF1 and β-CRY or ELL protein from non-pathogenic *Pythium oligandrum* induces plant resistance to bacterial wilt, powdery mildew and *P. parasitica* by activating the JA/ET-dependent elicitor signal transduction pathway characteristic of ISR and the expression of PR protein genes [138, 139, 142]. Thus, elicitins can be considered as effective elicitors of nonspecific SR, triggering both SA-dependent and JA/ET-dependent signaling.

Other elicitors of oomycetes. In addition to elicitins, elicitor proteins and peptides such as Pep-13, PB90, PaNie213, and CBEL are known in *Pythiaceae* species.

The Pep-13 peptide is a 13 amino acid motif responsible for the elicitor activity of the transglutaminase family protein GP42 (42 kDa) found in the cell wall of *P. sojae*, and CBEL is a late blight lectin that binds cellulose and is involved in adhesion to plant cells [7]. Pep-13 is highly conserved among *Phytophthora* species and is highly active. It induces typical PTI protective responses in potato and parsley cells at extremely low concentrations (~ 1 nM) [143]. Mutations that alter the composition of Pep-13 abolish both elicitor and transglucominase activities [143]. PaNie213 from *P. aphanidermatum* is capable of inducing HSR in Arabidopsis and other plants. The mature protein (25 kDa) consists of 213 amino acid residues, is synthesized as a 234 amino acid precursor with a signal peptide and has a proteinase cleavage site. It induces de novo synthesis of 4-hydroxybenzoic acid in cultured carrot cells, callus formation in Arabidopsis, and necrosis of tobacco and tomato leaves [144]. An elicitor protein similar to PaNie was isolated from *P. megasperma* f. sp. *glycinea*, *P. infestan*s and *P. parasitica* [145]. PB90 protein (90 kDa) was isolated from the culture filtrate of the cotton late blight pathogen *P. boehmeriae*. Its infiltration into the leaves of tobacco, which is not the host of this pathogen, causes HSR, induces the synthesis of ROS (H_2O_2), the accumulation of SA and an increase in the activity of protective enzymes peroxidase (POD) and PAL [32]. It is also reported that a glycoprotein (15 kDa) structurally similar to some elicitins was found in the cultural filtrate of *P. colocasiae*, the causative agent of late blight in *Colocasia esculenta*. A polypeptide fragment of this glycoprotein is responsible for its ability to induce an increase in the activity of POD, PAL and lipoxygenase [145].

Microbial enzymes with elicitor properties. When plants are infected, necrotrophic pathogens secrete various enzymes that break down the cellulosic and hemicellulosic components of the plant cell wall to make nutrients available. Some of the enzymes are recognized by plants and act as elicitors of defense responses, and the ability of these enzymes to induce resistance in the vast

majority of cases is not related to their enzymatic activity.

The best known examples of elicitor enzymes are fungal and bacterial xylanases. Thus, the ET biosynthesis-inducing endoxylanase EIX from *T. viride* has been well characterized. When it infiltrates tomato or tobacco leaves, early and late protective responses develop (HSR, ROS generation of and expression of PR protein genes, synthesis of fioalexins) [146]. It has been established that EIX is recognized by tomato plants as MAMP and binds to LRR-type receptors LeEix1 and LeEix2; The HSR and ET production are initiated only by contact with LeEix2, and LeEix1 acts as a decoy receptor [147]. The enzyme is synthesized as a 25 kDa precursor and undergoes post-translational glycosylation. The fungus secretes a mature protein (22 kDa), enriched in glycine, serine, tyrptophan, tyrosine and threonine and free of lysine, alanine, leucine and glutamine. Mutant forms of EIX that have lost endogluconase activity retain the ability to induce HSR. Plant defense responses are also induced by xylanase II from *T. reese*i and xylanases from *F. graminearum* [148]. The elicitor activity of xylanase is attributed to the TKLGE sequence, in which replacement of the terminal threonine (T) and glutamate (E) residues with valine and threonine, respectively, results in the loss of the protective properties of EIX. In endoxylonase Xyn11A from *B. cinerea*, a 25-residue peptide Xyn25 is responsible for elicitor activity, inducing HSR and the expression of a number of protective genes [149]. In turn, the protective activity of this peptide is due to a short conserved sequence of 4 amino acids, but not the catalytic activity [146].

Endocellulase EG1 from *R. solani* contains a putative 227 amino acid protein with a signal peptide and a glycosyl hydrolase domain. By replacing the aspartic acid residue at position 32 with alanine, a catalytically inactive enzyme was ptoduced. This altered enzyme was expressed in yeast and purified to homogeneity. The recombinant EG1 (rEG1), devoid of enzymatic activity, fully retained the properties of the elicitor. It induced cell death in the leaves of corn, tobacco and Arabidopsis, and also enhanced the expression of genes for SAR markers and a number of protective enzymes and PR proteins in corn and tobacco (PR1a, POD, PAL, chitinase, β-1,3-gluconase, etc.). In addition, rEG1 caused the accumulation of ROS, a reversible shift in extracellular pH to alkaline, the accumulation of Ca^{2+} ions and ethylene biosynthesis in a suspension of cultured tobacco cells. In experiments with infection of maize plants with *R. solani*, HSR cell death was also associated with EG1 expression [150].

The MF3 protein (microbial factor 3, 16.9 kDa) was isolated from the culture fluid and cell homogenate of *P. fluorescens*. This protein is capable of inducing a wide-range SR in plants against viruses, fungi, and nematodes [151]. It was identified as peptidyl-prolyl-cis/trans-isomerase, and a fragment of the conserved enzyme region consisting of 29 amino acid residues (IIPGLEKALEGK-AVGDDDLEVAVEPEDAYG) is responsible for its elicitor activity. The peptide retains the protective properties of the full-length protein [151] and can cause the accumulation of SA in tobacco leaves treated with it, possibly due to an increased expression of the isochorismate synthase gene, which catalyzes the SA synthesis.

Glycoproteins and peptidoglycans. Glycoproteins (GPs) result from post-translational glycosylation of proteins and regulate several biological processes vital for microorganisms. Phytopathogenic fungi secrete some of the GPs when infecting plants, using them as effectors that promote virulence. The carbohydrate moiety of such effectors regulates their stability, activity, conformational folding, target transport, and cellular localization [152], but may also determine the effector/elicitor activity of microbial GPs. Thus, GP Slp1 (syn. LysM 1) secreted by *M. oryzae* binds chitin and prevents the interaction of this PAMP with its receptor in rice cells (CEBiP), and only the glycosylated protein acts in this

way. Unglycosylated Slp1 is prone to rapid degradation, so unbound chitin becomes available again to CEBiP and triggers a defense response [153]. GP fractions of the cell walls and culture filtrate of *P oligandrum*, capable of inducing the expression of genes for resistance to phytopathogens and the accumulation of enzymes associated with it in wheat and sugar beet, lost this ability after autoclaving or enzymatic proteolysis. Howeve, periodate, which attacks the carbohydrate fragment of GP, did not affect the elicitor activity of these fractions [154].

Bacterial peptidoglycans represent a separate group of elistors from the category of complex proteins. Since the cell walls of almost all their species contain peptidoglycan, plants recognize it as a nonspecific elicitor. In particular, in Arabidopsis, tobacco, and rice, this bacterial PAMP was shown to activate early defense responses such as an increase in the concentration of calcium ions in the cytoplasm and the generation of ROS [155].

Effectors of phytoviruses. The interaction between viruses and plants serves as a convenient model for studying the molecular mechanisms of antiviral phytoimmunity. The introduction of viral particles into plant cells triggers defense mechanisms, including microwaves, leading to rapid isolation of the pathogen in the cells of plants resistant to it. These processes involve small interfering RNAs, which suppress or disrupt the synthesis of viral nucleic acids, preventing its replication. During evolution, phytoviruses acquired the ability to synthesize specific effectors, i.e., the viral suppressors of RNA silencing (VSR), the main role of which is to suppress RNA interference (RNAi). VSRs have been shown to cause plant gene silencing and thus allow viruses to overcome PTI [156]. VSR, like other viral gene expression products with diverse functions, are recognized by plant receptor R proteins, which leads to viral RNA silencing.

