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Development of Environmentally Friendly Protection Measures Against Pests and Diseases

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Abstract: The paper describes the monitoring of phytosanitary conditions in different regions of the Almaty region (Kazakhstan) and demonstrates the large population of cruciferous fleas (*Phyllotreta cruciferae*) on rapeseed plants, which reaches 7-9 spec./m^2, exceeding the economic injury level (3-5 spec./m^2). The disease is observed to spread on wheat and barley seeds. Proceeding from this, the study aims to develop effective protection measures against cruciferous fleas and to ecologize agricultural technologies using safe techniques and means of protection. The study uses methods of registration when monitoring the spread of pests and determining the effectiveness of the methods and tools applied. It is demonstrated that the treatment of wheat seeds with a protective and stimulating mixture using the preparation of bisolbisan 1 l/t + extrasol 1 l/t raised the intensity of crop growth to 94.2% compared to 70.9% in the control sample. High biological efficacy is noted from the use of actarophyte 1 l/ha mixed with extrasol against cruciferous fleas, the population of which after treatment is only 1 spec./m^2 against 7.8 spec./m^2 in the control case. The study also analyzes the results of releasing the *Trichogramma* ovipositor *(Trichogramma)* in the amount of 350 g/ha, *Habrobracon* (*Bracon hebetor Sa*y) at a rate of 500 spec./ha and green lacewings *(Chrysopidae)* at a rate of 1,500 eggs/ha to combat the cotton bollworm *(Helicoverpa armigera*) and the European corn borer (*Ostrinia nubilalis Hbn.)*. This first known treatment of rape crops using a drone against cruciferous fleas achieved a biological efficiency of 95.8-96.2%.

Keywords: Pests, Diseases, Organic Products, Biological Products, Entomophages

Introduction

In recent years, in the organization of protective measures against harmful organisms, the negative impact of pesticides on the environment and human health has become a major problem. In this regard, back in 2007, the parliament of the European union made a decision obliging all member countries to switch to biological crop protection methods that rule out the above-mentioned negative consequences (EPC, 2018).

Understanding the importance of the problem, the ministry of agriculture of the Republic of Kazakhstan, along with research institutions, directed their efforts to the greening of protective measures, reducing and in some cases eliminating the use of pesticides in agrocenoses to ensure phytosanitary safety and to obtain environmentally friendly products (Adilkhankyzy *et al*., 2022).

Many researchers have noted the advantages of ecologization of protective measures against harmful fauna, ensuring the preservation of soil fertility, landscape, and biodiversity of agrobiocenoses and ultimately, the creation of conditions for organic agriculture (Henning *et al*., 1991; Hole *et al*., 2005; Gomiero *et al*., 2011).

Kazakhstan's transition to this new paradigm focuses on improving the efficiency of agricultural production through the introduction of environmentally friendly systems of plant protection against pests, which provides a solution to the problem of organic agriculture production (Temreshev *et al*., 2023). We believe that the results of scientific research on the development of advanced agro technologies and the implementation of an environmentally friendly set of protective measures against pests will serve as an important step towards the production of environmentally friendly (organic) products (Alimbekova *et al*., 2021).

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The aim of this study is to develop efficient measures of protection against cruciferous fleas (*Phyllotreta cruciferae*) and improve the eco-friendliness of agro technologies through safe techniques and means of protection.

Materials and Methods

Location of the Study

Field research was conducted on the experimental fields of Svetlana LLP in the Karasai district, Almaty region, Kazakhstan, and laboratory work was carried out in the labs of the institute.

Study Design and Sample Collection

Treatment of seeds against diseases with protective and stimulating compositions was performed with an ordinary sprayer with a spraying nozzle. The laboratory experiments associated with the treatment of plants with protective and stimulating preparations were carried out in early spring before sowing the crop (April 15-20). Wheat, barley, corn, and rape were the plants on which common diseases related to fungal and bacterial microflora were studied. Field studies using unmanned aerial vehicles for treatments were conducted in the most vulnerable period in the development of cruciferous fleas, which in the years of the study were the 1st and 2nd 10-day periods of June.

Phytosanitary monitoring was conducted to identify the pests and their harmfulness during the main phases of plant growth: Tillering, stem formation, earing, and milk wax ripeness (Nasiyev *et al*., 2015; PRK, 2015).

