The loci of potato wart agent in Ukraine are surveyed, and their identification is carried out with the help of potato differentiating test-varieties of Ukrainian selection. In v. Beregomet, Vyzhnytsya district, Chernivtsi region, a common pathotype is identified — Д1; in Transcarpathian region, the aggressive pathotypes are identified: in v. Maydan, Mizhgirsky district -11 — Mizhgirsky aggressive pathotype; in v. Rahiv – 13 — Rahivsky aggressive pathotype; in v. Yasynya – 18 — Yasynivsky aggressive pathotype; in v. Bystrets, Verhovinian district, Ivano-Frankivsk region – 22 — Bystrets aggressive pathotype.

Potato wart Synchytrium endobioticum (Schilb.) Perc., pathotypes, identification, differentiation, potato test assortment.

Potato wart is a quarantine disease distributed in 55 countries of the world, which causes a significant damage to potato production and is capable of reducing yield by 80 – 90 %, particularly in crofts. The potato wart problem complication is due not only to the fact that the disease agent is difficult to root out of soil because of its zoosporangia high resistance to unfavorable environmental conditions, but also to its capability of changing its parasitic peculiarities in conditions favorable for such a process, under the natural phenomena — mutations, adaptations, hybridizations, etc. Such conditions exist in mountainous districts of Western regions of Ukraine. Here, the highest density of wart loci and its aggressive forms is observed. The favorable conditions influence the disease development and, together with this, are one of the reasons for the fungus forms changeability and new pathotypes formation. Such a phenomenon is observed at potato monoculture, especially at growing a mixture of varieties which differ by their wart-resistance [1].

A variety specialization is expressed on potato wart agent, related to a host plant – potato. According to the data from EPPO (European and Mediterranean Plant Protection Organization – 1982), in Europe, 20 pathotypes of the fungus are identified; the appearance of new ones which differ from the distributed common pathotype D1 is noted in Germany, Czechia, Slovakia, Peru, Canada (isl. Newfoundland), India, Italy [2].


By last years research [6] in Transcarpathian region, we have revealed the agent of potato wart in Velyko-Bereznyansky, Mizhgirsky, Rahivsky, Perechynsky, and Svalyavsky districts.

In Lviv region, the agent is detected in Turkivsky, Skolevsky, Striysky districts [7].

In Chernivtsi region, the disease agent is detected only in two districts: Vyzhnytsky and Putyvsky [8].
The wart agent aggressive pathotypes distributed in Mountainous-Carpathian zone of Ukraine, are able to infest potato assortment which is resistant to the common pathotype [3]. The data absence about pathotype belonging of this disease agent in Ukraine makes its spread prevention measures complicated to apply.

That is why the loci establishment and mapping, their infectious capacity on the territory of Ukraine is a necessary and important in modern condition for phytosanitary security. For establishing aggressive pathotypes, a differentiating potato test-assortment is investigated and present at UkrSRPQS PPI NAAS, which has been completed with Ukrainian selection varieties in recent years [4].

The wart agent pathotypes identification is also perspective, by developed and patented biochemical means, which allows in a short time to establish an isolate in a detected locus belonging to investigated and registered pathotypes in the world [5].

For such a disease spread prevention, one should conduct the consignment arrival surveillance at customs clearance of products under quarantine, as well as the infectious capacity analysis of the detected wart loci, and elaborate the disease agent control measures.

**Research aim** – checking the detected potato wart loci in border- and mountain zones of Ukraine, and conducting their identification.

**Research materials and methods.** For researches in 2013, the soil samples from wart loci were selected in v. Beregomet, Vyzhnytsya district, Chernivtsy region; v. Maidan, Mizhgirsky district, v. Surupy, v. Yasinya, Rahivsky district, Transcarpathian region, v. Bystrets, Verhovynsky district, Ivano-Frankivsk region. Soil samples for potato wart agent zoosporangia detection are selected by a standard envelope method (5 pits): according to SStU (State Standard of Ukraine) 3355-96 [9].

![Scheme 1. Soil samples selection (SStU 3355-96) for potato wart agent zoosporangia detection](image)

The zoosporangia isolation was performed by our self-developed flotation method, in 48,5% sodium iodide solution (Patent of Ukraine for invention №17049 from 15.09.2006 “Mode of potato wart agent *Synchytrium endobioticum* (Schilb.) Perc. zoosporangia isolation” [10].

Soil samples were rubbed with rubber pounder, weighed 1g, sifted through 3 sieves 3 with diameter 0,5; 0,25 and 0,03mm. Before the last sieve, the soil sample was treated with ether, for dissoluting organic matters, and passed to centrifugal vials where 35% sodium iodide with specific gravity 1,1 was added, and centrifugated for 3 minutes at 3000 revolvings per minute. At this, a light dash rised to the surface. A supernatant with dash was poured away, and the sodium iodide solution
was added to precipitation, but 48.5%, with specific gravity 1.4, and again centrifugated for 3 minutes at 3000 revolvings per minute. At this, the zoosporangia emerged to the surface; after this, they were collected onto a sentinel glass and counted under microscope (8 x 15). The zoosporangia viability determination was carried out by staining them with 0.5% Coomassie blue G-250 solution, which stains the living zoospores in blue, and the dead ones remain unstained [11]. They were counted under microscope. Potato wart agent *Synchytrium endobioticum* (*Schilb*) Perc. zoosporangia species identification was carried out.