Thus, tobacco plants of the species *N. glutinosa* have a set of proteins that cause RNA silencing of phytopathogenic viruses. Poleroviruses capable of infecting this species have a PO protein that functions as a VSR. The target of PO is the *ARGONAUTE1* gene product AGO1, a plant protein and a catalytic component of the RISC (RNA-induced silencing complex, a protein complex that ensures gene silencing via the RNAi mechanism) [157]. It was noted that the action of other VSRs is also directed towards AGO1. For example, its degradation is caused by P25 of the potato virus X, which leads to the suppression of the HSR in *N. glutinosa*.

The RNAi inhibitory properties of PO of different polerovirus isolates depend on intraspecific differences in its structure. When comparing the suppressive activity of PO from two different isolates of cereal yellow dwarf cereal yellow dwarf *Polerovirus* (amily *Luteoviridae*) CYDV-RPV, it was found that the presence of proline in the C-terminus reduces the stability of this protein and negatively affects its suppressor activity. It is likely that the high conformational rigidity of proline contributes to the structural destabilization of the protein molecule, which increases with increasing temperature. Replacing proline with other amino acids, such as serine, improves the structural stability of PO [158]. It is precisely this substitution that has been found in some natural isolates of poleroviruses that successfully infect cereals. It was also found that a mutation resulting in the replacement of the C-terminal proximal proline with serine in PO with low suppressive activity from the CYDV-RPV isolate which weakly infects plants at elevated temperatures, restores the activity of this protein [156-158]. Interestingly, in natural populations of poleroviruses, mutants spontaneously arise in which proline is replaced by serine. To induce HSR in plants, a complete functional protein PO is required. VSR mutants with amino acid substitutions in the F-box motif of this protein lost their suppressive activity and, as a result, could not cause HSR. Maintaining the structure of the protein F-box motif appears to be important for the ability of PO to cause degradation of the AGO1 protein, which leads to disruption

of the RNA silencing mechanism.

When studying the interaction of *N. glutinosa* plants with the turnip yellow virus (TuYV), the potato leaf roll virus (PLRV), and the cucurbit virus aphid borne yellows virus (CABYV), it was found that the resistance of this species to these viruses is associated with the ability of the PO proteins TuYV (POTu), PLRV (POPL), and CABYV (POCA) to induce HSR in cultivar TW59. Plants of other cultivars of this species recognize only POPL from PLRV [159]. Genetic analysis showed that in *N. glutinosa* the resistance gene *RPO1* (resistance to poleroviruses 1) inherited as a dominant allele is responsible for the ability to recognize POTu. Effector proteins have been identified in many phytoviruses, which, like PO, exhibit RNAi-inhibitory properties, suppressing the protective silencing of viral RNAs in infected plants, therefore silencing is considered the main mechanism of ETI to these pathogens.

Prospects for the use of elicitor proteins of microorganisms in agriculture and examples of protective drugs developed on their basis. Due to the variety of microbial proteins that can be perceived by plants as elicitors, causing rapid development of responses in low doses, activating the general or race-specific SR of plants and leading to long-term protection against phytopathogens, researchers are seeking to evaluate the possibilities of using these elicitors to increase crop yields [26, 160-162]. For example, the protective effectiveness of elicitor proteins from FS-94 has been confirmed by three-year field trials. It has been shown that pre-sowing treatment of spring wheat seeds significantly reduces the prevalence and weakens the symptoms of Fusarium and helminthosporium root rots throughout the growing season, resulting in increased yield [116]. From a practical point of view, the attractiveness of protein elicitors is also due to the fact that some of them increase plant resistance to abiotic stress and have a positive effect on their growth and development (https://doraagri.com/pro-duct/buy-harpin-protein/). In general, the prospects for including elicitors in environmentally friendly scenarios for protecting crops from diseases based on stimulating active phytoimmunity seem very promising. However, this research area still remains relatively new and under-demanded. So far, only a small number of elicitor proteins and commercial drugs created on their basis are known, which have found wide practical application.

The first protein elicitor-based drug for agricultural practeice was harpin. Under the trade name Messenger, by 2000 it was registered in the USA by the Eden Biotechnology Company and approved for use on all crops. The U.S. Environmental Protection Agency awarded the drug the Presidential Green Chemistry Challenge Award for plant protection and agricultural product safety in 2001. The drug was used to protect crops of tobacco, vegetables and fruits in the USA, Mexico, Spain and other countries. Messenger also was temporary registred in China, and since 2007 its use on tomato, pepper, tobacco and rapeseed has been approved.

In 2008, a license for the production of harpin was acquired by the Chinese company Dora Agri to produce the drug Dora Immune (https://doraagri.com/product/buy-harpin-protein/). Over the past five years, the drug has been supplied to the Chinese market, as well as to the markets of Central and South America and Western Europe. It is proposed to be used as a plant health factor, which activates the immune system and affects the expression of genes responsible for growth and activation of defense reactions. Dora Immune is used to treat seeds, roots and leaves and is approved for use on apple, plum, kiwi, nectarine, avocado, mango, citrus fruits, cherries, grapes, hemp, tobacco and some other plants. According to the manufacturer, treating plants with this drug improves product quality, increases yields, protection against diseases, the shelf life of fruits by 5-7 days and their

sugar content by 10-25%. However, such effectiveness is achieved only if the working solution is prepared no more than 6 hours before spraying, and is seeds are treated no more than 30 minutes before planting. In addition, when used together with preparations containing salts of heavy metals, the effectiveness of treatments may noticeably decrease, since the latter adsorb protein, preventing its contact with plant receptors.

Another convincing example is the PeaT1 protein from *A. tenuissima* [129, 130] which is also produced by *A. alternata* [160]. In 2014, the Institute of Plant Protection of China registered PeaT1 in China as the active ingredient of a protective drug with an immunostimulating effect, commercially named ATaiLing [160]. It is manyfactured by the Chinese company Zhongbao Chemicals Co., Limited. It has been shown that ATaiLing inhibits the expression of viral genes and repair pathological changes in plant tissues affected by viruses. In addition, ATaiLing induces multiple plant defense responses against insect pests. The drug is effective against rice streak disease, tomato yellow leaf curl virus, and tobacco mosaic virus. It is important to highlight that it provides protection against one of the most damaging citrus diseases, Huanglongbing (citrus greening disease, or pinyin) caused by *Candidatus* Liberibacter. Due to the high effectiveness of the drug, its sales since entering the market has reached 200 tons per year with an annual profit of 70 million yuan. To date, Atailing is used on 5 million hectares of cultivated areas in China. A number of agrochemical companies are negotiating the possibility of using the drug in America and Western Europe. Currently, in Russia, the Altbiotech company is registering a biological product based on peptidyl-prolyl cis/trans isomerase (MF3) the elicitor properties of which were first discovered and studied at the All-Russian Research Institute of Phytopathology [151]. The structure and elicitor functions of the protein were patented in Russia, European and Asian countries and in the USA [162]. Testing an MF3-based drug in three different agroclimatic zones showed its antiviral effectiveness on commercial potato plantings, the ability to improve the physiological state of plants and the possibility to increase harvest, in toxicological test, no toxicity was shown for laboratory animals. The protein elicitor AMEP412 from *B. subtilis* which induces systemic resistance to bacteria may also be promising as a biocontrol agent [163].

Thus, induced disease resistance becomes an attractive alternative to chemical pesticides in plant protection. The induced response is activated by elicitors, including proteins. Elicitors are recognized by various receptors, control several signaling pathways and induce various protective responses in plans. Despite the fact that a large number of specific and non-specific biogenic elicitors are currently known, only few are used in practice to protect agricultural crops. Nevertheless, a wide arsenal of elicitor proteins in microorganisms, the ongoing intensive search for new microbial proteins that activate plan immunity, and innovative methods for their screening suggest that new elicitor proteins will be identified to become the basis of drugs for crop protection.

REFERENCES

1. Nelson R., Wiesner-Hanks T., Wisser R., Balint-Kurti P. Navigating complexity to breed disease-resistant crops. *Nat. Rev. Genet.*, 2018, 19(1): 21-33 (doi: 10.1038/nrg.2017.82).
2. *Food and Agriculture Organization. New standards to curb the global spread of plant pests and diseases.* Available: https://www.fao.org/news/story/en/item/1187738/icode/. Accessed: 28.05.2023.
3. Hawkins N.J., Bass C., Dixon A., Neve P. The evolutionary origins of pesticide resistance. *Biol. Rev.*, 2019, 94(1): 135-155 (doi: 10.1111/brv.12440).
4. Fisher M.C., Hawkins N.J., Sanglard D., Gurr S.J. Worldwide emergence of resistance to antifungal drugs challenges human health and food security. *Science*, 2018, 360(6390): 739-742 (doi: 10.1126/science.aap7999).
5. Stuthman D.D., Leonard K.J., Miller-Garvin J. Breeding crops for durable resistance to disease.

