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Bioecology of the sea-buckthorn fly (*Rhagoletis batava obscuriosa* Kol.) and pest control treatment in Altai

Lyubov D. Shamanskaya

The Lisavenko Research Institute of Horticulture for Siberia

49 Zmeinogorsky Tract, 656045, Barnaul, Russia

e-mail: shamanskayald@gmail.com

Summary

Seabuckthorn fly is known as the main insect-pest of sea buckthorn on the territory of Russia. In Altai Krai, where seabuckthorn is the main industrial plant, crop losses from this insect-pest reach 100 % as larvae harm and destroy the pulp of fruit.

The sea buckthorn fly is a highly specialized insect-pest damaging only sea buckthorn berries. More than half of the varieties of Altay genotype are highly susceptible to the pest. Early varieties are damaged most strongly. We picked up only one pest resistant form and some steady-state forms which can be cultivated without carrying out protective measures.

Population of the seabuckthorn fly changes considerably year by year and depends on biotic and abiotic environmental factors.

The short-term forecast of the seabuckthorn fly's productivity developed at our institute allows to refuse from carrying out spraying against the pests in certain years.

In the years of mass development of seabuckthorn flies, a reliable protection of crop is provided by protective measures directed against a harming phase – larvae. Spraying is carried out within the period of larvae hatching when their number reaches 5-12 %. Any delay in terms of protection promotes considerable losses of crop and deterioration of its quality by the main rate – the content of oil.

A method of decreasing the dose of chemical and biological pesticides by 2-10 times was developed, allowing the reduction of content of toxic remains in fruits and receiving a crop without remains of pesticides.

Key words: Seabuckthorn (SB), seabuckthorn fly (SB fly), short-term prognosis, agents, efficiency, productivity, quality of fruit.

INTRODUCTION

The seabuckthorn is the main culture in industrial horticulture in Siberia. Owing to the high ecological plasticity and the richest biochemical structure of the fruit seabuckthorn cultivation has been started in many countries of the world. As a rule, insect-pests, among which the SB fly is the most harmful, follow SB in exploring new habitats.

On the territory of Russia crop losses because of this pest reaches 100 %. Universal distribution of SB flies in the midland of Russia became the main reason for withdrawing seabuckthorn berries cultivation from this region.

In Altai Krai, where the main industrial SB plantations are located, development of pest control measures began in 1960 and proceeds so far. The long-term practice of SB fly control showed that reliable crop protection is provided by chemical pesticides that does not guarantee receiving production without toxic remains (Goreeva, Prokofiev, 1996).This problem can be successfully solved due to cultivation of pest-resistant and relatively resistant varieties; however, the majority of varieties of the Altay genotype has high susceptibility to the pest (Shamanskaya, 2006).

Attempts to replace chemical pesticides by biological protection means (Kalvish, 1986; Hovalig, 2005), including a classical bio-method (Berger, Danilov, 1998), yet did not find wide application in practice.

Further research showed that using of biological product Phytoverm that was developed by the NBTs "Farmbiomed" (Moscow) against the SB fly, provides receiving eco-friendly yield (Shamanskaya, 2014).

However, high cost of this agent is an obstacle for its use on big areas.

Development of economically and ecologically reasonable system of seabuckthorn protection against the seabuckthorn fly became the main objective of our further research.

MATERIALS AND METHODS

The test of the agents against the seabuckthorn fly was carried out in a production-scale experiment and a small plot experiment with the background of one - and double-fold spraying. The expected losses of crop were counted by the original technique (Shamanskaya, Usenko, 2007).

Sprayer OP-200 with the consumption rate of 1000 l/ha was used in production-scale experiment and knapsack sprayer "Quasar" with the consumption rate of 0.6-1 l/tree in the small plot experiment. Control plants were no treated. The experiment was repeated three times. The age of plants in the small plot experiment was 5 years, in the production-scale experiment 5-6 years. The total area of small plot experiment was 0.7 ha and the total area of production experiment was 6-7 ha.

The efficiency of treatment was determined by accounting the death of larvae in 100 damaged fruits while viewing them under the microscope of MBS-10.