Potato differentiating varieties contamination with wart pathotypes was conducted in laboratory conditions by methods of Glynne A. [12] and Speickermann A. [13], improved by research workers of our research station [14].

For the research, potato differentiating varieties were used: 1 group – potato varieties which are infested with all the pathotypes of wart agent: Poliska pink; 2 group – potato varieties which are infested with only aggressive pathotypes: Slovyanka, Pirovska, Legenda; 3 group – potato varieties which are differentially infested with wart agent pathotypes: Kalynivska – infested only with the 11 (Mizhgirsky aggressive pathotype); Malynska white – infested with the 13 (Rahivsky aggressive pathotype); Shedryk – infested with the 18 (Yasinivsky aggressive pathotype), and Divo. Chervona Ruta – infested with the 22 (Bystretsksky aggressive pathotype); 4 group – potato varieties which are not infested with any wart agent pathotype in Ukraine: Bozhedar, Glazurna [15].

For conducting potato wart agent pathotypes differentiation, potato differentiating test-varieties were contaminated with wart agent summer zoospores of all the 5 pathotypes in laboratory conditions. For this, a paper ring was glued onto a potato tuber apex around the shoot part, with the help of paraphine and vaseline mixture (1:1). Into the ring, distilled water was poured, and inoculation was carried out with summer zoospores from fresh wart swells, of a common and 4 aggressive pathotypes, sized 0.5cm³. The inoculated samples were placed in a climatic chamber with increased humidity and temperature 11-13 °C, up to the disease symptoms.

### Scheme 2. Laboratory trials set on wart agent pathotypes differentiation and identification (1 – 11– potato test-varieties).

1 – Poliska pink; 2 – Slovyanka; 3 – Pirovska; 4 - Legenda; 5 – Kalynivska; 6 – Malynska white; 7 – Shedryk; 8 – Chervona Ruta; 9 - Divo; 10 – Bozhedar; 11- Glazurna.
emergence [8]. The contamination was performed simultaneously with zoospores of a common (Dalem), 11 — Mizhgirsky, 13 — Rahivsky, 18 — Yasinivsky, 22 — Bystretsky wart pathotypes.

![Potato samples inoculation with zoospores from fresh wart swells](image)

**Fig. 1. Potato samples inoculation with zoospores from fresh wart swells**

**Research results.** As a result of checking the infectious capacity of potato wart agent loci (common and aggressive pathotypes), from 54 to 72 viable zoosporangia per 1g of soil were detected (Table 1)

**1 – Infectious capacity of a common and aggressive wart pathotypes loci in Ukraine (2013)**

| Pathotypes                                                                 | Viable zoosporangia detected |  
|                                                                           | NaI 48,5% | 0,5% Coomassi blue G-250 |
|                                                                           | Quantity | Quantity | Effectiveness, % |
| 11 – Mizhgirsky aggressive (v. Maydan, Mizhgirsky district, Transcarpathian region) | 54,6 ± 0,6 | 58,3 ± 0,9 | 9,3 |
| 13 – Rahivsky aggressive (v. Surupy, Rahivsky district, Transcarpathian region) | 58,3 ± 0,3 | 68,3 ± 0,6 | 15,2 |
| 18 – Yasinivsky aggressive (v. Yasinya, Rahivsky district, Transcarpathian region) | 68,3 ± 0,6 | 76,6 ± 0,6 | 12,6 |
| 22 – Bystretsky aggressive (v. Bystrets, Verhovinian district, Ivano-Frankivsk region) | 54,6 ± 0,9 | 56,0 ± 0,6 | 2,3 |
| Дi – common (v. Beregomet, Vyzhnytsya district, Chernivtsy region) | 66,3 ± 0,8 | 74,6 ± 0,3 | 11,0 |
| HIP05 | 1,7 | 2,2 |
From the sample from v. Maydan, Mizhgirskey district, 54 - 58 viable zoosporangia were isolated; from v. Surupy – 58 - 68; from v. Yasinya – 68 - 76; from v. Bystrets – 54 - 56; from v. Beregomet – 66 - 74 zoosporangia of the disease agent (Table 1).