The examination of seeds evaluated their seeding qualities (germination energy on the 3rd day, laboratory germination on the 7th day). Seed sowing qualities were assessed in wet chambers. Fifty seeds were taken for the experiment from each of the crops in 4-fold replication and the number of diseased seeds and seedlings was registered. The species composition of fungal and bacterial microflora was established. The tests were performed according to N.A. Naumova's method, which consists in using two nutrient media (potato agar and synthetic Chapek's medium) to record the growth of infection. Identification of fungal and bacterial microflora was performed according to the morphological features of fungal and bacterial colonies and their pure cultures. The morphological features of fungi were examined via microscopy of sporonocytes and bacteria were studied based on their pathogenic properties.

Larger flying insects were detected visually as they took off from plants after the passage of the first observer, after which they were captured with an entomological net by the second observer. In addition, the observer independently conducted a continuous mowing of grass with an entomological net to find entomophages in the mass of captured insects. For the identified entomophages and phytophages, which were separate objects of the study, optimal conditions of laboratory maintenance (food, temperature regime, lighting, humidity, and special structures and devices for their accommodation) were created.

Biopreparations and Growth Stimulants

In selecting biopreparations and growth stimulants for the pre-sowing treatment of seeds and spraying of crops during the growing season, the account was taken of the degree of bacteria influence on the stimulation of growth processes and plant development, immunity enhancement, and increased resistance to stress.

Since infected seeds release toxins, to eliminate their negative effect on plants, special presowing treatment was performed using preparations with high fungicidal and bactericidal properties in combination with growth stimulators that activate physiological processes in the development of seeds.

For the treatment of grain crop seeds, various protective and stimulating preparations that favorably affect their sowing qualities (germination energy and laboratory germination, intensity of seedling, and root system growth) and effectively inhibit fungal and bacterial microflora were developed and tested.

In the laboratory conditions, we evaluated the effectiveness of the biopreparations and growth regulators extrasol, bisolbisan, phytosporin 350, actarophyte, green gold 0.3%, potassium humatophosphate and humate and determined the timing of treatments and product consumption rates.

Phytosporin, which is intended for the pre-sowing treatment of seeds and spraying of crops during the growing season, stimulates intensive growth and development, strengthens immunity, and increases plant resistance to stresses. Against crop pests, we used the systemic insecticide green gold, 0.3% (azadirachtin), which was also applied as a protective and stimulating agent when treating wheat and barley seeds.

Experiments with the use of protective and stimulating compositions for the studied crops accounting for the bactericidal and fungicidal properties of various biopreparations and their combination with bioinsecticides and growth regulators were developed and organized in the field setting.

To suppress the development of pests, we used such biopreparations as actarophyte 7,000 Tg/l, green gold 0.3% 3,8000 Tg/l, and extrasol 1,800 Tg/l. The main advantage of these biological agents is that they, unlike pesticides, do not cause addiction (resistance) in pests and resistance to diseases, while providing high efficiency at the accepted rates of consumption of the active substance.

For the first time in the practice of plant protection, treatments against cruciferous fleas with biological preparations actarophyte, green gold 0.3%, and extrasol were carried out with the use of an unmanned aerial vehicle (Fig. 1).

Use of the Entomophages

The ecologization of protective measures included the use of the entomophage *trichogramma*, *Habrobracon* (Fig. 2), and green lacewing against the pests. In corn crops to suppress the development of the cotton bollworm and the European corn borer, the parasitic *Trichogramma* was released. Additional releases of *Habrobracon* and green lacewings were performed at some sites in case of an increase in the number of pests. Lacewings were used to suppress the turnip sawfly and cruciferous fleas on rape crops.

Statistical Analysis

Statistical processing of the obtained data was carried out by the method of descriptive statistics. Calculations were performed using the excel software from the Microsoft office software package (Microsoft, USA).

