Prospects for biological methods of controlling oak pests

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Abstract. The article presents the results of experimental evaluation of biological control measures against oak adelgid, striking a young oak effectiveness. The material for the study were the young shoots of pedunculate oak (Quercus robur L.) affected Stomaphis quercus L.. The work was carried out on the basis of Undorovsky forest district of the Ulyanovsk region from May to September 2019. In the study area a detailed survey of the plantings for the major taxonomic characteristics was conducted. In determining the class status of the oak forests forest pathology technique was used. A forested area with the lowest productivity of young plantations was divided into 3 sample areas: the area of control; the area treated with biological product Bicol; the area of settlement of red ants. The area of each zone averaged 0,333 ha. In each of the trial sites 110 instances model trees affected by oak adelgid were selected, at the age of 10 years, with an average diameter of from 2 to 2.2 cm and a height of from 1.8 to 2.3 m, which were based on the number of this pest. The abundance of oak adelgid was calculated on the most populated internodes or on the whole tree, specifying the degree of damage to the tree. The dependence of the sanitary state of trees on the average number of pests has been established. The effectiveness of the biological product Bicol was 16.0 - 44%, depending on the duration of the experiment. Evaluation of the effectiveness of using red forest ants as a biological method for protecting young oak forests from Stomaphis quercus L. throughout the summer season allowed us to identify a growing trend in performance: from 10% in June to 20% in August, which suggests the relative stability of the developed biological protection system for young oak grove. The above statistical analysis confirms the positive effect of the use of the biological product Bikol and the promising nature of the method with the resettlement of red forest ants.

1. Introduction
Recently, the productivity decline of oak plantations has acquired a global dimension. Widespread tree mortality is mainly recorded in Europe, North America, and Asia. The most comprehensive reviews on the subject are given in many works of leading specialists and scientists, in particular: Hartman (1996); Delatour (1983); Steins (1980); Oleksin (1987); Leontovich (1987); Rysev (1979); Glebov (1985); Denisov (1989) and others.

Growing with each passing day, the urgency of the problem of loss of forest quality and condition oak plantations has caused an international outcry and intense anxiety. Quantitative losses of qualitative and highly productive oak groves, with a wide environmental adaptability and resistance to most stress factors reach unthinkable numbers. The need for urgent action and a quick search of ways to rescue the natural resources brought together the best scientists and experts all over the world. Largely the result of their research activities and awareness raising to this problem was the creation of international...
alliances and campaigns, which aimed at creating an integrated system that provides recovery and restoration of lost properties of this precious wood.

One such organization, dealing with the disappearance of deciduous forests, including oak, is an International Social Ecological Union (ISEU). More than 30 years of ISEU research and scientific activities has borne fruit. Years of experience exchange and analysis of the situation created prerequisites for the elaboration of a common strategy in solving such global problems, the basic component of which was the development and application of methods of combating pests [1],[2],[3],[4],[5].

More than 500 species of insects (the vast majority of Hymenoptera and Diptera) at different stages in their development as a result of their life damage the aerial organs of trees: shoots, seeds, flowers, leaves, stems. Despite the fact that oak is recognized as the most valuable breed that combines the ruggedness and resistance to adverse conditions, recent years show an increase in injuries and pathologies caused by phytophages, which are detrimental to its condition. This leads to a plus seed trees reduction – the bases of the gene pool of the region, on which the whole modern forest seed is based. The most common type in the Ulyanovsk region, damaging young oak groves, is considered to be adelgid black oak (Northern or stomates oak) - Stomaphis quercus L. The pest has a high degree of resistance to various factors and natural conditions that makes it dangerous not only for the plantations of Quercus robur L., but other species of oak trees [6],[7]. Adelgid feed on phloem sap of plants, injecting plant tissue saliva, causing warping and desiccation of the plant and as a consequence his subsequent death [8]. During mass reproduction of pests, not only young plants, but mature trees are dying as well, caught in range of multiple negative factors. Despite the knowledge of the main pests of oak, there are not sufficient detailed studies of the damaging effect of Stomaphis quercus L. on young plants of Quercus robur, although questions on the fight against adelgid are becoming increasingly important.

