BIOLOGICAL CONTROL OF GRAPE CROWN GALL

METODĂ BIOLOGICĂ DE COMBATERE A CANCERULUI BACTERIAN LA VIŢA DE VIE

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Abstract. *Genetic date transfer T-DNA from a pathogen to a plant cell occure the process of tumefaction on vineyard. Treatment of woundings by biologic preparate Paurine (suspension of bacterials cells of Pseudomonas fluorescens) deteriorate the interaction of bacteria-pathogen with the cell of host plant. Application of this preparat decrease the quantity of plants with tumors.*
Key words: crown gall, biological control, grape

Rezumat. *Cercetările se referă la transmiterea informaţiei genetice de la patogen către celula vegetală exprimă procesul de formare a tumorilor la viţa de vie. Prelucrarea leziunilor vegetale cu biopreparatul Paurin (suspenzia de celule a bacteriei Pseudomonas fluorescens) preintimpină penetrarea acestei bacterii in celula vegetală a plantei. Aplicarea preparatului reduce numărul de viţe altoite şi tufe contaminate cu tumori.*
Cuvinte cheie: cancer bacterian, control biologic, viţa de vie

INTRODUCTION

Among the chronic diseases of perennial plants crown gall is a very dangerous ones, which caused considerable waste of productions in the industrial vineyards of the Republic of Moldova. First of all the danger is determined by the situation that pathogenic agent could survived a lot of time in vascular plant system in latent form, without provocation of any tumors on the aerial grape plant surfaces. Crown gall pathogen agent is represented by pathogenic species *Agrobacterium*- bacteria bacilare gramm negativ - *A. tumefaciens* which penetrate in whole vascular system of plants throw freeze induced wounds, periderm deteriorations on new developed roots, mechanical lesions of stem ramification as well as on the trunk. It is preserved there for long time in latent form, without tumors provocation. Grape initiation of tumorogenesis coincide with the period of „bleeding" , during the spring running, when it is favourable penetration of enormous quantities of pathogenic bacteria throw new wounds. As a result of complex biological processes with participation of bacteria land plant cell enzymes a portion of bacterial gene- plasmid T i is incorporated in chromosome of vegetal cell, which starts cell division according the ways of tumorous proliferation. Once being contaminated, grapes vine plants began chronic manifestation of diseases. Tumors represent a consequence of contamination, and as a rule are formated after 1-3 years after theirs establishments on plants at permanent place (Chernin L.S., 1997; ***, 1990).

There is no exist resistant to crown gall grapes vine varieties. This disease is

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spread all over the zones of grape cultivation in the Republic of Moldova. Maximal cancerogenesis intensity there are presented on the sites and microareas characterized by cold winter, freeze injures, as well as in the cases of provocation of additional freezes, induced wounds, graft unions, mechanically produced wounds.

According to our long-term observations on the development of infected plants in the vineyards every years 6% of plants became infected, in the same time varieties Cabernet et Merlot -9,2%. In the nursery 10-20% of plants are already infected, and developments of shoots is reduced at the level of 36%. Therefore 30 plants for one thousand could not be standardized. Depending on type of soil during 10 years plantation 8-19% of plants are lost (Lemanova N.B., 1991). Members of the *Agrobacterium* genus naturally have the ability to transfer a segment of DNA from a plasmid hosted by the bacterium into the genome of a cell of a living plant. The DNA transferred into the plant (the T-DNA) causes the plant cells to initiate two activities. One activity is to manufacture a class of chemicals, called opines, which can be metabolized by the bacteria as a food source. The other activity is to initiate the growth of a tumorous mass referred to as a crown gall. It is not necessary to create a competition relationships between bacterial disease and antagonists in the first step of pathogenesis. In this case *Agrobacterium* could not be attached to walls of plant cells. For this scope it is indispensable to obtain lesions of another bacteria, which is responsible for introduction of pathogen agent. In this situation it is impossible to transfer parasite T-ADN transferation into mother cell.

Rizoferial bacteria from genus *Pseudomonas* are the most important for production of biologically active compounds – bacteriocine, antibiotics, enzymes. That is an explanation referring theirs utilization like antagonists for ecological production of microorganisms with biological origin.

MATERIAL AND METHOD

The researches were carried out in different representative sites of moldavian vineyards within the Experimental Station of Research Institute for Horticulture and Alimentary Technologies, Chisinau. Experiences were consecrated to application of preparation Paurin for biological control of grape crown gall. „Paurin" (***, 1990) represent semitransparent liquid, color of milk, mat, with specific smell, non-significant sediment, which disappear on the process of agitation. Ingredients of preparation: suspension of live cells of *Pseudomonas fluorescens*, cultural media within metabolic compounds of bioagent. Content of 1mL of preparation - 10mld bacterian cells. It is necessary to agitate 3-5 minutes diluted preparation before treatment. In case of the production of grafted material plants (scion and rootstock) are treated before paraffination and stratification during 10 sec. using the work solution of 300 ml of Paurin preparate and 100ml water. Before plantation in field duration of treatment of grafted plants is 30 min. Irrigation of planted material should contain 2L of Paurin in 1 t of water.