Fig. 1: Monitoring and treatment of fields using unmanned aerial vehicles

(a)

(b)

Fig. 2: (a) *Habrobracon* cassettes prepared for release; (b) *Habrobracon* cassettes placed on crop crops

Results

Infestation of Wheat and Barley Seeds

During the phytoexpertese of wheat and barley, fungal and bacterial microflora dominated and the total contamination rate of the seeds reached 90-100%. All samples had fungi of the genera *Alternaria* and *Fusarium.*

There also were saprophytic species of fungi, genera *Mucor* and *Aspergillius*, that cause mold in seeds. Almost all the analyzed seeds were infected with saprophytic and pathogenic microflora, in the range of 28-58.1%. Subsequently, during the growing season, the infection of seeds by diseases led to the development of root rot, *Fusariosis*, *Alternariosis*, and *Bacteriosis*, thereby impairing the sowing quality of seeds and lowering the germination energy and productivity of plants (Table 1 and Fig. 3).

The Efficiency of Protective and Stimulating Preparations

Analysis of the sowing qualities of seeds treated with biopreparations demonstrates that their germination energy on the 3rd day varies depending on the various characteristics between 47.8 and 86.7%. and the germination rate is 92.5-100%. The percentage of diseased seeds and seedlings in the control group is as high as 97.5-100%, while in the experimental variants treated with the developed protective and stimulating compositions, no diseases are found. For example, the percentage of infection in wheat varieties does not exceed 1.7%. The best performance is demonstrated by the variant phytosporin 5.0 l/t + green gold 0.3% 0.3 l/t, which provides an increased intensity of seedling growth and a high percentage of laboratory germination (Table 2).

The records indicate that the seeds of all varieties exhibit high growth energy and laboratory germination on moistened sand (Table 3).

Table 1: Infestation of wheat and barley seeds with fungal and bacterial microflora (nutrient medium)

Variety	Number of diseased seeds and germs, pcs.	Fungal microflora, %						Bacterial microflora, %
		Alternaria	*Fusarium*	*Mucor*	*Aspergillius*	*Penicillium*	*Bipolaris*	
Wheat								
Almaly	100	64.2	-	35.7	-	-	-	28.0
Glassy	100	42.8	-	14.2	14.2	-	-	33.0
Barley								
Arna	100	47.6	4.7	33.3	-	-	9.5	54.6
Baisheshek	100	25.8	2.7	-	-	-	1.8	58.1

Table 2: Efficiency of treatment of wheat and barley seeds with protective and stimulating preparations (in a moist chamber)

Variant	Growth energy, %	Laboratory germination, %	The intensity of germ growth at day 5, %			Infestation by fungal and bacterial microflora, %
			Low	Medium	High	
Wheat, variety Glassy 24						
Control	51.1	95.7	23.3	70.9	-	97.5
Extrasol, 1.0 l/t + potassium humatophosphate, 20 mL/t + tumat, 50 mL/t	52.2	92.5	5.0	88.5	-	1.7
BisolbiSan, 1.0 l/t + extrasol, 1 l/t	66.7	97.5	2.5	94.2	-	0.0
Extrasol, 2.0 l/t + green gold 0.3%, 0.3 l/t	64.2	98.8	1.7	91.7	-	0.8
phytosporin, 5.0 l/t + green gold 0.3%, 0.3 l/t	63.1	97.3	1.5	91.1	-	0.2
wheat, variety Almaly						
Control	47.8	95.5	2.5	94.2	-	100.0
Extrasol, 1.0 l/t + potassium humatophosphate, 20 mL/t + tumat, 50 mL/t	76.7	99.2	4.2	95.8	-	0.0
BisolbiSan, 1.0 l/t + extrasol, 1 l/t	67.8	96.7	2.5	94.2	-	0.0
Extrasol, 2.0 l/t + green gold 0.3%, 0.3 l/t	82.5	100.0	0.8	95.0	5.0	0.8
Phytosporin 5.0 l/t + green gold 0.3%, 0.3 l/t	83.3	100.0	0.5	97.1	-	0.0
barley, variety arna						
Control	70.0	94.5	15.0	81.7	-	100.0
Extrasol, 1 l/t + potassium humatophosphate, 20 mL/t + tumat, 50 mL/t	85.5	98.3	3.3	94.4	-	0.0
BisolbiSan, 1.0 l/t + extrasol, 1 l/t	86.7	98.3	11.7	86.7	-	0.0
extrasol, 2.0 l/t + green gold 0.3%, 0.3 l/t	79.2	99.2	3.8	85.4	10.0	0.0
Phytosporin, 5.0 l/t + green gold 0.3%, 0.3 l/t	88.2	100.0	5.5	96.3	-	0.0
barley, variety baisheshek						
Control	80.3	92.3	12.5	80.8	5.0	100.0
Extrasol, 2.0 l/t + potassium humatophosphate, 20 mL/t + tumat 50 mL/t	82.2	95.3	5.0	92.5	-	0.0
BisolbiSan, 1.0 l/t + extrasol, 1 l/t	83.3	94.7	10.8	86.7	-	0.0
Extrasol, 2.0 l/t + green gold 0.3%, 0.3 l/t	83.3	98.3	6.6	81.1	10.6	0.0
Phytosporin, 5.0 l/t + green gold 0.3%, 0.3 l/t	87.7	98.6	5.9	92.4	-	0.0