Advances in Agronomy, 2007, 95: 319-367 (doi: 10.1016/s0065-2113(07)95004-x).
6. Conrath U., Beckers G. J., Flors V., Garcia-Agustin P., Jakab G., Mauch F., Newman M.-A., Pieterse C.M.J., Poinssot B., Pozo M.J., Pugin A., Schaffrath U., Ton J., Wendehenne D., Zimmerli L., Mauch-Mani B. Priming: getting ready for battle. *MPMI*, 2006, 19(10): 1062-1071 (doi: 10.1094/MPMI-19-1062).
7. Wiesel L., Newton A.C., Elliott I., Booty D., Gilroy E.M., Birch P.R.J., Hein I. Molecular effects of resistance elicitors from biological origin and their potential for crop protection. *Front. Plant Sci.*, 2014, 21(5): 655 (doi: 10.3389/fpls.2014.00655).
8. Klessig D.F., Choi H.W., D'Maris D.A. Systemic acquired resistance and salicylic acid: past, present, and future. *MPMI*, 2018, 31(9): 871-888 (doi: 10.1094/MPMI-03-18-0067-CR).
9. Conrath U. Molecular aspects of defence priming. *Trends in Plant Science*, 2011, 16(10): 524-531 (doi: 10.1016/j.tplants.2011.06.004).
10. Pastor V., Luna E., Mauch-Mani B., Ton J., Flors V. Primed plants do not forget. *Environmental and Experimental Botany*, 2013, 94: 46-56 (doi: 10.1016/j.envexpbot.2012.02.013).
11. Vlot A.C., Pabst E., Riedlmeier M. Systemic signalling in plant defence. In: *eLS*. John Wiley & Sons Ltd, Chichester, 2017.
12. Andersen E.J., Ali S., Byamukama E., Yen Y., Nepal M.P. Disease resistance mechanisms in plants. *Genes*, 2018, 9(7): 339 (doi: 10.3390/genes9070339).
13. Kamle M., Borah R., Bora H., Jaiswal A.K., Singh R.K., Kumar P. Systemic acquired resistance (SAR) and induced systemic resistance (ISR): role and mechanism of action against phytopathogens. In: *Fungal biotechnology and bioengineering*. A.L. Hesham, R. Upadhyay, G. Sharma, C. Manoharachary, V. Gupta (eds.). Springer, Cham, 2020.
14. Jones J.D., Dangl J.L. The plant immune system. *Nature*, 2006, 444(7117): 323-329 (doi: 10.1038/nature05286).
15. Newman M.A., Sundelin T., Nielsen J.T., Erbs G. MAMP (microbe- associated molecular pattern) triggered immunity in plants. *Front. Plant Sci.*, 2013, 4: 139 (doi: 10.3389/fpls.2013.00139).
16. Abdul Malik N.A., Kumar I.S., Nadarajah K. Elicitor and receptor molecules: orchestrators of plant defense and immunity. *Int. J. Mol. Sci.*, 2020, 21(3): 963 (doi: 10.3390/ijms21030963).
17. Medina-Puche L., Rufián J.S. Role of receptor-like kinases in plant-pathogen interaction. In: *Plant receptor-like kinases*. S.K. Upadhyay, Shumayla (eds.). Academic Press, Cambridge, 2023. (doi: 10.1016/B978-0-323-90594-7.00014-4).
18. Meng X., Zhang S. MAPK cascades in plant disease resistance signaling. *Annual Review of Phytopathology*, 2013, 51: 245-266 (doi: 10.1146/annurev-phyto-082712-102314).
19. Kadota Y., Shirasu K., Zipfel C. Regulation of the NADPH oxidase RBOHD during plant immunity. *Plant and Cell Physiology*, 2015, 56(8): 1472-1480 (doi: 10.1093/pcp/pcv063).
20. Yuan P., Tanaka K., Du L., Poovaiah B.W. Calcium signaling in plant autoimmunity: a guard model for AtSR1/CAMTA3-mediated immune response. *Molecular Plant*, 2018, 11(5): 637-639 (doi: 10.1016/j.molp.2018.02.014).
21. Zhang Y., Li X. Salicylic acid: biosynthesis, perception, and contributions to plant immunity. *Current Opinion in Plant Biology*, 2019, 50: 29-36 (doi: 10.1016/j.pbi.2019.02.004).
22. Zhong C.-L., Zhang C., Liu J.-Z. Hetero-trimeric G protein signaling in plant immunity. *Journal of Experimental Botan*, 2018, 70(4): 1109-1118 (doi: 10.1093/jxb/ery426).
23. Dodds P.N., Rathjen J.P. Plant immunity: towards an integrated view of plant—pathogen interactions. *Nat. Rev. Genet.*, 2010, 11(8): 539-548 (doi: 10.1038/nrg2812).
24. Pruitt R.N., Gust A.A., Nürnberger T. Plant immunity unified. *Nature Plants*, 2021, 7(4): 382-383 (doi: 10.1038/s41477-021-00903-3).
25. Remick B.C., Gaidt M.M., Vance R.E. Effector-triggered immunity. *Annual Review of Immunology*, 2023, 41: 453-481 (doi: 10.1146/annurev-immunol-101721-031732).
26. Mejía-Teniente L., Torres-Pacheco I., González-Chavira M.M., Ocampo-Velazquez R.V., Herrera-Ruiz G., Chapa-Oliver A.M., Guevara-González R.G. Use of elicitors as an approach for sustainable agriculture. *African Journal of Biotechnology*, 2010, 9(54): 9155-9162.
27. Naito K., Taguchi F., Suzuki T., Inagaki Y., Toyoda K., Shiraishi T., Ichinose Y. Amino acid sequence of bacterial microbe-associated molecular pattern flg22 is required for virulence. *MPMI*, 2008, 21(9): 1165-1174 (doi: 10.1094/MPMI-21-9-1165).
28. Zipfel C. Early molecular events in PAMP-triggered immunity. *Current Opinion in Plant Biology*, 2009, 12(4): 414-420 (doi: 10.1016/j.pbi.2009.06.003).
29. Wang B., Yang X., Zeng H., Liu H., Zhou T., Tan B., Yuan J., Guo L., Qiu D. The purification and characterization of a novel hypersensitive-like response-inducing elicitor from *Verticillium dahliae* that induces resistance responses in tobacco. *Appl. Microbiol. Biotechnol.*, 2012, 93(1): 191-201 (doi: 10.1007/s00253-011-3405-1).
30. Xu Y., Chen H., Zhou X., Cai X. Induction of hypersensitive response and nonhost resistance by a *Cladosporium fulvum* elicitor CfHNNI1 is dose-dependent and negatively regulated by salicylic acid. *Journal of Integrative Agriculture*, 2012, 11(5): 1665-1674 (doi: 10.1016/S2095-3119(12)60169-5).
31. Mishra A.K., Sharma K., Misra R.S. Elicitor recognition, signal transduction and induced resistance in plants. *Journal of Plant Interactions*, 2012, 7(2): 95-120 (doi: 10.1080/17429145.2011.597517).