The biochemical analysis of fruits was carried out by the standard techniques in biochemical laboratory of The Lisavenko Research Institute, the toxicological analysis was carried out in Central research- and-production veterinary radiological laboratory (Barnaul) by a method of gas-liquid chromatography. The remains of Aversectin C after treatment by biological agent Phytoverm were determined in NBTs "Farmbiomed" (Moscow) by a method of fluorescent highly effective liquid chromatography. Experimental data was processed by a method of dispersive analysis (Dospehov, 1973).

RESULTS

The seabuckthorn fruit fly belongs to the order *Diptera,* family *Tophritidae.* On the first figure are pre- sented the original pictures of imago, larvae and pupae of the seabuckthorn fly (Fig.1)

SB fly has a hidden lifestyle and a complex developmental circle. Puparium (false cocoon) overwinter under the canopy of damaged trees, mainly at the depth of 1-5 cm. Pupation of larvae occurs approximately one week before the beginning of fly flight.

The fly belongs to thermophile insects. The timing when the flight of adult insects starts depends on the temperature conditions in the previous weeks. Hatching of larvae lasts up to 6 weeks; the most ac- tive hatching lasts about a week and usually occurs in the first half of July.

Adult insects feed on seabuckthorn berries, piercing their skin by ovipositor. Flight, mating and oviposition occur only in warm weather. In 6-14 days after the beginning of flight, females start laying eggs, placing them one by one under the skin of fruit. Sometimes in one berry 2-3 eggs can be laid. The fertility of one female reaches 200 eggs and more.

A

B

C

D

Figure 1. Seabuckthorn fly: A – imago, B – larva inside of berry, C – larvae before pupation, D – pupae

In the conditions of Altai Krai, larvae are the most harmful. Hatching begins in the first or second decade of July and ends in the second or third decade of August. Larvae of the early age are usually small and transparent, later they have yellowish colour and reach 7 mm in length. On the forepart of their body two black chitin jaws (Maxillae) are situated and on the back-end a pair of light brown suction cups is presented which hold larvae on the surface of fruit. Within 3-4 weeks larvae feed on pulp of fruits, leaving the thin skin untouched. After destruction of one fruit, the larva gets over in the next one, damaging up to 50-100 % of fruits in one whorl (fig. 2).

The fruits occupied by larvae are softened, gradually deformed, darken and dried up without falling down (fig. 3).

Figure 2. Berries damaged by larvae

A

B

Figure 3. Character of fruits damaged by the seabuckthorn fly: A – in a period of active nutrition of larvae, B – after larvae leaving to pupation

SB fly is a highly specialized pest. It damages only seabuckthorn berries. The long-term assessment of seabuckthorn varieties of the Altay genotype showed different extent of their damage by the SB fly. Among them, 55 % are highly susceptible to the pest. They are, first of all, varieties of the early term of maturing. There is one form of a highly pest-resistant variety and some relatively resistant varieties which at a certain planting scheme can be cultivated without carrying out protective measures against the SB fly (Shamanskaya, 2014). The majority of relatively resistant varieties are small-fruited, red- fruited and late-ripening varieties (Fig. 4).

A

B

C

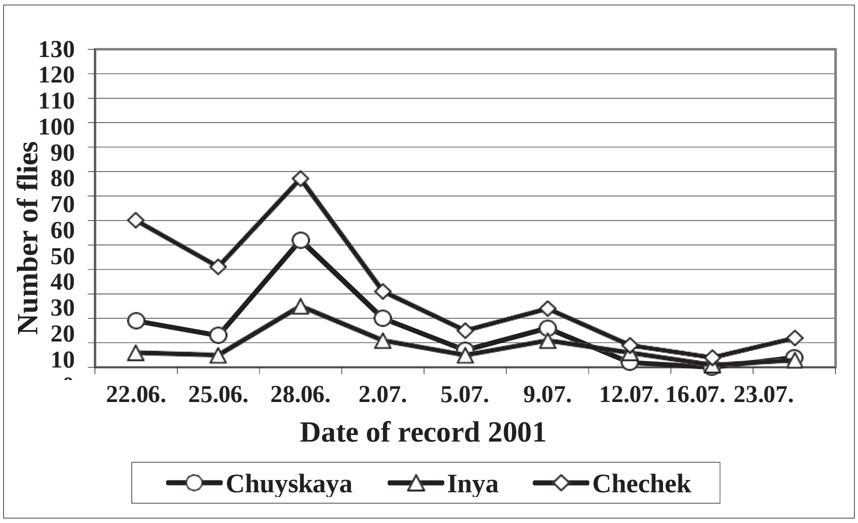
Figure 4. Varieties of seabuckthorn berries: A – highly-resistant to seabuckthorn fly, B – relatively resistant red-fruited, C – relatively resistant late-ripening