Table 3: Effect of the protective and stimulating mixture (phytosporin, 5.0 l/t + green gold, 0.3%, 0.3 l/t) on growth energy and laboratory germination of wheat seeds (wet sand experiment)

	Indicator, %	
Variety	Energy of growth	Laboratory germination
T-4	68.5	94.0
1675-149	97.2	99.7
2046-1	96.7	99.0
1127	89.0	96.2
2041	96.5	98.7
Turkey-12	65.2	89.5
1675-170	98.2	98.7

Table 4: Efficiency of treatment of wheat seeds with a protective and stimulating mixture of Phytosporin, 5.0 l/t + green gold 0.3%, 0.3 l/t

Variety	The intensity of germ development, %	Number of diseased seeds and germs, %
T-4	+++	100.0
1675-149	+++	10.0
2046-1	++	70.0
1127	++	13.3
2041	++	0.0
Turkey-12	+	86.6
1675-170	++	10.0

Note: + low intensity, ++ medium intensity, +++ high intensity of development

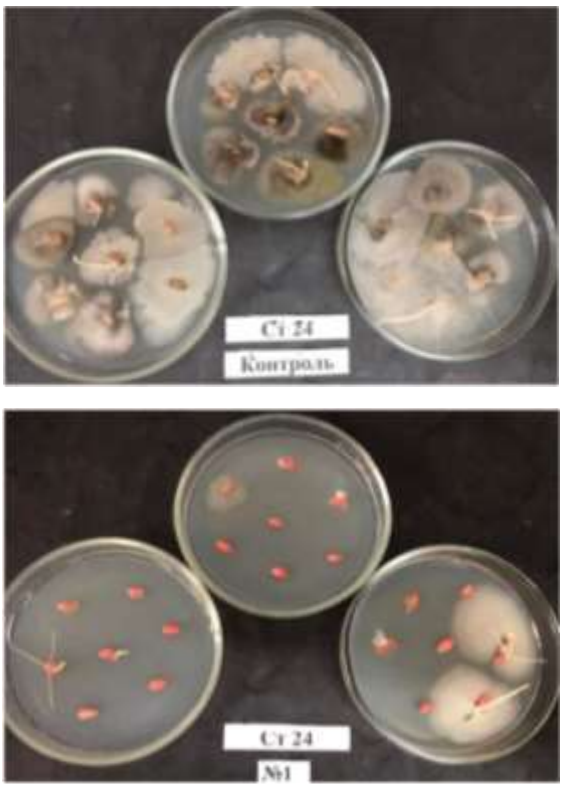

Fig. 3: Effect of the protective and stimulating mixtures on the fungal and bacterial microflora of wheat seeds (on a nutrient medium)