32. Erbs G., Newman M.A. The role of lipopolysaccharide and peptidoglycan, two glycosylated bacterial microbe-associated molecular patterns (MAMPs), in plant innate immunity. *Molecular Plant Pathology*, 2012, 13(1): 95-104 (doi: 10.1111/j.1364-3703.2011.00730.x).
33. Albert M. Peptides as triggers of plant defence. *Journal of Experimental Botany*, 2013, 64(17): 5269-5279 (doi: 10.1093/jxb/ert275).
34. Boller T., He S.Y. Innate immunity in plants: an arms race between pattern recognition receptors in plants and effectors in microbial pathogens. *Science*, 2009, 324(5928): 742-744 (doi: 10.1126/science.1171647).
35. Tyuterev S.L. *Vestnik zashchity rasteniy*, 2015, 1(83): 3-13 (in Russ.).
36. Khavkin E.E. *Fiziologiya rasteniy*, 2021, 68(2): 115-131 (doi: 10.31857/S001533032102007X) (in Russ.).
37. Popova E.V., Domnina N.S., Sokornova C.V., Kovalenko N.M., Tyuterev S.L. Novel hybrid modulators of plant immune responses based on chitosan and bioactive anti-oxidants and pro-oxidants. *Sel'skokhozyaistvennaya biologiya* [*Agricultural Biology*], 2021, 26(1): 158-170 (doi: 10.15389/agrobiology.2021.1.158eng).
38. Slavokhotova A.A., Shelenkov A.A., Andreev Ya.A., Odintsova T.I. *Uspekhi biologicheskoy khimii*, 2017, 57: 209-244 (in Russ.).
39. Shcherbakova L.A., Dzhavakhiya V.G. *Izvestiya Samarskogo nauchnogo tsentra Rossiyskoy akademii nauk*, 2013, 15(3-5): 1705-1709 (in Russ.).
40. Ayliffe M., Sørensen C. Plant non-host resistance: paradigms and new environments. *Current Opinion in Plant Biology*, 2019, 50: 104-113 (doi: 10.1016/j.pbi.2019.03.011).
41. Guo J., Cheng Y. Advances in fungal elicitor-triggered plant immunity. *Int. J. Mol. Sci.*, 2022, 23(19): 12003 (doi: 10.3390/ijms231912003).
42. Gómez-Gómez L., Boller T. Flagellin perception: a paradigm for innate immunity. *Trends in Plant Science*, 2002 7(6): 251-256 (doi: 10.1016/s1360-1385(02)02261-6).
43. Lu Y., Swartz J.R. Functional properties of flagellin as a stimulator of innate immunity. *Sci. Rep.*, 2016, 6: 18379 (doi: 10.1038/srep18379).
44. Gómez-Gómez L., Boller T. FLS2: an LRR receptor-like kinase involved in the perception of the bacterial elicitor flagellin in *Arabidopsis*. *Molecular Cell*, 2000, 5(6): 1003-1011 (doi: 10.1016/s1097-2765(00)80265-8).
45. Boller T., Felix G. A renaissance of elicitors: perception of microbe-associated molecular patterns and danger signals by pattern-recognition receptors. *Annual Review of Plant Biology*, 2009, 60: 379-406 (doi: 10.1146/annurev.arplant.57.032905.105346).
46. Shiu S.-H., Karlowski W.M., Pan R., Tzeng Y.-H., Mayer K.F., Li W.-H. Comparative analysis of the receptor-like kinase family in *Arabidopsis* and rice. *The Plant Cell*, 2004, 16(5): 1220-1234 (doi: 10.1105/tpc.020834).
47. Jelenska J., Davern S.M., Standaert R.F., Mirzadeh S., Greenberg J.T. Flagellin peptide flg22 gains access to long-distance trafficking in *Arabidopsis* via its receptor, FLS2. *Journal of Experimental Botany*, 2017, 68(7): 1769-1783 (doi: 10.1093/jxb/erx060).
48. Fliegmann J., Felix G. Immunity: flagellin seen from all sides. *Nature Plants*, 2016, 2(9): 16136 (doi: 10.1038/nplants.2016.136).
49. Hind S.R., Strickler S.R., Boyle P.C., Dunham D.M., Bao Z., O'Doherty I.M., Baccile J.A., Hoki J.S., Viox E.G., Clarke C.R. Vinatzer B.A., Schroeder F.C., Martin G.B. Tomato receptor FLAGELLIN-SENSING 3 binds flgII-28 and activates the plant immune system. *Nature Plants*, 2016, 2(9): 1-8 (doi: 10.1038/nplants.2016.128).
50. Chinchilla D., Zipfel C., Robatzek S., Kemmerling B., Nürnberger T., Jones J.D., Felix G., Boller T. A flagellin-induced complex of the receptor FLS2 and BAK1 initiates plant defence. *Nature*, 2007, 448(7152): 497-500 (doi: 10.1038/nature05999).
51. Macho A.P., Zipfel C. Plant PRRs and the activation of innate immune signaling. *Molecular Cell*, 2014, 54(2): 263-272 (doi: 10.1016/j.molcel.2014.03.028).
52. Wei Y., Caceres-Moreno C., Jimenez-Gongora T., Wang K., Sang Y., Lozano-Duran R., Macho A.P. The *Ralstonia solanacearum* csp22 peptide, but not flagellin-derived peptides, is perceived by plants from the *Solanaceae* family. *Plant Biotechnol. J.*, 2018, 16(7): 1349-1362 (doi: 10.1111/pbi.12874).
53. Cai R., Lewis J., Yan S., Liu H., Clarke C.R., Campanile F., Almeida N.F., Studholme D.J., Lindeberg M., Schneider D., Zaccardelli M., Setubal J.C., Morales-Lizcano N.P., Bernal A., Coaker G., Baker C., Bender C.L., Leman S., Vinatzer B.A. The plant pathogen *Pseudomonas syringae* pv. *tomato* is genetically monomorphic and under strong selection to evade tomato immunity. *PLOS Pathog.*, 2011, 7(8): e1002130 (doi: 10.1371/journal.ppat.1002130).
54. Moroz N., Tanaka K. FlgII-28 is a major flagellin-derived defense elicitor in potato. *MPMI,* 2020, 33(2): 247-255 (doi: 10.1094/MPMI-06-19-0164-R).
55. Haney C.H., Ausubel F.M., Urbach J.M. Innate immunity in plants and animals: differences and similarities. *The Biochemist*, 2014, 36(5): 40-45 (doi: 10.1042/BIO03605040).
56. Kunze G., Zipfel C., Robatzek S., Niehaus K., Boller T., Felix G. The N terminus of bacterial elongation factor Tu elicits innate immunity in *Arabidopsis* plants. *The Plant Cell*, 2004, 16(12): 3496-3507 (doi: 10.1105/tpc.104.026765).