The methodological and technical base of plant protection is quite diverse and has many directions. Nevertheless, at the moment there is no single universal and suitable method for controlling pests and plant diseases [9]. The chemical method of controlling pests and diseases has been the most popular and advanced over the years. Unfortunately, the irrational and uncontrolled use of pesticides has led to dire consequences. It is proved that the chemical protective agents accumulated in natural systems cause poisoning of animals and humans, some of which have serious consequences, including death. Every year, the concentration of pesticides in groundwater, soil and living organisms is growing. This is primarily due to the fact that pests become resistant to pesticides, which reduces the effect of their use and leads to a multiple increase in the dose of the chemical. A large number of beneficial organisms die simultaneously with pests [10],[11]. Therefore, since the use of chemical control measures in forest park areas with a strong recreational load is undesirable, it is necessary to search and develop new biological protection methods, the action of which is based on regulating the number of pests with the help of natural enemies and biological products.

Black oak adelgid lives in close symbiosis with the ants Lasius fuliginosus [12],[13],[14],[15]. Their partnership is quite justified because the adelgid supplies fuliginosus nourishing sweet nectar (the pad). The fact that the juices on which it feeds adelgid, are composed of an exceptionally large percentage of glucose and sucrose, but protein content, which is necessary for processes of life and development is significantly less. That is why in the summer stomatis almost non-stop suck the juices of young trees in order to stock up on protein component. As a result, they pass through excess juice, which is converted in the form of honeydew, which is so necessary to fuliginosus for an active lifestyle. In response, they care and protect their supporters from enemies and adverse conditions [16],[17]. Fuliginosus strongly protect the adelgid, as well as create all conditions for comfortable holding wintering [18],[19],[20],[21]. Thus, the insulation Stamatis oak from black ants (Lasius fuliginosus) is harmful for them and requires more attention from researchers. Since fuliginosus strongly interferes with the destruction of adelgid, as evidenced by a number of publications, it was necessary to resort to the introduction of a stronger “adversary”, which if it had not destroyed the population of L. fuliginosus, it would at least reduce it. A suitable predator is the red forest ant (Formica rufa Linnaeus), as evidenced by many works of foreign
researchers [22], [23], [24], [25]. Many years of observations by staff foresters farms discovered an interesting pattern: in areas of life of old ant hills of Formica rufa Linnaeus (ages 5 - 10 years), adelgid is not observed at all, or its population is small and is isolated from 10 to 40 individuals. Based on these data, it was decided to test the resettlement of red wood ants for the protection of young stands of pedunculate oak from Stomaphis quercus L.

2. Materials and methods

The research material was young growth of pedunculate oak (Quercus robur L.), affected by Stomaphis quercus L. The work was performed on the basis of the Undorovsky forest district of the Ulyanovsk region. The studies were carried out from May to September 2019.

At the end of May 2019, a detailed survey of the stands on the main taxation characteristics was conducted on the study area. When determining the status category of oak groves, the forest pathological technique was used [26]. The result of the survey was the division of trees into categories of healthy, weakened, and lost vitality. Areas where tree specimens that lost their ability to restore predominated were subject to inspection by forest pathologists and sanitary cutting.

Examination of young growth was carried out in the same way as the examination of adult plantings. The assessment was carried out with special tools (measuring forks, altimeters, etc.) according to the methods adopted by many researchers: bonitet, crown length and extension, growth, and development indicators (Yurtsev, 1997; Yurtsev et al., 2002, 2004; Khanina, 2004). The shape, quality and projection of the plantations were established ocular, adhering to the botanical indications and gradations described in the methodological manuals and determinants of the region's woody vegetation.

| Indicators | Density | 2 and above | 3 | 4 | 5 | 5A – 5B | Total ha /% | Middle class bonitet |
|------------|---------|-------------|---|---|---|---------|-------------|-------------------|
|            | 03 – 04 | 0.3         | 1.9 | 3.0 | 1.1 | - | 6.3/5 | 3.8 |
|            | 05      | 0.5         | 3.4 | 5.8 | 2.2 | - | 11.9/10 | 3.8 |
|            | 06      | 1.1         | 10.6 | 12.8 | 3.3 | 0.2 | 28.2/24 | 3.6 |
|            | 07      | 3.1         | 19.7 | 17.8 | 4.1 | 0.2 | 44.9/38 | 3.5 |
|            | 08      | 2.0         | 9.4 | 8.3 | 1.9 | - | 21.6/19 | 3.4 |
|            | 09 – 1.0| 0.7         | 1.9 | 1.8 | 0.4 | - | 4.8/19 | 3.4 |
| Total ha /%| 7.7/7   | 46.9/40 | 49.5/42 | 13.0/11 | 0.4/- | 117.5/100 | 3.6 |