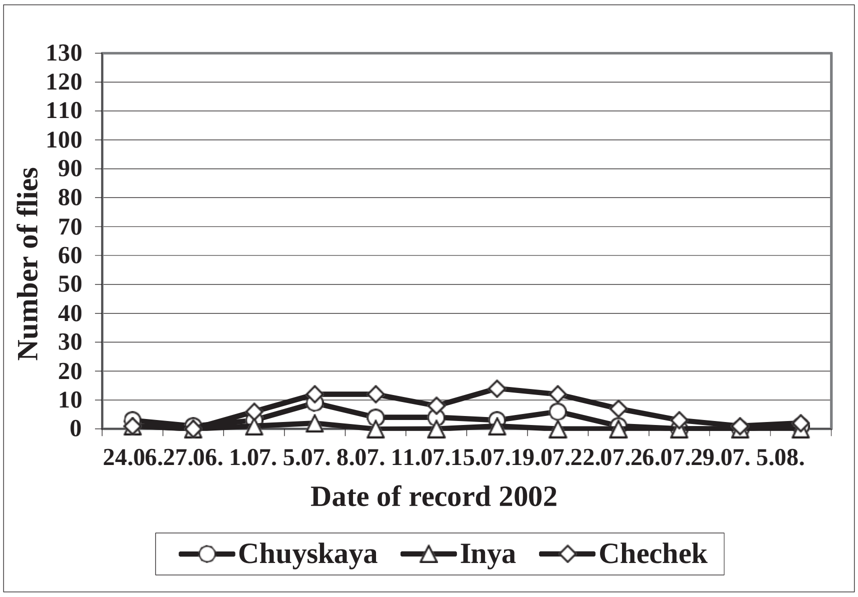
Long-term observations have shown that the number of SB flies significantly changes year by year and depends on biotic and abiotic environmental factors. In certain years up to 70 % of puparium are catched by the parasite of fruit flies (*Opius rhagoleticollis*). In wet years up to 40 % of puparium can be damaged by diseases. Much greater influence on the change of SB fly population size in conditions of sharp and continental climate of Altai territory is caused by abiotic environmental factors. Population size of pests decreases in 2-6 times during long (2-3 weeks) flooding of seabuckthorn areas; due to the freezing of soil surface in the years with low snow cover; sharp cold events during the flight of adult insects and their additional nutrition, and also in conditions of abnormal heat while eggs are developing. Many larvae perish when they go into the soil to form pupae. We believe that not less than 90 % of the population perishes due to natural reasons.

Within 10 years we carried out observations of population dynamics of SB flies on the experiment site where pesticides were applied. Dynamics of adult insect flight was determined by the usage of yel- low glutinous traps (fig. 5).

Figure 5. Yellow glutinous trap

Year 2001 was the first year of monitoring under favorable conditions for pests and a lot of SB flies were noticed (Fig. 6).