57. Xu G., Greene G.H., Yoo H., Liu L., Marqués J., Motley J., Dong X. Global translational reprogramming is a fundamental layer of immune regulation in plants. *Nature*, 2017, 545(7655): 487-490 (doi: 10.1038/nature22371).
58. Wei Z.-M., Laby R.J., Zumoff C.H., Bauer D.W., He S.Y., Collmer A., Beer S.V. Harpin, elicitor of the hypersensitive response produced by the plant pathogen *Erwinia amylovora*. *Science*, 1992, 257(5066): 85-88 (doi: 10.1126/science.1621099).
59. He S.Y., Huang H.C., Collmer A. *Pseudomonas syringae* pv. *syringae* harpin$_{Pss}$: a protein that is secreted via the Hrp pathway and elicits the hypersensitive response in plants. *Cell*, 1993, 73(7): 1255-1266 (doi: 10.1016/0092-8674(93)90354-s).
60. Kim J.G., Jeon E., Oh J., Moon J.S., Hwang I. Mutational analysis of *Xanthomonas* harpin HpaG identifies a key functional region that elicits the hypersensitive response in nonhost plants. *Journal of Bacteriology*, 2004, 186(18): 6239-6247 (doi: 10.1128/JB.186.18.6239-6247.2004).
61. Tarafdar P.K., Vedantam L.V., Kondreddy A., Podile A.R., Swamy M.J. Biophysical investigations on the aggregation and thermal unfolding of harpin$_{Pss}$ and identification of leucine-zipper-like motifs in harpins. *Biochimica et Biophysica Acta (BBA) — Proteins and Proteomics*, 2009, 1794(11): 1684-1692 (doi: 10.1016/j.bbapap.2009.07.023).
62. Choi M.-S., Kim W., Lee C., Oh C.-S. Harpins, multi-functional proteins secreted by gram-negative plant-pathogenic bacteria. *MPMI*, 2013, 26(10): 1115-1122 (doi: 10.1094/MPMI-02-13-0050-CR).
63. Dong H.-P., Yu H., Bao Z., Guo X., Peng J., Yao Z., Chen G., Qu S., Dong H. The ABI2-dependent abscisic acid signalling controls HrpN-induced drought tolerance in *Arabidopsis*. *Planta*, 2005, 221(3): 313-327 (doi: 10.1007/s00425-004-1444-x).
64. Yang Y., Chen T., Dai X., Yang D., Wu Y., Chen H., Zheng Y., Zhi Q., Wan X., Tan X. Comparative transcriptome analysis revealed molecular mechanisms of peanut leaves responding to *Ralstonia solanacearum* and its type III secretion system mutant. *Front. Microbiol.*, 2022, 13: 998817 (doi: 10.3389/fmicb.2022.99881).
65. Xie L., Liu Y., Wang H., Liu W., Di R., Miao W., Zheng F. Characterization of harpin Xoo induced hypersensitive responses in non host plant, tobacco. *J. Plant Biochem. Biotechnol.*, 2017, 26(1): 73-79 (doi: 10.1007/s13562-016-0363-9).
66. Zou L.-F., Wang X.-P., Xiang Y., Zhang B., Li Y.-R., Xiao Y.-L., Wang J.-S., Walmsley A.R., Chen G.-Y. Elucidation of the *hrp* clusters of *Xanthomonas oryzae* pv. *oryzicola* that control the hypersensitive response in nonhost tobacco and pathogenicity in susceptible host rice. *Applied and Environmental Microbiology*, 2006, 72(9): 6212-6224 (doi: 10.1128/AEM.00511-06).
67. Cho H.-J., Park Y.-J., Noh T.-H., Kim Y.-T., Kim J.-G., Song E.-S., Lee D.-H., Lee B.-M. Molecular analysis of the *hrp* gene cluster in *Xanthomonas oryzae* pathovar *oryzae* KACC10859. *Microbial Pathogenesis*, 2008, 44(6): 473-483 (doi: 10.1016/j.micpath.2007.12.002).
68. Liu Y., Zhou X., Liu W., Xiong X., Lv C., Zhou X., Miao W. Functional regions of HpaXm as elicitors with specific heat tolerance induce the hypersensitive response or plant growth promotion in nonhost plants. *PLoS ONE*, 2018, 13(1): e0188788 (doi: 10.1371/journal.pone.0188788).
69. Miao W.-G., Song C.-F., Wang Y., Wang J.-S. HpaXm from *Xanthomonas citri* subsp. *malvacearum* is a novel harpin with two heptads for hypersensitive response. *J. Microbiol. Biotechnol.*, 2010, 20(1): 54-62.
70. Haapalainen M., Engelhardt S., Kuefner I., Li C.M., Nuernberger T., Lee J., Romantschuk M., Taira S. Functional mapping of harpin HrpZ of *Pseudomonas syringae* reveals the sites responsible for protein oligomerization, lipid interactions and plant defence induction. *Molecular Plant Pathology*, 2011, 12(2): 151-166 (doi: 10.1111/j.1364-3703.2010.00655.x).
71. Wu H., Wang S., Qiao J., Liu J., Zhan J., Gao X. Expression of HpaGXooc protein in *Bacillus subtilis* and its biological functions. *J. Microbiol. Biotechnol.*, 2009, 19(2): 194-203 (doi: 10.4014/jmb.0802.154).
72. Chen G.-Y., Zhang B., Wu X.-M., Zhao M.-Q., Wei S., Wu X.B. Cloning and characterization of an harpin encoding gene from *Xanthomonas axonopodis* pv. *glycines* required for hypersensitive response on nonhost plant tobacco. *Wei Sheng Wu Xue Bao*, 2005, 45(4): 496-499.
73. Palmieri A.C.B., Amaral A.M., Homem R.A., Machado M.A. Differential expression of pathogenicity- and virulence-related genes of *Xanthomonas axonopodis* pv. *citri* under copper stress. *Genet. Mol. Biol.*, 2010, 33(2): 348-353 (doi: 10.1590/S1415-47572010005000030).
74. Wang X., Zhang L., Ji H., Mo X., Li P., Wang J., Dong H. Hpa1 is a type III translocator in *Xanthomonas oryzae* pv. *oryzae*. *BMC Microbiol.*, 2018, 18(1): 105 (doi: 10.1186/s12866-018-1251-3).
75. Niu X.-N., Wei Z.-Q., Zou H.-F., Xie G.-G., Wu F., Li K.-J., Jiang W., Tang J.-L., He Y.-Q. Complete sequence and detailed analysis of the first indigenous plasmid from *Xanthomonas oryzae* pv. *oryzicola*. *BMC Microbiol.*, 2015, 15(1): 233 (doi: 10.1186/s12866-015-0562-x).
76. Solé M., Scheibner F., Hoffmeister A.-K., Hartmann N., Hause G., Rother A., Jordan M., Lautier M., Arlat M., Büttner D. *Xanthomonas campestris* pv. *vesicatoria* secretes proteases and xylanases via the Xps type II secretion system and outer membrane vesicles. *Journal of Bacteriology*, 2015, 197(17): 2879-2893 (doi: 10.1128/JB.00322-15).
77. Fu M., Xu M., Zhou T., Wang D., Tian S., Han L., Dong H., Zhang C. Transgenic expression

of a functional fragment of harpin protein Hpa1 in wheat induces the phloem-based defence against English grain aphid. *Journal of Experimental Botany*, 2014, 65(6): 1439-1453 (doi: 10.1093/jxb/ert488).
78. Kvitko B.H., Ramos A.R., Morello J.E., Oh H.-S., Collmer A. Identification of harpins in *Pseudomonas syringae* pv. *tomato* DC3000, which are functionally similar to HrpK1 in promoting translocation of type III secretion system effectors. *Journal of Bacteriology*, 2007, 189(22): 8059-8072 (doi: 10.1128/JB.01146-07).
79. Tampakaki A.P., Panopoulos N.J. Elicitation of hypersensitive cell death by extracellularly targeted $HrpZ_{Pph}$ produced in planta. *MPMI,* 2000*,* 13(12): 1366-1374 (doi: 10.1094/MPMI.2000.13.12.1366).
80. Reboutier D., Frankart C., Briand J., Biligui B., Laroche S., Rona J.-P., Barny M.-A., Bouteau F. The $HrpN_{ea}$ harpin from *Erwinia amylovora* triggers differential responses on the nonhost *Arabidopsis thaliana* cells and on the host apple cells. *MPMI*, 2007, 20(1): 94-100 (doi: 10.1094/MPMI-20-0094).
81. Li P., Zhang L., Mo X., Ji H., Bian H., Hu Y., Majid T., Long J., Pang H., Tao Y., Ma J., Dong H. Rice aquaporin PIP1;3 and harpin Hpa1 of bacterial blight pathogen cooperate in a type III effector translocation. *Journal of Experimental Botany*, 2019, 70(12): 3057-3073 (doi: 10.1093/jxb/erz130).
82. Grant S.R., Fisher E.J., Chang J.H., Mole B.M., Dangl J.L. Subterfuge and manipulation: type III effector proteins of phytopathogenic bacteria. *Annual Review of Microbiology*, 2006, 60: 425-449 (doi: 10.1146/annurev.micro.60.080805.142251).
83. Engelhardt S., Lee J., Gabler Y., Kemmerling B., Haapalainen M.-L., Li C.-M., Wei Z., Keller H., Joosten M., Taira S., Nurnberger T. Separable roles of the *Pseudomonas syringae* pv. *phaseolicola* accessory protein HrpZ1 in ion-conducting pore formation and activation of plant immunity. *The Plant Journal*, 2009, 57(4): 706-717 (doi: 10.1111/j.1365-313X.2008.03723.x).
84. Oh C.S., Beer S.V. AtHIPM, an ortholog of the apple HrpN-interacting protein, is a negative regulator of plant growth and mediates the growth-enhancing effect of HrpN in *Arabidopsis*. *Plant Physiology*, 2007, 145(2): 426-436 (doi: 10.1104/pp.107.103432).
85. Chang X., Nick P. Defense signaling triggered by flg22 and harpin is integrated into a different stilbene output in *Vitis* cells. *PLoS ONE, 2012,* 7(7): e40446 (doi: 10.1371/journal.pone.0040446).
86. Sang S., Li X., Gao R., You Z., Lь B., Liu P., Ma Q., Dong H. Apoplastic and cytoplasmic location of harpin protein Hpa1Xoo plays different roles in H_2O_2 generation and pathogen resistance in Arabidopsis. *Plant Mol. Biol.*, 2012, 79(4-5): 375-391 (doi: 10.1007/s11103-012-9918-x).
87. Pavli O.I., Kelaidi G.I., Tampakaki A.P., Skaracis G.N. The *hrpZ* gene of *Pseudomonas syringae* pv. *phaseolicola* enhances resistance to rhizomania disease in transgenic *Nicotiana benthamiana* and sugar beet. *PLoS ONE,* 2011, 6(3): e17306 (doi: 10.1371/journal.pone.0017306).
88. Dong H.-P., Peng J., Bao Z., Meng X., Bonasera J.M., Chen G., Beer S.V., Dong H. Downstream divergence of the ethylene signaling pathway for harpin-stimulated Arabidopsis growth and insect defense. *Plant Physiology*, 2004, 136(3): 3628-3638 (doi: 10.1104/pp.104.048900).
89. Chen L., Zhang S.-J., Zhang S.-S., Qu S., Ren X., Long J., Yin Q., Qian J., Sun F., Zhang C. Wang L., Wu X., Wu T., Zhang Z., Cheng Z., Hayes M., Beer S.V., Dong H. A fragment of the *Xanthomonas oryzae* pv. *oryzicola* harpin $HpaG_{Xooc}$ reduces disease and increases yield of rice in extensive grower plantings. *Phytopathology*, 2008, 98(7): 792-802 (doi: 10.1094/PHYTO-98-7-0792).
90. Wang D., Wang Y., Fu M., Mu S., Han B., Ji H., Cai H., Dong H., Zhang C. Transgenic expression of the functional fragment Hpa110-42 of the harpin protein Hpa1 imparts enhanced resistance to powdery mildew in wheat. *Plant Disease*, 2014, 98(4): 448-455 (doi: 10.1094/PDIS-07-13-0687-RE).
91. Sands L.B., Cheek T., Reynolds J., Ma Y., Berkowitz G.A. Effects of harpin and flg22 on growth enhancement and pathogen defense in *Cannabis sativa* seedlings. *Plants*, 2022, 11(9): 1178 (doi: 10.3390/plants11091178).
92. Chang X., Seo M., Takebayashi Y., Kamiya Y., Riemann M., Nick P. Jasmonates are induced by the PAMP flg22 but not the cell death-Inducing elicitor harpin in *Vitis rupestris*. *Protoplasma*, 2017, 254(1): 271-283 (doi: 10.1007/s00709-016-0941-7).
93. Joshi J.B., Senthamilselvi D., Maupin-Furlow J.A., Uthandi S. Microbial protein elicitors in plant defense. In: *Microbial biocontrol: sustainable agriculture and phytopathogen management*. A. Kumar (ed.). Springer, Cham, 2022 (doi: 10.1007/978-3-030-87512-1_10).
94. Dong H., Delaney T.P., Bauer D.W., Beer S.V. Harpin induces disease resistance in Arabidopsis through the systemic acquired resistance pathway mediated by salicylic acid and the *NIM1* gene. *Plant J.*, 1999, 20(2): 207-215 (doi: 10.1046/j.1365-313x.1999.00595.x).
95. Shoi M.S., Kim W., Lee C., Oh C.S. Harpins, multifunctional proteins secreted by gram-negative plant-pathogenic bacteria. *MPMI*, 2013, 26(10): 1115-1122 (doi: 10.1094/MPMI-02-13-0050-CR).
96. Zhang N., Zhou S., Yang D., Fan Z. Revealing shared and distinct genes responding to JA and SA signaling in *Arabidopsis* by meta-analysis. *Front. Plant Sci.*, 2020, 11: 908 (doi:

10.3389/fpls.2020.00908).
97. Skabkin M.A., Skabkina O.V., Ovchinnikov L.P. *Uspekhi biologicheskoy khimii*, 2004, 44: 3-52 (in Russ.).
98. Eshwar A.K., Guldimann C., Oevermann A., Tasara T. Cold-shock domain family proteins (Csps) are involved in regulation of virulence, cellular aggregation, and flagella-based motility in *Listeria monocytogenes*. *Front. Cell. Infect. Microbiol.*, 2017, 7: 453 (doi: 10.3389/fcimb.2017.00453).
99. Felix G., Boller T. Molecular sensing of bacteria in plants. The highly conserved RNA-binding motif RNP-1 of bacterial cold shock proteins is recognized as an elicitor signal in tobacco. *Journal of Biological Chemistry*, 2003, 278(8): 6201-6208 (doi: 10.1074/jbc.M209880200).
100. Wang L., Albert M., Einig E., Fürst U., Krust D., Felix G. The pattern-recognition receptor CORE of *Solanaceae* detects bacterial cold-shock protein. *Nature Plants*, 2016, 2: 16185 (doi: 10.1038/nplants.2016.185).
101. Djavakhia V.G., Nikolaev O.N., Voinova T.M., Battchikova N.A., Korpela T., Khomutov R.M. DNA sequence of gene and amino acid sequence of protein from *Bacillus thuringiensis*, which induces nonspecific resistance of plants to viral and fungal diseases. *Journal of Russian Phytopathological Society*, 2000, 1: 75-81.
102. Shcherbakova L.A. Some natural proteinaceous and polyketide compounds in plant protection and their potential in green consumerization. In: *Natural products in plant pest management*. N.K. Dubey (ed.). CABI International, Boston, 2011 (doi: 10.1079/9781845936716.0109).
103. Kromina K.A., Dzhavakhiya V.G. *Molekulyarnaya genetika*, *mikrobiologiya i virusologiya*, 2006, 1: 31-34 (in Russ.).
104. de Wit P.J., Laugé R., Honée G., Joosten M.H., Vossen P., Kooman-Gersmann M., Vogelsang R., Vervoort J.J. Molecular and biochemical basis of the interaction between tomato and its fungal pathogen *Cladosporium fulvum*. *Antonie Van Leeuwenhoek*, 1997, 71(1-2): 137-141 (doi: 10.1023/a:1000102509556).
105. Ökmen B., de Wit P.J.G.M. *Cladosporium fulvum*-tomato pathosystem: fungal infection strategy and plant responses. In: *Molecular plant immunity*. G. Sessa (ed.). John Wiley & Sons Ltd., West Sussex, 2013 (doi: 10.1002/9781118481431.ch10).
106. Wan J., Zhang X.-C., Neece D., Ramonell K.M., Clough S., Kim S.-Y., Stacey M.G., Stacey G. A LysM receptor-like kinase plays a critical role in chitin signaling and fungal resistance in *Arabidopsis*. *Plant Cell*, 2008, 20(2), 471-481 (doi: 10.1105/tpc.107.056754).
107. Liebrand T.W., van den Berg G.C., Zhang Z., Smit P., Cordewener J.H., America A.H., Sklenar J., Jones A.M., Tameling W.I., Robatzek S., Thomma B.P., Joosten M.H. Receptor-like kinase SOBIR1/EVR interacts with receptor-like proteins in plant immunity against fungal infection. *Proc. Natl. Acad. Sci. USA*, 2013, 110(24): 10010-10015 (doi: 10.1073/pnas.1220015110).
108. Wulff B.B., Chakrabarti A., Jones D.A. Recognitional specificity and evolution in the tomato–*Cladosporium fulvum* pathosystem. *MPMI*, 2009, 22(10): 1191-1202 (doi: 10.1094/MPMI-22-10-1191).
109. Sánchez-Vallet A., Saleem-Batcha R., Kombrink A., Hansen G., Valkenburg D.J., Thomma B.P., Mesters J.R. Fungal effector Ecp6 outcompetes host immune receptor for chitin binding through intrachain LysM dimerization. *eLife*, 2013, 2: e00790 (doi: 10.7554/eLife.00790).
110. Thomma B.P., Nürnberger T., Joosten M.H. Of PAMPs and effectors: the blurred PTI-ETI dichotomy. *The Plant Cell*, 2011, 23(1): 4-15 (doi: 10.1105/tpc.110.082602).
111. Kombrink A., Sanchez-Vallet A., Thomma B.P. The role of chitin detection in plant-pathogen interactions. *Microbes Infect.*, 2011, 13(14-15): 1168-1176 (doi: 10.1016/j.micinf.2011.07.010)
112. Rep M., van der Does H.C., Meijer M., van Wijk R., Houterman P.M., Dekker H.L., de Koster C.G., Cornelissen B.J.C. A small, cysteine-rich protein secreted by *Fusarium oxysporum* during colonization of xylem vessels is required for I-3-mediated resistance in tomato. *Molecular Microbiology*, 2004, 53(5): 1373-1383 (doi: 10.1111/j.1365-2958.2004.04177.x).
113. Takken F., Rep M. The arms race between tomato and *Fusarium oxysporum*. *Molecular Plant Pathology*, 2010, 11(2): 309-314 (doi: 10.1111/j.1364-3703.2009.00605.x).
114. Shcherbakova L., Odintsova T., Stakheev A., Fravel D. Zavriev S. Identification of a novel small cysteine-rich protein in the fraction from the biocontrol *Fusarium oxysporum* strain CS-20 that mitigates *Fusarium* wilt symptoms and triggers defense responses in tomato. *Front. Plant Sci.*, 2015, 6: 1207 (doi: 10.3389/fpls.2015.01207).
115. Shcherbakova L.A., Nazarova T.A., Mikityuk O.D., Fravel D.R. *Fusarium sambucinum* isolate FS-94 690 induces resistance against *Fusarium* wilt of tomato via activation and priming of a salicylic acid-dependent signaling system. *Russ. J. Plant Physiology*, 2011, 58: 808-818 (doi: 10.1134/S1021443711050207).
116. Shcherbakova L.A., Nazarova T.A., Mikityuk O.D, Istomina E.A., Odintsova T.I. An extract purified from the mycelium of a tomato wilt-controlling strain of *Fusarium sambucinum* is able to protect wheat against *Fusarium* and common root rots. *Pathogens*, 2018, 7(3): 61 (doi: 10.3390/pathogens7030061).
117. Keates S.E., Kostman T.A., Anderson J.D., Bailey B.A. Altered gene expression in three plant species in response to treatment with Nep1, a fungal protein that causes necrosis, *Plant Physiology*, 2003, 132(3): 1610-1622 (doi: 10.1104/pp.102.019836).

