

## SHORT COMMUNICATION

### VIRUS DISEASES IN COLLECTION OF GENETIC RESOURCES OF GARLIC IN THE CZECH REPUBLIC

#### VIROVÉ CHOROBY U KOLEKCE GENETICKÝCH ZDROJŮ ČESNEKU V ČESKÉ REPUBLICE

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The first results of detailed testing of the garlic genetic resources collection in Olomouc by ELISA showed a huge occurrence of all four viruses (OYDV, LYSV, GCLV, SLV) – from 104 tested genotypes only one, was found to be not infected. 50 genotypes (at least one

plant from 10 tested plants) were, on the other hand found to be infected by all four viruses. These results are very unfavourable and it can't be expected either, that the rest of the collection will be better.

Key words: garlic, genetic resources, virus diseases, OYDV, LYSV, GCLV, SLV, restoration to health

In the Czech Republic there is probably the largest collection of long day, vegetatively propagated garlic (*Allium sativum* L.) in the world. In the collection there are currently 623 accessions which present genotypes from different countries and continents. Unfortunately, the health condition of the collection appeared to have been not very good and therefore the estimation of the current state of health of garlic in the collection at the Olomouc department of Crop Research Institute was evaluated. All the collection (10 plants per genotype) was tested for the presence of four virus diseases (OYDV, LYSV, GCLV, SLV) using the ELISA method. The aim of this paper was to present the first results of this testing.

#### MATERIAL AND METHODS

The testing of the garlic collection for occurrence of virus diseases took place between 5<sup>th</sup> April and 17<sup>th</sup> May 2007. This time was set up by developing phase of plants (plants with at least 3 leaves) and climatic condition in Olomouc (when the outside temperature exceeded 28°C some viruses are getting undetectable by ELISA method due to its thermolability).

Accessions in the garlic collection in Olomouc are consisted of by 19 plants and to eliminate possible methodical mistakes 10 plants of this set were analysed for virus occurrence in each accession. This amount, which represented more than 50% of accession, was stated be-

cause the concentration of viruses is influenced by the development phase of plants as well as in particular morphological parts of plants and current climatic conditions and in the case of low concentration of virus in plant sample the ELISA method showed a negative reaction with antibodies and plant is determined as virus-free although it was already infected by virus.

Fresh samples of garlic leaves (about 10 g) were used for testing. It was crushed together with extraction buffer (Homogenizer hand model; Bioreba) in plastic bags and then the occurrence of onion yellow dwarf potyvirus (OYDV), leek yellow stripe potyvirus (LYSV), garlic common latent carlavirus (GCLV) and shallot latent carlavirus (SLV) was detected by Bioreba reagent sets and common chemicals according to the manufacturer's instructions. The colour reaction was analysed by ELISA reader (BIO-TEK EL800-PC) in wave length  $\lambda = 405$  nm. Each sample was analysed only in one repetition (one cup) at the ELISA plate and it was evaluated as positive when spectroscopy result of absorbance exceed 0.1.

Up to now 104 genotypes (1040 plants) from Olomouc garlic collection have been tested. This set was chosen randomly, but it represents all three garlic morphological types presents in the collection (garlic produces the scape with topset, garlic without scape and group garlic - semi-bolters) and also different geographical origin of accessions. The accessions were evaluated as "fully infected" by particular virus (10 plants from 10 tested plants showed positive

reaction), "partly infected" (at least 1 plant from 10 tested plants showed positive reaction) and "no-infected" (no any plant from 10 tested plants showed positive reaction). This evaluation was made separately for each of the 4 tested virus diseases in each accession. An important notice is, that the result "no-infected" really does not mean that the plant is virus-free. It is easily possible, that the plant was already infected by a virus but that the concentration of virus was in the plant tissue during testing time was too low for its detection by the ELISA method – it means that the virus concentration was under the detection limit of the used method.

