"Influence of plant resistance inducers on post-transcriptional gene silencing (PTGS) process and its suppression during viral infection"

Plants have evolved a number of immune mechanisms during evolution, including physical (like the cell wall structures), and chemical, enabling them to fight pathogens (*e.g.,* fungi, bacteria, or viruses). Although plants do not have the immune system like animals, after all, they do not remain passive in the face of pathogenesis. The virus, entering the plant cell, releases its genetic material, which is a signal for the plant to start defending itself. The plant begins the synthesis of defense factors, such as salicylic acid or pathogenesis-related proteins (PR), which are aimed at weakening the virus and preventing the further spread of the infection. In addition, the plant activates the gene silencing process, the so-called post-transcriptional gene silencing (PTGS), which degrades foreign genetic material, including virus RNA.

The mechanism of PTGS consists of the production of short (about 21-24 nucleotide) small RNAs (sRNAs) using enzymes from the group of RNases (RNase III) (called DCL proteins (Dicer-like protein). Then sRNA is methylated and transported from the nucleus to cytoplasm, where connects with proteins Argonaute (AGO*)* forming a protein RISC (*RNA- induced silencing complex*). This complex recognizes small homologous fragments of viral genetic material and inhibits their further synthesis. A silencing signal is sent from the infection site to other plant tissues, preparing the plant organism to inhibit the spread of the viral pathogen.

However, viruses have developed their own mechanisms to protect them against the silencing process. They synthesize special factors called silencing suppressors that block PTGS at various stages. One of the best-known suppressors are cucumber mosaic virus (CMV) 2b protein and potato virus Y (PVY) HC-Pro protein.

To support plants in the fight against pests and pathogens, various commercially available pesticides are used. It is not always 100% effective, and their frequent use exerts significant evolutionary pressure on pathogens, which can cause the development of their highly resistant forms. Therefore, interest in the use of integrated plant protection and green chemistry (natural substances that the plant synthesizes (but in insufficient quantities) or synthetically created compounds, which are usually their analogues or polymers) increased. These compounds include resistance inducers (RI) such as benzothiadiazoles (BTH, salicylic acid analogue) and chitosan (CHT, β-1,4-glucose polymer). Plants treatment with these compounds leads to activation of SAR (Systemic Acquired Resistance), through the synthesis of many defense-related proteins or phytohormones (plant hormones), even before a pathogen appears in the environment.

Our previous studies have shown a positive effect of the use of RI on the plant's response to subsequent viral infection. A reduced level of viral RNA accumulation was found in tobacco plants (*Nicotiana tabacum)* treated with BTH. In our earlier transcriptome studies of the tomato (*Solanum lycopersicum*), we noted that plants treatment with BTH caused an increased expression of genes associated with the PTGS like mentioned before the gene that encodes the DCL protein and a significant increase in the level of synthesis of abscisic acid ABA, whose indirect influence on the gene silencing process has also been described. These results drew our attention and encouraged us to take a closer look at the role of RI in the process of post-transcriptional gene silencing in plants.

In our research, we want to check the effect of using two types of RIs: BTH and CHT on tobacco plants (*Nicotiana benthamiana*), using wild-type plants and previously prepared selected mutants (with deletion of *DCL* genes) in which PTGS is significantly impaired. In addition, it is assumed to use two viruses with well-known and very potent PTGS suppressors (CMV and PVY). We expect that we explain how RIs act on the PTGS process and viral suppressors of PTGS. The results obtained from the presented research project will deepen the knowledge in the field of plant resistance induced by RIs and also will influence the development of such disciplines as plant pathology, plant protection, and virology.

A better understanding of mechanisms of RIs action can contribute to the inclusion of resistance inducers for agricultural practice in the future and help to reduce the use of aggressive chemistry, improving plant resistance and reducing crop losses.