Age classes:

| Age classes | Total ha /% |
|-------------|-------------|
| young stands | 7.7/7 | 46.9/40 | 49.5/42 | 13.0/11 | 0.4/- | 117.5/100 | 3.6 |
| middle-aged maturing | 2.8 | 4.4 | 3.5 | 0.6 | - | 11.3/10 | 2.9 |
| exploitably maturing | 2.6 | 25.5 | 19.5 | 6.3 | 0.1 | 54.0/46 | 3.6 |
| declining | 1.2 | 10.3 | 12.0 | 4.1 | 0.1 | 27.7/23 | 3.7 |
| Total ha /% | 7.7/7 | 46.9/40 | 49.5/42 | 13.0/11 | 0.4/- | 117.5/100 | 3.6 |

As can be seen from the data in table 1, the most representative group are middle-aged stands, much less ripening and a small proportion of young growths. For all age groups with any completeness, low productivity of stands is characteristic.
The forest park zone with the lowest productivity of young stands was conditionally divided into 3 trial plots: No. 1 - background zone or control zone; No. 2 - was treated with biological product Bicol; No. 3 - was inhabited by red forest ants. The area of each zone averaged 0.333 ha.

Observations were carried out selectively. In each of the test sites, 110 specimens of model trees affected by oak adelgid were selected, on which the number of this pest was recorded (the total number of which was 10,230 specimens for the entire duration of the experiment). In total, 330 trees at the age of 10 years were selected, with an average diameter of 2 to 2.2 cm and a height of 1.8 to 2.3 m. The number of pests was calculated on the most populated internodes or on the whole tree, specifying the degree of damage to the tree.

The number was counted from the end of May to September 2019 on 330 model trees growing along the entire periphery of the outbreak; the number of black oak adelgid on the whole aerial part of the tree (shoots, branches, trunks) was determined manually. Observations and quantitative accounting of adelgid were carried out every day / every second day in the daytime and recorded in the logbook of the pest population (number of specimens / tree).

On each trial plot, the trees were divided into categories of conditions that were distinguished in accordance with the “Guidelines for the planning, organization and management of forest pathological examinations” [26]. The degree of damage to tree crowns by insects gnawing needles and leaves is determined by eye examination in percentage (weak type damage - up to twenty five percent, medium type - up to fifty percent, strong type - up to seventy-five percent, full type - over seventy-five percent).

In order to artificially resettle the anthill nests in the experimental plot No. 3, it was necessary to form a base in the form of large nests, from which it was possible to take layering [27], [28], [29]. In the technique of taking the lay, strict regulation was adhered to: the anthill was nominally divided into four equal sectors, two of which were shifted by shovels into a container of 100 litters; material must be taken into the cuttings, both of the cover layer and the inner cone of the nest, moreover, without destroying the remaining dome. The inner cone was covered with the material of the surface layer, giving the rounded shape of the nest [30].

Layers were resettled with the help of employees of the forest park zone at the end of the third decade of May 2019 in the trial plot No. 3 in a checkerboard pattern, every 20 m near the nests of black ants, the spatial distribution of which was noted everywhere. Nesting material was carefully poured out of the container, forming a compact dome 40-50 cm high and giving it a light conical shape, considering that the minimum size of a viable anthill is at least 50 cm in the diameter of the dome and 30 cm in height [31]. Resettlement was carried out on a sunny day, in the morning at a temperature of at least 20 degrees. In the places of the settlement, the layering took into account good illumination, since the ant hill should be under the sun for at least two hours a day. All of the above factors created ants optimal conditions for the completion of the uterine nest, while maintaining in it the temperature regime necessary for the development of juveniles, which undoubtedly ensured their accelerated survival in a new place.