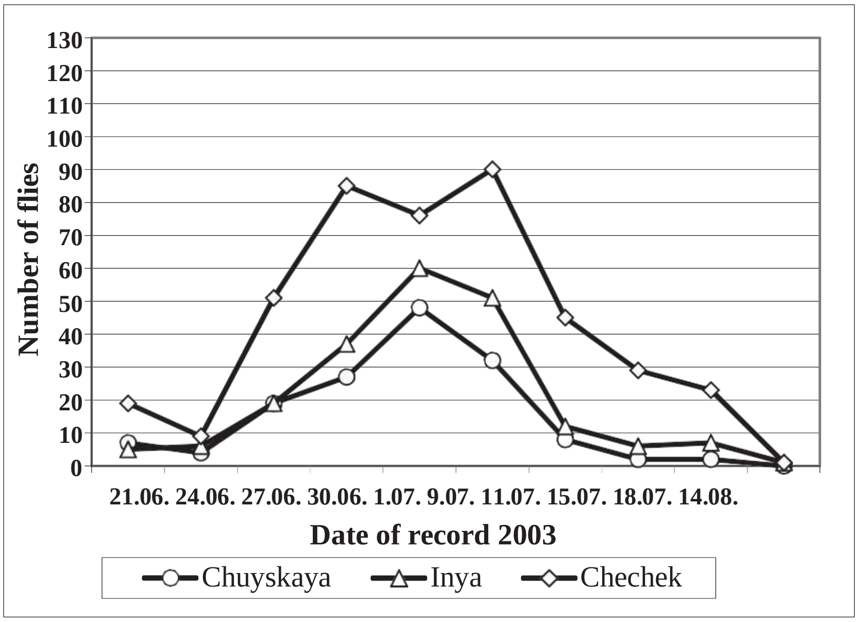


Figure 6. Dynamics of SB flies flight, 2001-2003.

In 2002 sharp temperature drops with the fall of air temperature down to 10-11^0 C during flight of adult insects led to reduction of pest population. The period of flight of adult insects was stretched and weak. In these conditions the fertility of females decreased sharply. Dry and hot weather at the end of spring – at the beginning of summer 2003, promoted an active reactivation of pupae, mass adult insect flight and substantial increase of pest population.

In 2004, population of SB flies remained high. This was caused by hot and dry weather in spring and in the beginning of summer. The flight of adult insects began early – on the 10th of June, but it was suspended by pouring rains. In connection to this the timing of pest development was shifted for later period, but it did not reduce their harmfulness.

During the next 4 years (2005, 2006, 2007 and 2008) the population of SB fly in the experimental site considerably decreased, which is connected with the adverse weather conditions during the various periods of pest development.

In 2009, due to the favourable weather conditions, the increase of pest population was observed. In 2010 population of SB flies decreased again in connection with incessant pouring rains during the flight period of adult insects.

Despite considerable fluctuations of SB fly population, its harmfulness remains at the rather high level due to their ability for a long-term diapause. However, in certain years with a well-functioning forecast system of SB fly population it is possible to abandon spraying against the pest.

We developed and patented a short-term forecast of population of SB flies, based on the accounting of total number of adult insects on the yellow glutinous traps (Shamanskaya, Usenko, 2007) where the expected losses of yield are calculated with the use of equation of regression:

Y = 0.45 x X – 0.1496, where

Y – the expected loss of yield, %

X – the total number of adult insects on the average on 1 trap

0.45 - regression factor

0.1496 - constant regression factor

The use of a short-term forecast of SB fly population in practice allows to abandon spraying against the pest on the area of tens hectares of industrial plantations in certain years. So, for example, in 2014 SB flies treatments were cancelled on the area of 92 ha.

The decrease of seabuckthorn fly population in 2014 was connected with the long influence of low temperatures in the reactivation period of pupae and the negative impact of abiotic stress – the air temperature increased up to +38 °C before larvae hatched from the eggs which led to their mass death.

When developing a short-term forecast for SB fly population, it is necessary to keep in mind that in the other regions the fertility of females can vary considerably in comparison with pest population in Altay, therefore in each case the adjustment in calculations of the expected losses of a yield is necessary.

SB flies move extremely actively from the old plantations to young plantations. In the first year of fruit-bearing highly susceptible varieties can already be damaged by 99.9%. Thus, the varieties located in close proximity to the source of infection become populated most intensively. Our research findings showed that the most active settling of plants by the SB fly in 2004 was noticed on the variety Altayskaya.

In the second year of fruit-bearing happened a redistribution of pests on the site and more intensive flight was noticed on the variety Elizaveta that is located at the maximum distance from the source of infection. During next years the variety Elizaveta remained leader in susceptibility to the seabuckthorn fly on this site.

The long-term practice of the seabuckthorn fly control showed that the reliable crop protection is provided by protective measures that are directed against a harming phase – larvae, with the use of chemical pesticides. Actellic (Pirimiphos-methyl) showed a good result among them. It reliably protects crop but does not guarantee production without toxic remains.

Further research work was directed on decreasing the dose of Actellic due to combined use with bio-supplements developed at our institute.