## RESULTS AND DISCUSSION

The first results of testing of the garlic genetic resources collection in Olomouc by ELISA showed a huge occurrence of all four viruses (OYDV, LYSV, GCLV, SLV) – from 104 tested genotypes from which only one was found as "no-infected" by any of the testing viruses. On the other hand 26 accessions were "fully infected" by all four viruses and another 24 genotypes were by all four viruses infected at least "partly". 33 accessions were "fully" or "partly infected" by three viruses and 18 accessions were "fully" or "partly infected" by two viruses in different combinations. Another overview about a degree of virus infection within garlic genetic resources in Olomouc is present

T a b l e 1

The first results of testing of garlic genetic resources collection in Olomouc  
(104 tested accessions = 1040 tested plants)

Degree of virus infection	OYDV	LYSV	GCLV	SLV
„no-infected“ accessions (0 plants from 10 tested ones)	16	39	6	18
„partly infected“ accessions (1–9 plants from 10 tested ones)	16	23	6	23
„fully infected“ accessions (10 plants from 10 tested ones)	72	42	92	63
% infected plants (from 1040 plants)	76.7	50.5	89.7	69.0
% infected accessions (min. 1 plant/accession; from 104 accessions)	83.2	72.9	97.4	83.2

OYDV – onion yellow dwarf potyvirus  
 LYSV – leek yellow stripe potyvirus  
 GCLV – garlic common latent carlavirus  
 SLV – shallot latent carlavirus

in Table 1. Only two genotypes showed a positive reaction on only one virus disease – it was accession ECN 09H0100072 – cv. P-61, origin: Czechoslovakia (infected by OYDV) and ECN 09H0100236 - landrace (Medea 3), origin: Algeria (infected by GCLV). The only one “no-infected” accession was found ECN 09H0100786 – landrace (Kalonda 1) originally from the Slovak Republic.

The first reference about the occurrence of viruses in the garlic genetic resources collection in Olomouc was presented by Klukáčková et al. (2004). During a four year survey they analysed 57 randomly chosen genotypes (only one plant per accession) and in accordance with our results they discovered that most of these genotypes are affected by a complexity of viruses. They found a mixed infection of four ELISA tested viruses in 18% of samples and infection with a complexity of three viruses was noticed in 16% of samples. In detail 95% of tested plants were positive with OYDV, 61% with LYSV, 93% with GCLV and 49% with SLV. These scores were a starting reason for a detailed study of virus occurrence in the collection.

Our monitoring of virus occurrence in the Olomouc garlic collection only confirms these results and only less occurrence of OYDV within the accessions (83.2%) compared to the previous testing (95%) is surprising, because actually even a higher degree of infection with all four viruses was expected. However, this difference can be easily explained by the non identical set of analysed accessions and it could be changed in next set of samples.

## CONCLUSIONS

A detailed testing of garlic genetic resources collection in Olomouc by ELISA confirmed a huge occurrence of viruses OYDV, LYSV, GCLV and SLV.

Most of the tested genotypes are affected by a complexity of these viruses.

The highest number of accessions is affected by GCLV (97.4%) and the lowest by LYSV (72.9%).

Only one of the 104 tested accessions was found as “no-infected” by any of the testing viruses but in spite of this, it is not possible to say that the plants are virus-free. It is easily possible, that the virus concentration was just under the detection limit of the used method.

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## SOUHRN

V České republice je soustředěna pravděpodobně největší kolekce dlouhodobního vegetativně množeného česneku na světě. V kolekci je v současné době 623 položek, které reprezentují genotypy z různých zemí a světadílů. Zdravotní stav olomoucké kolekce VÚRV, v.v.i. vykazoval jisté problémy, a proto v současné době probíhá jeho testování. Celá kolekce (10 rostlin z každého genotypu) je metodou ELISA testována na přítomnost čtyř virových chorob (OYDV, LYSV, GCLV, SLV) a již první výsledky ukazují vysoký výskyt všech těchto virů. Z prvních 104 testovaných genotypů byl nalezen pouze jeden, který nevykázal žádnou přítomnost virů. Na druhé straně 50 položek (nejméně 1 rostlina z 10 testovaných) bylo infikováno kompletní sadou všech 4 sledovaných virů. Lepší výsledky se nedají očekávat ani u dalších genotypů v kolekci.

Klíčová slova: česnek, genetické zdroje, virové choroby, OYDV, LYSV, GCLV, SLV, ozdravení