The treatment of sample plot No. 2 with an area of 0.332 hectares with Bikol biological product at a concentration of 1% was carried out with an interval of 15 days starting May 27, 2019. Given that the organisms are based on living organisms, the option of treating the experimental plot in the morning or evening, avoiding the active sun [32] was followed.

The biological product is not toxic and has no adverse effects because of their selectivity; safe for endotherms, human and entomophages and will not lead to dangerous pollution of the biosphere at an acceptable concentration in the solution. Since the flow rate of the biological product is 2 kg/ha, of working liquid - 200-250 l/ha, the drug was dissolved in pure water in the proportion 0.335 kg powder in 33 litters of water. The treatment of aboveground parts of plants was carried out with the adjoining sides, evenly spraying the drug, adhering to the fine spray, no more than 20 microns – drop.

For high accuracy of results calculation of averages, the error (standard deviation), the parametric St’yudent's t-test, calculation of the use of red ants biological effectiveness and the performance of the biological product Bicol action was carried out using the computer program Microsoft Excel.
3. Results and discussion
Field studies revealed that the average abundance of black oak adelgid at the beginning of experimental observations and counting (in June) had a temporary linear trend of a quantitative increase in all areas. The intensity of the population of oak groves with black adelgid in the spring - summer period is related to the seasonal breeding season, which corresponds to the peculiarities of the biology of the pest development.

Nevertheless, in areas where Bikol was used and the red forest ants were resettled, a reduction in the number of pests was observed. The minimum abundance on the first day of the experiment was 306 copy/wood on a plot treated with Bicol. In this case, the largest population growth was observed in the control zone and averaged up to 365 ± 51.42 copy/wood (figure 1).

![Figure 1. Average number of black oak adelgid in June.](image)

The maximum abundance of Stomaphis quercus L. over the entire observation period was recorded on July 14 at the control site and amounted to 968 copy/wood. In all the studied areas, the peak of adelgid abundance was observed during the period from July 9 to July 21. In this case, the average abundance was minimal in the area treated with Bikol and amounted to 514 ± 140.59 copy/wood (figure 2).

![Figure 2. Average number of black oak adelgid in July.](image)
At the same time, the number of oak adelgid in August (figure 3) gradually decreased at all test plots to varying degrees and varied from $318 \pm 131.68$ specimens/tree (control zone) to $404 \pm 155.05$ specimens/tree (plot treated with Bicol).

![Figure 3. Average number of black oak Hermes in August.](image)

Figure 3. Average number of black oak Hermes in August.

A similar situation was observed in September. The number of this pest rapidly died away in the test plots treated with the Bikol biological product and in the area inhabited by red forest ants and varied on average from $14 \pm 35.53$ to $19 \pm 44.71$ specimens/tree. To a lesser extent, a decrease in the number was observed in the control zone, amounting to $30 \pm 58.01$ specimens/tree. The absence of adelgid populations was noted for the first time in the area treated by Bicol on September 8th. The next day, a similar trend was detected in the area inhabited by red forest ants. A day later, a zero abundance was also noted in the control zone, which was apparently primarily due to a sharp cooling and the end of the vegetative period of pedunculate oak (Quercus robur L.). Thus, in August, according to an inventory of black hermes in all areas, its natural biological attenuation of the life cycle was revealed.

A comparative analysis of the change in the number of black adelgid with a 5-day interval in all test plots revealed a significant decrease in the number of pests on plants after treatment with the Bikol and during the resettlement of ants ($p <0.05$), followed by a similar trend until June 30 (figure 4). A similar situation was noted in the following months.
Figure 4. Average number of black oak Hermes in June ((interval-5 days).

An analysis of the data obtained revealed the dynamics of the number of Stomaphis quercus L. depending on the applied biological protective measures in the studied areas. In young oak groves growing in the control zone, where no effective measures to protect young oak groves were taken, the largest abundance of adelgid was observed during each month of the experiment (figure 5). The opposite situation was revealed in the remaining test plots, where the number was significantly (p <0.05) less, which indicates a positive result of measures taken to protect the pedunculate oak.

Figure 5. Dynamics of the number of black oak adelgid.