In case of low damage level caused by the seabuckthorn fly, with the expected loss of crop/yield 1.3-5.0%, the efficiency of spraying at the level of 96-100% is received by decreasing the agent dose from 1 liter to 100 ml/ha that is 10 times less (Table 1).

Table 1. Efficiency of a microdose of Actellic against seabuckthorn flies. Results of production experiment. Treatment 27.07.2007

Treatment	***Variety***	***Death of larvae, %***
Without spraying - control	Chuyskaya Inya Zhemchuzhnitsa	19.3 25.4 18.6
Actellic – 1 l/ha (standard)	Chuyskaya	100
Actellic – 0.1 l/ha + bio-supplement	Chuyskaya Inya Zhemchuzhnitsa	96.0 100 100
LSD $_{0.5}$	-	3.6

In case of high damage where the expected loss of crop was 44%, the total death of pests was received by decreasing Actellic dose from 1 liter to 400 ml that is 2.5 times less (Table 2).

According to the records of plant productivity in the second experiment, the productivity in control was 4.65 t/ha, and in Actellic spraying with total and lowered dose the productivity was more than 11 t/ha. The yield increase compared to control was 151% and 149% respectively (Table 3).

Table 2. Efficiency of reduced doses of Actellic against seabuckthorn flies. Results of production experiment (variety Chuyskaya) on 30.07.2010

Treatment	***Death of larvae, %***		
	2-3.08	***9-10.08***	***19.08***
Without spraying - control	2.4	3.0	0.1
Actellic – 1 l/ha (standard)	93.0	100	100
Actellic – 0,5 l/ha + bio-supplement	85.0	97.0	100
Actellic – 0,4 l/ha + bio-supplement	82.0	95.0	100
Actellic – 0,3 l/ha + bio-supplement	70.7	82.5	96.2
Actellic – 0,2 l/ha + bio-supplement	59.2	62.0	84.8
LSD $_{0.5}$	0.98	0.86	0.84

Table 3. Productivity of seabuckthorn with reduced Actellic doses. Results of production experiment (variety Chuyskaya) from the year 2010.

Option	***Productivity***		***Increase compared to control***	
	kg/bush	***t/ha***	***t/ha***	***%***
Without spraying - control	3.94	4.65	-	-
Actellic– 1 l/ha (standard)	9.91	11.71	7.06	151
Actellic – 0,5 l/ha + bio-supplement	9.65	11.41	6.76	138
Actellic – 0,4 l/ha + bio-supplement	9.80	11.58	6.93	149
Actellic – 0,3 l/ha + bio-supplement	8.60	10.16	5.50	118
Actellic – 0,2 l/ha + bio-supplement	6.50	7.68	3.03	65
$LSD_{0.5}$	1.63	0.06	-	-

Results of biochemical analysis of fruit showed that combined use of reduced dose of Actellic and bio-supplement improved some indicators. The content of sugars, acids and the amount of carotenoids were higher in comparison with the full dose of the agent.

Decrease in dose of Actellic from 1 to 0.4 l/ha in experiment in 2010 reduced the quantity of toxic remains in fruit in 1.4 times. In certain years this kind of treatment allows to receive production without toxic remains.

Among biological agents a good effect was shown by Phytoverm (1%), developed in Farmbiomed scientific and production association (Moscow) on the basis of not pathogenic soil fungus *Streptomyces avermetilis* strain VNIISKHM-54 or strain VNIISKHM-51. In the growth process the culture of primary producer forms a complex of “close to chemical” substances that are named avermectin, which have biological activity. Phytoverm does not pollute environment, it quickly dissipates in water and soil. Expectation time from the use on various cultures is 3 days, on the sea-buckthorn – 2 weeks.

A single Phytoverm spraying against SB fly provides a short-term protective effect; therefore for a reliable protection of crop the double treatment is needed.

Phytoverm is an expensive agent and it is an obstacle for its wide implementation into production chain. A combined use of Phytoverm with bio-supplement allows to halve the dose. The table 4 shows the efficiency of full and reduced dose of Phytoverm in the production experiment which in the first case was 100 % and in the second case was 97.7 %.

