# Stress Caused by Plant Virus Infection in Presence of Heavy Metals

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## Abstract

Due to increased heavy metal content in Ukrainian soils, purpose of the work was to study relations between presence of heavy metals in soil and their effect on development of phytoviral infection. Experiments were conducted in *Nicotiana tabacum – Potato virus X* model system. Soluble salts of Cu, Zn and Pb were deposited in soil separately at the limiting concentrations simultaneously with virus infection of plants. Infected plants grown on usual soil showed symptoms of disease on 16 dpi as well as plants grown on soil with metals deposited. Contrary, combined effect of heavy metals and virus infection caused an increase of chlorophyll content comparing with control plants, therefore effect of heavy metals partially compensated the effect of virus infection on experimental plants.

Keywords: *Potato virus X* (PVX); *Nicotiana tabacum* cv. Trapeson; heavy metals; maximum permissible concentration; chlorophyll; photosynthesis; stress

# **INTRODUCTION**

Due to intensive industry development, increased heavy metal content is clearly observed in environment nowadays. Therefore, agrocenoses are seriously affected by stresses both of biotic and abiotic nature. Increase of heavy metal content leads to various physiological changes of plants, the most common from them could be membrane damages, changes in enzyme activity, inhibition of roots' growth, etc. (Foy et al. 1978; BARCELO & POSCHENRIEDER 1990). Virus infection can be an inductor of biotic stresses in plants. Owing to serious pollution of Ukrainian soils with heavy metal compounds, the point of these experiments was to reveal a separate effect of every single heavy metal tested on the development of plant virus infection, and to show possible changes in chlorophyll concentration that actually reflect photosynthetic activity and condition of a plant in general.

# MATERIALS AND METHODS

Experiments were conducted in *Potato virus* X – *Nicotiana tabacum* cv. Trapeson plants model system. Copper, lead and zinc were deposited in pot soil in

form of soluble salts (CuSO<sub>4</sub> × 5H<sub>2</sub>O, ZnSO<sub>4</sub>, PbNO<sub>3</sub>) (KABBATA-PENDIAS & PENDIAS 1986) in concentration corresponding to the estimated level of maximum permissible concentrations (MPC) determined for these metals in Ukrainian soils (KOSTYSHYN *et al.* 1995). Plants were mechanically inoculated with PVX in concentration of 50  $\mu$ g/ml simultaneously with metal deposit. Virus concentration in plant extracts has being determined every week during the experiment by indirect ELISA with polyclonal rabbit antiserum to PVX, as described in CROWTHER (1995). Chlorophyll concentration has being measured by Arnon's method (WELLBURN 1994) simultaneously with determining of virus content in plants.

#### RESULTS

Results of the experiments showed that symptoms on virus-infected plants have been developed by 16 day post infection (dpi) independently from depositing (or not depositing) of heavy metal salts in soil. There were typical PVX symptoms of mild mosaic with following black ringspots on tobacco plants. Analysis of chlorophyll contents dynamics demonstrated that, in the absence of heavy metals, virus-infected plants



Figure 1. Total chlorophyll content in virus-infected plants



Figure 3. Total chlorophyll content in virus-infected plants grown in soil polluted by heavy metals in MPC

accumulated less total chlorophylls comparing to intact non-infected plants (Figure 1) that correlated with gradual increase of virus concentration.

As it was revealed (ULYNETS *et al.* 2001), inhibition of photosynthetic activity is shown to be a characteristic distinguishing feature of systemic virus infection at the level of plant organism. Contrary, in the absence of virus infection, plants growing in soil with deposited heavy metal had higher chlorophyll content in comparison with healthy plants w/o metal added (Figure 2). Moreover, all infected plants growing in deposited soil demonstrated higher content of chlorophylls comparing to even intact ones (no virus, no metal) (Figure 3). This means that, surprisingly, combined effect of heavy metal together with virus infection caused an increase of chlorophyll concentration in experimental plants.



Figure 2. Total chlorophyll content in plants grown in soil polluted by heavy metals in MPC

# DISCUSSION

It is well known that zinc and copper are the components of plant enzymes and participate in the processes of photosynthesis. Hence, their presence in soil in permanently high concentrations lower than MPC is normal (KABBATA-PENDIAS & PENDIAS 1986). Deposit of separate metals in soil leads to the increase of total chlorophyll content in the absence of viral infection (Figure 2), proving an intensive uptake of microelements by young plants during their development. The possible explanation lays in possible compensation of microelements shortage which is due to the active plant growth. Also, its demonstrated that zinc and cobalt are included in complex compounds increasing plant resistance to viral infections. Therefore, in low doses these metals may stimulate plant defence responses (RAUSER & DUMBROFF 1981).

Therefore, presence of separate heavy metals in concentration close to MPC partially compensated the impact of virus infection on plants. This may lead to the conclusion that a slight increase of some heavy metal content in soil may grade in part the influence of the biotic stress caused by plant virus infection.

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