TRANSGENIC POTATO PLANTS WITH GENES FOR SYNTHESIS OF CECROPIN-MELLITIN ANTIMICROBIAL PEPTIDES

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The objective is to obtain the lines of transgenic potato plants with enhanced resistance to phytopathogenic organisms due to the expression of cecropin-mellitin antimicrobial peptides on the basis of Belarusian potato varieties.

Cecropin-mellitin antimicrobial peptides are recombinant molecules constructed on the basis of cecropin A of the moth *Hyalophora cecropia* and mellitin of the honeybee *Apis mellifera*.

In this work two hybrids of cecropin-mellitin antimicrobial peptides have been used: MsrA1 and CEMA (MBI 28).

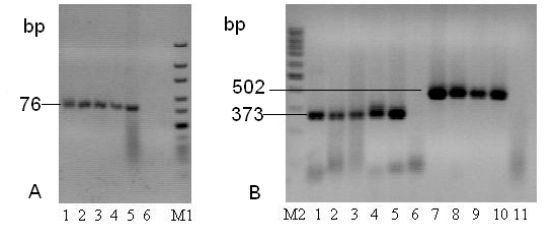
Genes encoding these antimicrobial peptides were introduced into the cells of potato plants as a part of the binary vector systems, based on the pBIΔGUS1-MsrA1 and pBIΔGUS1-CEMA plasmids (T. Gapeeva et al., 2008).

CEMA: *Met- Lys- Trp- Lys- Leu- Phe- Lys- Lys- Ile- Gly- Ile- Gly- Ala-Val- Leu- Val- Leu- Thr- Thr- Gly- Leu- Pro- Ala- Leu- Lys- Leu- Thr- Lys*

MsrA1: *Met- Ala- Leu- Glu- His- Met- Lys- Trp- Lys- Leu- Phe- Lys- Lys- Ile- Gly- Ile- Gly- Ala- Val- Leu- Val- Leu- Thr- Thr- Gly- Leu- Pro- Ala- Leu- Lys- Leu- Thr- Lys*

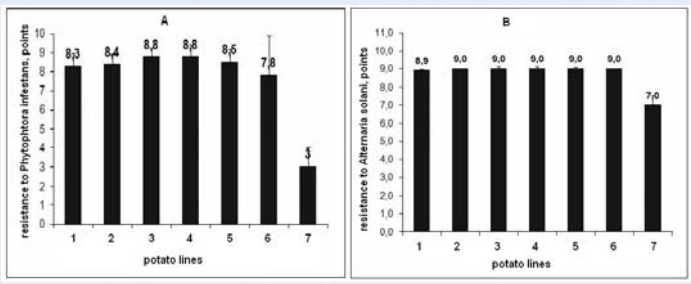
The sequqnces of MsrA1 and CEMA (M. Osusky, 2000)

The selection of regenerated plants was conducted on a medium containing kanamycin at a concentration of 25 mg/l. Analysis of the resulting plant transformation was carried out by RNA-PCR and showed the presence of mRNA transcripts of marker and target genes in 80% of the transformed plants.



RNA-PCR analysis of potato plants after transformation:
A – PCR with primers for MsrA1gene;
B – PCR with primers for NPT II (373 bp) and potato actin (502 bp) genes.

Transgenic potato plants with MsrA1 and CEMA genes were subjected to testing in vitro using fungal pathogens ***Phytophtora infestans, Alternaria solani***



A – resistance to ***Phytophtora infestans***,
B – resistance to ***Alternaria solani***:
1, 2, 3, 4, 5, 6 – potato lines with expression of MsrA1gene;
7 – control line (nontransgenic).
Values are means ± SE for 10 plants.

Estimation of resistance to ***Phytophtora infestans*** of the transgenic potato plants with antimicrobial peptides MsrA1 и CEMA genes:

A – transgenic potato with CEMA gene (very high resistance to the pathogen, almost no damages of leaves);
B, D – transgenic potato with MsrA1 gene (high resistance to the pathogen, minor damage of leaves);
C, E – control potato (significant damages).

CONCLUSION: Gene expression of antimicrobial cecropin-melittin peptides MsrA1 and CEMA in the cells of transgenic potato plants of Belarusian varieties is accompanied by increased resistance to fungal infection (***Phytophtora infestans, Alternaria solani***).