118. Oomea S., Raaymakersa T.M., Cabrala A., Samwela S., Böhm H., Albert I., Nürnberger T., van den Ackerveken G. Nep1-like proteins from three kingdoms of life act as a microbe-associated molecular pattern in *Arabidopsis*. *Proc. Natl. Acad. Sci. USA*, 2014, 111(47): 16955-16960 (doi: 10.1073/pnas.1410031111).
119. van't Slot K. A., van den Burg H.A., Kloks C.P., Hilbers C.W., Knogge W., Papavoine C.H. Solution structure of the plant disease resistance-triggering protein NIP1 from the fungus *Rhynchosporium secalis* shows a novel beta-sheet fold. *Journal of Biological Chemistry*, 2003, 278(46): 45730-45736 (doi: 10.1074/jbc.M308304200).
120. Djonović S., Pozo M.J., Dangott L.J., Howell C.R., Kenerley C.M. Sm1, a proteinaceous elicitor secreted by the biocontrol fungus *Trichoderma virens* induces plant defense responses and systemic resistance. *MPMI*, 2006, 19(8): 838-853 (doi: 10.1094/MPMI-19-0838).
121. Seidl V., Marchetti M., Schandl R., Allmaier G., Kubicek C.P. Epl1, the major secreted protein of *Hypocrea atroviridis* on glucose, is a member of a strongly conserved protein family comprising plant defense response elicitors. *The FEBS Journal*, 2006, 273(18): 4346-4359 (doi: 10.1111/j.1742-4658.2006.05435.x).
122. Wang Y., Song J., Wu Y., Odeph M., Liu Z., Howlett B.J., Wang S., Yang P., Yao L., Zhao L., Yang Q. Eplt4 proteinaceous elicitor produced in *Pichia pastoris* has a protective effect against *Cercosporidium sofinum* infections of soybean leaves. *Appl. Biochem. Biotechnol.*, 2013, 169(3): 722-737 (doi: 10.1007/s12010-012-0015-z).
123. Ruocco M., Lanzuise S., Lombardi N., Woo S.L., Vinale F., Marra R., Varlese R., Manganiello G., Pascale A., Scala V., Turrà D., Scala F., Lorito M. Multiple roles and effects of a novel *Trichoderma* hydrophobin. *MPMI*, 2015, 28(2): 167-179 (doi: 10.1094/MPMI-07-14-0194-R).
124. Zhang W., Fraiture M., Kolb D., Löffelhardt B., Desaki Y., Boutrot F.F., Tör M., Zipfel C., Gust A.A., Brunner F. *Arabidopsis* receptor-like protein and receptor-like kinase suppressor of BIR1-1/EVERSHED mediate innate immunity to necrotrophic fungi. *The Plant Cell*, 2013, 25(10): 4227-4241 (doi: 10.1105/tpc.113.117010).
125. Zhang Y., Yang X., Liu Q., Qiu D., Zhang Y., Zeng H., Yuan J., Mao J. Purification of novel protein elicitor from *Botrytis cinerea* that induce disease resistance and drought tolerance in plants. *Microbiological Research*, 2010, 165(2): 142-151 (doi: 10.1016/j.micres.2009.03.004).
126. Peng D.-H., Qiu D.-W., Ruan L.-F., Zhou C.-F., Sun M. Protein elicitor PemG1 from *Magnaporthe grisea* induces systemic acquired resistance (SAR) in plants. *MPMI*, 2011, 24(10): 1239-1246 (doi: 10.1094/MPMI-01-11-0003).
127. Chen M., Zeng H., Qiu D., Guo L., Yang X., Shi H., Zhou T., Zhao J. Purification and characterization of a novel hypersensitive response-inducing elicitor from *Magnaporthe oryzae* that triggers defense response in rice. *PLoS ONE*, 2012, 7(5): e37654 (doi: 10.1371/journal.pone.0037654).
128. Kulye M., Liu H., Zhang Y., Zeng H., Yang X., Qiu D. Hrip1, a novel protein elicitor from necrotrophic fungus, *Alternaria tenuissima*, elicits cell death, expression of defence-related genes and systemic acquired resistance in tobacco. *Plant Cell Environ.*, 2012, 35(12): 2104-2120 (doi: 10.1111/j.1365-3040.2012.02539.x).
129. Liu W.P., Zeng H.M., Liu Y.F., Yuan J.J., Qiu D.W. Expression of *Alternaria tenuissima* PeaT2 gene in *Pichia pastoris* and its function. *Wei Sheng Wu Xue Bao*, 2007, 47(4): 593-597.
130. Mao J., Liu Q., Yang X., Long C., Zhao M., Zeng H., Liu H., Yuan J., Qiu D. Purification and expression of a protein elicitor from *Alternaria tenuissima* and elicitor-mediated defence responses in tobacco. *Annals of Applied Biology*, 2010, 156(3): 411-420 (doi: 10.1111/j.1744-7348.2010.00398.x).
131. Zhang W., Li H., Wang L., Xie S., Zhang Y., Kang R., Zhang M., Zhang P., Li Y., Hu Y., Wang M., Chen L., Yuan H., Ding S., Li H. A novel effector, CsSp1, from *Bipolaris sorokiniana*, is essential for colonization in wheat and is also involved in triggering host immunity. *Molecular Plant Pathology*, 2022, 23(2): 218-236 (doi: 10.1111/mpp.13155).
132. Wang J., Liu S., Ren P., Jia F., Kang F., Wang R., Xue R., Yan X., Huang L. A novel protein elicitor (PeSy1) from *Saccharothrix yanglingensis* induces plant resistance and interacts with a receptor-like cytoplasmic kinase in *Nicotiana benthamiana*. *Molecular Plant Pathology*, 2023, 24(5): 436-451 (doi: 10.1111/mpp.13312).
133. Tarallo M., McDougal R.L., Chen Z., Wang Y., Bradshaw R.E., Mesarich C.H. Characterization of two conserved cell death elicitor families from the Dothideomycete fungal pathogens *Dothistroma septosporum* and *Fulvia fulva* (syn. *Cladosporium fulvum*). *Front. Microbiol.*, 2022, 13: 964851 (doi: 10.3389/fmicb.2022.964851).
134. Xu Q., Hu S., Jin M., Xu Y., Jiang Q., Ma J., Zhang Y., Qi P., Chen G., Jiang Y., Zheng Y., Wei Y. The N-terminus of a *Fusarium graminearum*-secreted protein enhances broad-spectrum disease resistance in plants. *Molecular Plant Pathology*, 2022, 23(12): 1751-1764 (doi: 10.1111/mpp.13262).
135. Wang S., Yang S., Dai K., Zheng W., Zhang X., Yang B., Ye W., Zheng X., Wang Y.The effector Fg62 contributes to *Fusarium graminearum* virulence and induces plant cell death. *Phytopathol. Res.*, 2023, 5: 12 (doi: 10.1186/s42483-023-00167-z).
136. Janků M., Činčalová L., Luhová L., Lochman J., Petřivalský M. Biological effects of oomycetes