The calculation of the biological effectiveness of the measures taken to protect young oak groves from black oak adelgid was carried out in accordance with the guidelines of E Yu Veretelnik (2012), by determining the percentage of pest mortality on the basis of which they judge the effectiveness of the measures taken [33].

Based on the results of the studies, the biological effectiveness of the biological product Bicol was from 16% to 44% (figure 6). As already noted, Bicol is a biological insecticide, the preparation of which is based on the microbiological synthesis of gram-positive spore-forming soil bacteria Bacillus thuringiensis var. Thuringiensis. Bicol differs in intestinal type of action. Once in the body of the pest Bacillus thuringiensis var. Thuringiensis produces toxins, leading to disorders of the digestive system of the insect and ultimately causes its death.

The selectivity of the drug is provided by a combination of specificity of the enzymatic and digestive systems of the pests, on which the action of the drug is directed. Multiple field trials of Bicol against different types of aphids proved its high biological effectiveness.
Figure 6. Effectiveness of biological measures taken.

The use of red forest ants as a biological means of protecting young oak groves from Stomaphis quercus L. showed the following results: 10% in June, 15% in July and 20% in August (figure 6). This positive trend is an excellent result for the first year of the experiment. Obviously, to complete the uterine nest and take root in a new place, ants require significantly more time. We can assume a rather high efficiency of this bio method in these areas in subsequent years.

A comparative analysis of the dynamics of tree damage showed a strong tendency of trees to weaken and lose homeostatic stability with a high number of adelgid, which manifested itself in the form of darkening and drying of the shoots. This is more typical of oaks growing in the control zone, where>40% of damaged trees were found (figure 7). At the same time, it was revealed that the crown of young model trees was damaged, on average, by 20% by June 15. According to the results of observations during the first and second decades of June, it was possible to note a rather high level of oak damage with intensive population of adelgid. This is true both in relation to the number of damaged trees and in relation to the number of damaged leaves, which indicates the presence of favourable conditions for mass development in the spring - summer period.

Figure 7. The degree of severe shrinkage and damage to shoots (more than 30%) on trees in the study areas (in%).

Young shoots darkened and acquired a brown hue, then the young growth dried up, which eventually spread throughout the shoot, which subsequently died and fell. To a lesser extent, this manifestation of reduced tree viability affected the area treated by the Bikol, where the percentage of severely damaged trees did not exceed 13%. On the trial plot with ants, this indicator was <20%.

4. Conclusion

In the studies shown above, the biological effectiveness of a biological product Bicol was 16.0 – 44 % depending on the time duration of the experiment, which is a fairly low value of efficiency, given the more significant positive effect, according to the manufacturer. It may be necessary to carry out the testing of the application of a stronger concentration of biological product with a change of frequency of treatment of trees.

Evaluation of the effectiveness of the use of red ants as a biological method of protection of young oak from Stomaphis quercus L. during the summer season allowed us to identify an increasing trend of productivity from 10% in June to 20% in August. This positive trend is a result for the first year of the experiment, given the fact that some time passed since the resettlement of the ants. Obviously, for the completion of the breeding nests and sewing in a new place, the ants takes more calendar time. Therefore, we can assume a fairly high efficiency of this biological control in subsequent years. This
method is most promising due to its versatility, as forest ants have an extensive range of applications in forestry, as evidenced by many studies.

A detailed survey and analysis of oak stands in the end of the experiment showed a strong tendency of the trees to weaken and reduce resistance with a high number of adelgid in the control plot, which was manifested in the form of darkening and drying of the shoots.

Based on the study, we can conclude about the positive effect of the biological product Bicol and the promising method with the resettlement of red forest ants. Nevertheless, a number of issues remain unresolved and the final decision on the appropriateness of introducing these methods should be taken after 2-3 years of testing in several forestry enterprises, which differ in a number of factors: temperature and humidity conditions, light exposure, crown density, water regime, soil type and composition, and other agroecological conditions.

Thus, without taking effective measures to preserve oak groves, there is a real possibility of a complete loss of oak plantations in the region. It is necessary to develop new methods of protection based on the use of biological agents, where the main principle is to regulate the number of pests with the help of natural enemies and biological products.

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