Productivity at full dose of Phytoverm was 9.92 t/ha, at the lowered – 9.45 t/ha. The increase of crop was respectively 162% and 150% (Table 5).

Table 4. Efficiency 1% EC of Phytoverm against seabuckthorn flies. Results of production experiment (variety Chuyskaya) in 2009

Treatment	***Number of sprayings***	***Death of larvae, %***		
		28.07	***4.08***	***11.08***
Without spraying - control	-	8.6	12.4	5.1
Actellic – 1 l/ha (standard)	1	76.2	100	100
Phytoverm – 3 l/ha	1	11.1	67.5	84.7
Phytoverm – 3 l/ha	2	-	89.6	100
Phytoverm – 1.5 l/ha+ bio-supplement	1	15.6	57.4	68.1
Phytoverm – 1.5 l/ha + bio-supplement	2	-	95.9	97.7
$LSD_{0.5}$	-	3.8	3.4	1.3

Table 5. Yield of seabuckthorn when used various Phytoverm doses. Results of production experiment (variety Chuyskaya) in 2009

Treatment	***Number of sprayings***	***Productivity***		***Crop increase***	
		kg / bush	***t/hectare***	***t/hectare***	***%***
Without processing - control	-	3.2	3.78	-	-
Actellic – 1 l/ha (standard)	1	8.2	9.69	5.91	156
Phytoverm – 3 l/ha (standard)	1	8.4	9.21	5.43	143
Phytoverm – 3 l/ha	2	7.8	9.92	6.14	162

Phytoverm - 1,5 l/ha + bio-supplement	1	4.9	5.79	2.01	53
Phytoverm - 1,5 l/ha + bio-supplement	2	8.0	9.45	5.67	150
$LSD_{0.5}$	-	2.0	0.31	-	-

The biochemical analysis of fruits showed that combined use of Phytoverm with bio-supplement improves the quality of fruit according to the content of dry soluble substances, sugars, vitamin C and oil.

The toxicological analysis of fruit showed that the use of biological agent Phytoverm in seabuckthorn fly control allows cultivation of eco-friendly production.

The advantage of using biologically active supplement for decreasing a consumption rate of pesticides in years with arid weather conditions should be especially marked because one of its components increases the resistance of plants to the drought. For example in 2012 when there were arid weather conditions in a small plot experiment, the maximum increase of crop 138-150 % in relation to control was received in treatments with bio-supplement (Table 6).

Table 6. Yield and weight of seabuckthorn berries (planting 2008) associated with different spraying doses. Results of production experiment (variety Chuyskaya) in 2012

Experiment	***No. of sprayings***	***Mass of 100 fruits g***	***Productivity***		***Increase of crop compared to control, %***
			kg/bush	***t/hectare***	
Without spraying - control	-	50.1	1.5	1.8	-
Actellic – 0.1 % (standard)	1	51.0	3.2	3.9	116
Actellic – 0.04 % + bio-supplement	1	53.1	3.6	4.5	150
Phytoverm (1 % EC) – 0.3 %	2	51.4	2.6	3.2	77
Phytoverm (1 % EC)- 0.15 % + bio- supplement	2	54.5	3.5	4.3	138
$LSD_{0.5}$	-	0.7	0.8	0.98	-

Weight increase of fruit from 51.0 g (Actellic full dose) to 53.1 g (Actellic + bio-supplement) and from 51.4 g (Phytoverm full dose) to 54.5 g (Phytoverm + bio-supplement) was observed.

Calculation of economic efficiency showed a low level of profitability of fruit production when using various seabuckthorn fly sprayings in 2012. It was connected with the high costs of manual harvesting and incomplete accession of plants into fruit-bearing phase. The highest profitability – 72.3% was received when using Actellic in a complex with bio-supplement (Table 7).

Profitability increased by 11.5% compared to the full rate of application of the agent. In case of double decrease of dose of expensive agent Phytoverm, the profitability of fruit production was 62.5 % which exceeds the ratio of full dose by 29.5% unit. The analysis of the received data showed that the use of full dose of Phytoverm is not economically justified, as profitability of fruit production was 1/3 less than in the control treatment.

The biochemical analysis of fruit showed the increase in content of carotenoids from 18.50-18.97 mg/100g (use of full doses of the agent) to 20.08-22.10 mg/100 g (a combined use of reduced doses of Phytoverm and bio-supplement). The content of oil increased from 3.97-4.41% to 4.59-4.93% respectively.