elicitins. *Plant Prot. Sci.*, 2020, 56(1): 1-8 (doi: 10.17221/21/2019-PPS).
137. Derevnina L., Dagdas Y.F., de la Concepcion J.C., Bialas A., Kellner R., Petre B., Domazakis E., Du J., Wu C. H., Lin X., Aguilera-Galvez C., Cruz-Mireles N., Vleeshouwers V.G., Kamoun S. Nine things to know about elicitins. *New Phytol.*, 2016, 212 (4): 888-895 (doi: 10.1111/nph.14137).
138. Noman A., Aqeel M., Irshad M.K., Qari S.H., Hashem M., Alamri S., AbdulMajeed A.M., Al-Sadi A.M. Elicitins as molecular weapons against pathogens: consolidated biotechnological strategy for enhancing plant growth. *Critical Reviews in Biotechnology*, 2020, 40(6): 821-832 (doi: 10.1080/07388551.2020.1779174).
139. Qutob D., Huitema E., Gijzen M., Kamoun S. Variation in structure and activity among elicitins from Phytophthora sojae. *Molecular Plant Pathology*, 2003, 4(2): 119-124 (doi: 10.1046/j.1364-3703.2003.00158.x).
140. Starý T., Satková P., Piterková J., Mieslerová B., Luhová L., Mikulík J., Kašparovský T., Petřivalský M., Lochman J. The elicitin β-cryptogein's activity in tomato is mediated by jasmonic acid and ethylene signalling pathways independently of elicitin-sterol interactions. *Planta*, 2018, 249: 739-749 (doi: 10.1007/s00425-018-3036-1).
141. Keller H., Bonnet P., Galiana E., Pruvot L., Friedrich L., Ryals J., Ricci P. Salicylic acid mediates elicitin-induced systemic acquired resistance, but not necrosis in tobacco. *MPMI*, 1996, 9: 696-703 (doi: 10.1094/MPMI-9-0696).
142. Kawamura Y., Hase S., Takenaka S., Kanayama Y., Yoshioka H., Kamoun S., Takahashi H. INF1 elicitin activates jasmonic acid- and ethylene-mediated signalling pathways and induces resistance to bacterial wilt disease in tomato. *Journal of Phytopathology*, 2009, 157: 287-297 (doi: 10.1111/j.1439-0434.2008.01489.x).
143. Brunner F., Rosahl S., Lee J., Rudd J.J., Geiler C., Kauppinen S., Rasmussen G., Scheel D., Nürnberger T. Pep-13 a plant defense inducing pathogen-associated pattern from *Phytophthora* transglutaminases. *EMBO J.*, 2002, 21(24): 6681-6688 (doi: 10.1093/emboj/cdf667).
144. Veit S., Worle J. M., Nurnberger T., Koch W., Seitz H.U. A novel protein elicitor (PaNie) from *Pythium aphanidermatum* induce duel defense responses in carrot and *Arabidopsis*. *Plant Physiology*, 2001, 127: 832-841.
145. Frías M., González M., González C., Brito N. A 25-residue peptide from *Botrytis cinerea* xylanase BcXyn11A elicits plant defenses. *Front. Plant Sci.*, 2019, 10: 474 (doi: 10.3389/fpls.2019.00474).
146. Mishura A.A., Sharma K., Misra R.S. Purification and characterization of elicitor protein from *Phytophthora colocasiae* and basic resistance in *Colocasia esculenta*. *Microbiological Research*, 2009, 64: 688-693 (doi: 10.1016/j.micres.2008.09.001).
147. Bar M., Sharfman M., Avni A. LeEix1 functions as a decoy receptor to attenuate LeEix2 signaling. *Plant Signaling & Behavior*, 2011, 6(3): 455-457 (doi: 10.4161/psb.6.3.14714).
148. Tundo S., Moscetti I., Faoro F., Lafond M., Giardina T., Favaron F., Sella L., D'Ovidio R. *Fusarium graminearum* produces different xylanases causing host cell death that is prevented by the xylanase inhibitors XIP-I and TAXI-III in wheat. *Plant Science*, 2015, 240: 161-169 (doi: 10.1016/j.plantsci.2015.09.002).
149. Noda J, Brito N, González C. The *Botrytis cinerea* xylanase Xyn11A contributes to virulence with its necrotizing activity, not with its catalytic activity. *BMC Plant Biol.*, 2010, 10: 38 (doi: 10.1186/1471-2229-10-38).
150. Ma Y., Han C., Chen J., Li H., He K., Liu A., Li D. Fungal cellulase is an elicitor but its enzymatic activity is not required for its elicitor activity. *Molecular Plant Pathology*, 2015, 16(1): 14-26 (doi: 10.1111/mpp.12156).
151. Shumilina D., Krämer R., Klocke E., Dzhavakhiya V. MF3 (peptidyl-prolyl cis/trans isomerase of FKBP type from *Pseudomonas fluorescens*) — an elicitor of non-specific plant resistance against pathogens. *Phytopathol. Pol.*, 2006, 41: 39-49.
152. Struwe W.B., Robinson C.V. Relating glycoprotein structural heterogeneity to function in sights from native mass spectrometry. *Current Opinion in Structural Biology*, 2019, 58: 241-248 (doi: 10.1016/j.sbi.2019.05.019).
153. Chen X.-L., Shi T., Yang J., Shi W., Gao X., Chen D., Xu X., Xu J.-R., Talbot N. J., Peng Y. L. N-glycosylation of effector proteins by an α-1,3-mannosyltransferase is required for the rice blast fungus to evade host innate immunity. *Plant Cell*, 2014, 26(3): 1360-1376 (doi: 10.1105/tpc.114.123588).
154. Takenaka S., Nakamura Y., Kono T., Sekiguchi H., Masunaka A., Takahashi H. Novel elicitin-like proteins isolated from the cell wall of the biocontrol agent *Pythium oligandrum* induce defence-related genes in sugar beet. *Molecular Plant Pathology*, 2006, 7(5): 325-339 (doi: 10.1111/j.1364-3703.2006.00340.x).
155. Gust A.A., Biswas R., Lenz H.D., Rauhut T., Ranf S., Kemmerling B., Götz F., Glawischnig E., Lee J., Felix G., Nürnberger T. Bacteria-derived peptidoglycans constitute pathogen-associated molecular patterns triggering innate immunity in *Arabidopsis*. *Journal of Biological Chemistry*, 2007, 282(44): 32338-32348 (doi: 10.1074/jbc.M704886200).
156. Jin Y., Zhao J.-H., Guo H.-S. Recent advances in understanding plant antiviral RNAi and viral suppressors of RNAi. *Curr. Opin. Virol.*, 2021, 46: 65-72 (doi: 10.1016/j.coviro.2020.12.001).
157. Wang K.D., Empleo R., Nguyen T.T., Moffett P., Sacco M.A. Elicitation of hypersensitive

responses in *Nicotiana glutinosa* by the suppressor of RNA silencing protein P0 from poleroviruses. *Molecular Plant Pathology*, 2015, 16(5): 435-448 (doi: 10.1111/mpp.12201).
158. Cai L., Dang M., Yang Y., Mei R., Li F., Tao X., Palukaitis P., Beckett R., Miller W.A., Gray S.M., Xu Y. Naturally occurring substitution of an amino acid in a plant virus gene-silencing suppressor enhances viral adaptation to increasing thermal stress. *PLoS Pathog.*, 2023, 19(4): e1011301 (doi: 10.1371/journal.ppat.1011301).
159. Pumplin N., Voinnet O. RNA silencing suppression by plant pathogens: defence, counter-defence and counter-counter-defence. *Nat. Rev. Microbiol.*, 2013, 11(11): 745-760 (doi: 10.1038/nrmicro3120).
160. Dewen Q., Yijie D., Yi Z., Shupeng L., Fachao S. Plant immunity inducer development and application. *MPMI*, 2017, 30(5): 355-360 (doi: 10.1094/MPMI-11-16-0231-CR).
161. Sokolov Yu. A. *Izvestiya Natsional'noy akademii nauk Belarusi. Seriya khimicheskikh nauk*, 2015, 4: 109-121 (in Russ.).
162. Dzhavakhia V., Filippov F., Skryabin K., Voinova T., Kouznetsova M., Shulga O., Shumilina D., Kromina K., Pridanniko M., Battchikova N., Korpela T. *Proteins inducing multiple resistance of plants to phytopathogens and pests. Intern. Pat. Classification: C07K 14/21. Intern. applic. number: PCT/FI2004/000766. Intern. Filing date: 17 December 2004 (17.12.2004). Priority data: 20031880 22 December 2003. Intern. Public. number: WO 2005/061533 A1*
163. Shen Y., Li J., Xiang J., Xiang J., Wang J., Yin K., Liu Q. Isolation and identification of a novel protein elicitor from a *Bacillus subtilis* strain BU412. *AMB Expr.*, 2019, 9: 117 (doi: 10.1186/s13568-019-0822-5).