Obtained mature plants for industrial plantations should be treated with Paurin during 2 hours (proportion: 3l of Paurin and 1t. of water). Treatment in established new plantations annualy in the period of intensive circulation of liquids in shoots: 3 L/1ha and 1liter of Paurin to 100 l water for treatment of individual plants. In the vineyards were are affected trunks and principal shoots of plants it is used Paurin in the proportion 5L to 1 ton of water two times during the vegetation.

RESULTS AND DISCUSSIONS

The importance of the method for controlling crown gall disease in grapes vine (*Vitis vinifera*) plants using an effective quantity of Paurin occurs from table 1.

Table 1

Effects/Influence of grapes plant treatment with Paurin variety Pinot Franc (after sever winter of 2009-2010)

Spring tilling	Number of planting places	Autumn 2010					
		Killed trunks		Trunks with tumors		Healthy trunks	
		nr	%	nr	%	nr	%
2008-2009 years	88	9	10	2	2,3	79	90,9
2009	88	25	26,4	4	6,4	63	71,6
Without treatment	88	35	39,7	24	45,3	53	60,2

Our investigations and experimentations demonstrate that utilization of Paurin in the production of grafted and non grafted vegetative material in the Republic of Moldova, Krasnodar region of Russia, Herson region of Ukraine reduced percentage of affected plants of varieties Leana, Moldova, Dekabriskii, Strashemskiii, Vostorg from1to 5% comparatively with control (non treatment) – 31-86% (fig. 1). Treatment of infected variety Cabernet, grafted on rootstocks S x B-41 B and Kober 5BB before stratification contributed to increase production of good vegetative material in the first field around 56%, comparatively with 42% in control. Biopreparation Paurin have been utilization for replantation of six grapes varieties in the period of vegetation within 900 thousand grafted plants in nursery and in greenhouse. Experimentally are established effectiveness of Paurin in the process of irrigation of plant on containers before plantation and for rooted plants in greenhouse and in open field. Plantation established in 1990-1994 using pretreatment with Paurin actually are economically efficient, without compromise from diseases.

On the basis of bacterial metabolic products in Australia (A. Kerr), USA (Xi-o Pu, Gudman C.) and China (Sule S., 2005; Chen F. et al., 2007; Burr T., 2004) there are created analogical biofungicides for prevention of bacterial tumors development. In Western Europe (Bazzi C., Bini F., 2005; Chen F. et al., 2007) because of mild climate manifestation of tumors are reduced, respectively there are no used treatment in the first stage of plant production. In the same time a special attention there are provied to selection of healthy material for vegetative reproduction (scion and rootstock).

Likewise Paurin is efficient for stopped tumorous proliferation in vineyards. Our investigation demonstrated, that early spring Paurin treatment of experimental vineyards of our Institute before the bleeding vines consecutively 2 years (2008-2009) conducted to reduction of intension of tumor formation (tab.1) in the conditions of cold, air temperature -26^0C. For this consideration variety are very important. The incidence of crown gal land its importance in local moldavian vineyards appears to be on the increase. Especially because the enlargement of varietal conveeier, climate change etc.

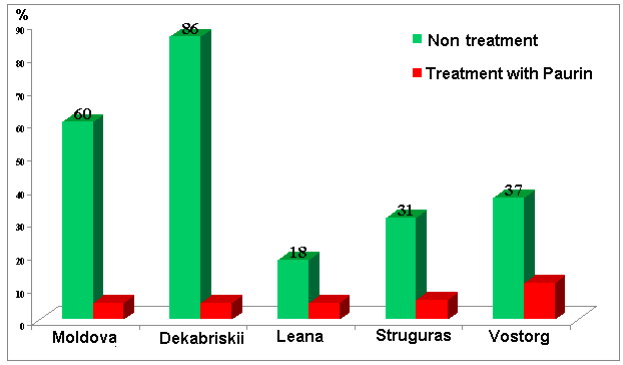
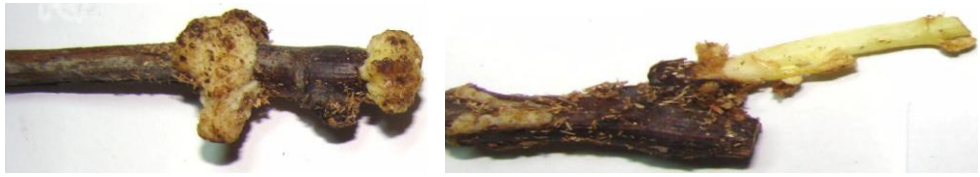


Fig. 1 –Susceptibility of grapes varieties to crown gall and efficiency of Paurin utilization



a) **b)**

Fig. 2 – Tumors and necrosis caused by *Agrobacterium vitis*: **a** – non treatment; **b** – treatment with "Paurin"

CONCLUSIONS

1. Early spring infected grapes plants treatment using biologic preparate "Paurin" provoke effects of inhibition of tumorous development, have an anti-stress effect for whole plants, stopped grows of tumorous within the injuries.

2. In the scope of reducing the occurrence of crown gall it is necessary to plant winter hardy grape vine varieties.

3. Cultural control strategies include mounding soil around grape vine plants with multiple trunks.

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