As a result of differentiating varieties contamination with winter- and summer zoospores from v. Beregomet, Vyzhnytsya district, Chernivtsy region, the variety Poliska pink was infested in laboratory conditions. At contaminating with the 11 – Mizhgirskey aggressive pathotype, v. Maydan, Mizhgirskey district, Transcarpathian region – the varieties Poliska pink, Slovyanka, Pirovska, Legenda, Kalynivska were infested. As a result of contamination with the 13 – Rahivsky aggressive pathotype from v. Surupy, Rahivsky district, Transcarpathian region, the differentiating varieties Poliska pink, Slovyanka, Pirovska. Legenda and Malynska white were infested. As a result of the pathogen effect from v. Yasinya, Rahivsky district, Transcarpathian region (the 18 – aggressive pathotype), among the differentiating varieties, Poliska pink, Slovyanka, Pirovska. Legenda and Malynska white were infested. At contaminating the differentiating varieties with zoospores from v. Bystrets, Verhovinian district, Ivano-Frankivsk region (the 22 – aggressive pathotype), among the differentiating varieties, Poliska pink, Slovyanka, Pirovska, Legenda, Divo and Chervona Ruta were infested. The variety Bozhedar and Glazurna was not infested with any potato wart agent pathotype (Table 2). Thus, as a result of the performed research in laboratory conditions, it is established, that in mountainous zones of Ukraine, a process of potato wart agent aggressive pathotypes formation is taking place, and such pathotypes infest potato varieties which are resistant to a common (D1) pathotype of the disease agent. The new selected test-assortment of Ukrainian selection potato allows to identify the existing pathotypes of the disease agent. In perspective, it is necessary to carry out the identification of potato wart Ukrainian pathotypes, using the European potato test-assortment.

Fig. 2. Potato variety Poliska pink, infested with the common (D1) potato wart agent pathotype from v. Beregomet, Vyzhnytsya district, Chernivtsy region
Fig. 3. Potato variety Divo, infested with 22 – potato wart agent aggressive pathotype from v. Bystrets, Verhovinian district, Ivano-Frankivsk region

2 – Potato test-varieties reaction on wart agent pathotypes contamination (2013-2014)

<table>
<thead>
<tr>
<th>№</th>
<th>Differentiating variety name</th>
<th>Pathotypes</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>D1 – common</td>
</tr>
<tr>
<td>1</td>
<td>Poliska pink</td>
<td>+</td>
</tr>
<tr>
<td>2</td>
<td>Slovyanka</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>Pirovska</td>
<td>-</td>
</tr>
<tr>
<td>4</td>
<td>Legenda</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>Kalynivska</td>
<td>-</td>
</tr>
<tr>
<td>6</td>
<td>Malynska white</td>
<td>-</td>
</tr>
<tr>
<td>7</td>
<td>Shedryk</td>
<td>-</td>
</tr>
<tr>
<td>8</td>
<td>Divo</td>
<td>-</td>
</tr>
<tr>
<td>9</td>
<td>Chervona ruta</td>
<td>-</td>
</tr>
<tr>
<td>10</td>
<td>Bozhedar</td>
<td>-</td>
</tr>
<tr>
<td>11</td>
<td>Glazurna</td>
<td>-</td>
</tr>
</tbody>
</table>
CONCLUSIONS
1. As a result of checking the infectious capacity with potato wart zoosporangia in various disease loci, from 58 to 72 viable zoosporangia per 1g of soil are detected.
2. The test-assortment of Ukrainian selection potato allows to identify the existing pathotypes and new detected wart agent isolates in Ukraine.
3. As a result of the existing potato wart pathotypes identification in Ukraine, a common pathotype (D1), and 4 aggressive pathotypes of the disease agent: 11 – Mizhgirsky; 13 – Rahivsky; 18 – Yasinivsky, and 22 – Bystretsky, are identified.
4. The Ukrainian potato wart agent pathotypes identification is necessary to carry out with the usage of European potato differentiating varieties, for conducting their international classification.

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15. Зеля А.Г. Стійкість картоплі проти збудника раку Synchytrium endobioticum (Schilb.) Perc., методи його виявлення і диференціації. Автореферат кандидатської дисертації на здобуття наукового ступеня кандидата біологічних наук зі спеціальності 06.01.11 – фітопатологія. - К.-2009.-24 с/
Ідентифікація патотипів збудника раку картоплі в Україні

Обстежено вогнища збудника раку картоплі в Україні і проведено їх ідентифікацію за допомогою тест-сортів-диференціаторів картоплі української селекції. В н. п. Берегомет Вижницького району Чернівецької області ідентифіковано звичайний патотип – Д1; в Закарпатській області ідентифіковано агресивні патотипи: в н.п. Майдан Міжгірського району - 11 – Міжгірський агресивний патотип; в н.п. Рахов – 13 – Рахівський агресивний патотип; в н.п. Ясінія – 18 – Ясінівський агресивний патотип; в н. п. Бистрець Верховинського району Івано-Франківської області – 22 – Бистрецький агресивний патотип

рак картоплі Synchytrium endobioticum (Schilb.) Perc., патотипи, ідентифікація, диференціація, тест-сортичент картоплі