These results got confirmation in a production experiment where Actellic (0.4 l/ha) was used in a complex with bio-supplement. The experiment was carried out in the same year on the area of 28 hec- tares. In this case the amount of carotenoids increased from 17.94 mg /100 g to 20.14 mg/100 g when using lowered dose of Actellic and bio-supplement instead of full dose of Actellic, and the content of oil increased from 4.91% to 5.64% respectively.

The toxicological analysis showed 4 times less toxic remains in fruits after combined use of Actellic with bio-supplement in a small plot experiment. In production-scale experiment toxic remains in fruits were not found.

Table 7. Economic efficiency of seabuckthorn fruit production associated with different spraying backgrounds.

Treatment	***Yield, t/ha***	***Costs of 1 ha, thousand roubles***		***Prime cost, thousand rub/ha***	***Earnings thousand rub/ha***	***Profit, thousand rub/ha***	***Level of profitability, %***
		total	***protective treatments***				
Without treatment - control	1.8	79.7	0	44.2	82.8	38.6	48.4
Actellic-1 l/ha (standard)	3.9	111.5	1.22	28.5	179.4	67.9	60.8
Actellic – 0.4 l/ha + bio-supplement.	4.5	120.1	0.49	26.6	207.0	86.9	72.3
Phytoverm - 6 l/ha	3.2	110.6	9.60	34.5	147.2	36.6	33.0
Phytoverm - 3 l/ha + bio-supplement.	4.3	122.3	4.80	28.4	198.8	76.5	62.5

During carrying out protective measures against a harming phase – larvae, it is very important to define optimum timing of spraying. This allows destroying the pest at early stage of its development and provides active regeneration of the damaged tissue and minimum loss of fruit weight. It is possible to destroy larvae at late stage of development, but the part of fruit is heavily deformed, its weight is decreased and that conducts to the considerable loss of crop (Fig. 9).

If the quantity of such fruits is 10% - the loss of crop is 3.5%, but if their quantity increase to 50 percent - losses of crop increase to 14% (Table 8).

The content of oil in damaged fruits decreases to 4.84%, in comparison with outwardly healthy fruits where content is 5.55%.

A

B

Figure 9. Fruits of sea buckthorn, variety Inya: A – healthy, B – damaged by SB flies

Table 8. Change in yield weight at various amounts of damaged fruits. Variety Inya in 2009.

Indicators	***Healthy fruit***	***Content of damaged fruit in yield weight, %***									
		1	***10***	***20***	***30***	***40***	***50***	***60***	***70***	***80***	***90***	***100***
Weight of 100 fruits, g	92.7	92.5	89.5	86.7	84.0	81.8	79.8	73.5	70.3	67.1	63.9	60.5
% compared to con-trol	-	99.7	96.5	93.5	90.6	88.2	86.0	79.2	75.8	72.3	68.9	65.2
Loss of yield, %	-	0.3	3.5	6.5	9.4	11.8	14.0	20.8	24.2	27.7	31.1	34.8

CONCLUSION

In the conditions of sharp and continental climate of Altai Krai population of SB flies varies considerably from year to year. Population forecast of pests developed by the Lisavenko Scientific and Research Institute allows abandoning application of pesticides on scores of hectares of SB industrial plantations in certain years.

During the days of mass development of seabuckthorn flies, combined use of insecticide Actellic and biological product Phytoverm with bio-supplement is ecologically reasonable and economically sound. Bio-supplement allows reducing the agents in 2-10 times compared to standard provides the maximum increase of crop by 138-150 % and improves the basic fruit quality index – carotenoid and oil content.

Combined use of bio-supplements and insecticide Actellic allows lowering the content of toxic remains in fruits by 1.4-4 times, and in some cases to receive production without any remains of pesticides.

Application of biological pest control agent Phytoverm for crop protection allows getting crop without toxic remains. In the changing conditions of market economy the usage of this agent in complex with bio-supplements is economically justified.

The maximum safety of a crop is provided by the optimum time of spraying against a harming phase – the larvae, which number by the beginning of treatment should not exceed 5-12%.

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