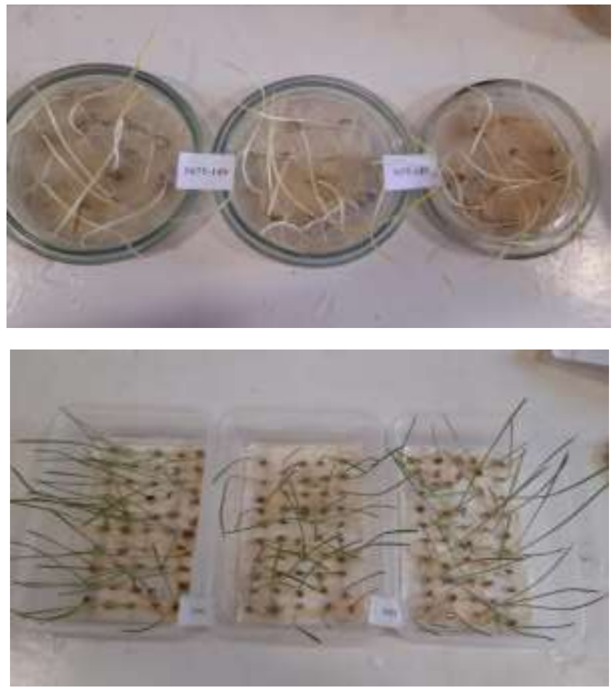

Fig. 4: Wheat seed growth in a moist chamber

Records indicate that of all seed varieties, only the varieties T-4 and Turkey-12 show growth energy of 68.5 and 65.2%, respectively. However, the reduction of growth energy has little effect on the laboratory germination of the indicated variety Turkey-12, which amounts to 89.5%.

The same experiment examined the intensity of germ development and the number of infected seeds and germs depending on the protective and stimulating compounds used (Table 4 and Fig. 4).

The results obtained prove phytosporin to have virtually no suppressing effect on the development of fungal microflora, as the number of deceased germs of the varieties T-4, 2046-1, and Turkey-12 ranges between 70.0-100%, while the varieties 1675-149, 1675-170 and 1127 have only about 10.0-13.3% of affected germs. High effectiveness of the agent is observed on the variety 2041 on which no signs of the disease are detected at all.

Phytoexpertise of seeds on a nutrient medium reveals that all analyzed wheat varieties are infected with fungal and bacterial microflora. The dominant ones are fungi from the genera *Fusarium*, *Helminthospoium*, and *Alternaria*, which cause plant damage by Alternariosis (black mold), Fusariosis, and root rot. Saprophytic fungi from the genera *Mucor*, *Penicillium*, and *Aspergilus* are found to cause seed molding, while Pseudomonas atrofacions bacteria are the causative agents of basal bacteriosis, *Xanthomonos translucens* causes black chaff of wheat and Erwinia *Carotovora* leads to root system diseases.

Table 5: Biological effectiveness of products against cruciferous fleas on rape crops (Karasai district, Svetlana LLP)

Experiment variant	Repetition	Flea population, spec./m^2 Before treatment	On the reporting day 1	3	7	Decrease in population, % on the reporting day 1	3	7
Actarophyte, 1 l/ha + extrasol, 2.0 l/ha	1	5.1	3.90	1.8	1.1			
	2	5.5	3.10	2.0	0.9			
	Mean	5.3	3.50	1.9	1.0	51.7	75.3	87.1
Green gold 0.3%, 0.3 l/t + extrasol, 2.0 l/t	1	5.9	3.50	2.1	1.3			
	2	5.1	3.30	2.1	1.1			
	Mean	5.1	3.40	2.1	1.2	53.1	72.7	84.6
Control (without treatment)	1	5.1	7.20	7.5	8.0	-	-	-
	2	5.0	7.30	7.9	7.6	-	-	-
	Mean	5.0	7.25	7.7	7.8	-	-	-

Table 6: Efficiency of entomophages against the cotton bollworm and the European corn borer

Entomophages	Decrease in the pest population, %
Green lacewing *(Chrysopidae)*	48
Trichogramma ovipositor *(Trichogramma)*	62
Habrobracon (*Bracon hebetor* say)	70

Effectiveness of Products Against Cruciferous Fleas

Monitoring of rape crops during the emergence of seedlings and up to the phase of the second pair of true leaves on the crops, cruciferous fleas (*Phyllotreta cruciferae*) was highly widespread, with their population reaching 7-9 spec./m^2, which exceeds the economic injury level (3-5 spec./m^2).

Meanwhile, flea population density during the season depended significantly on weather conditions, in particular the average daily air temperature. The maximum number of phytophages (up to 500 spec./m^2) was typically reported in the 2^{nd} 10-day period of May. The biological efficiency of the treatment of crops with biological agents via unmanned aerial vehicles against cruciferous fleas amounts to 84.6-87.1%.

The obtained data on the treatments performed are shown in Table 5.

The results of recordings indicate that 7 days after the treatment, the population of fleas on the control plot on average amounts to 7.8 spec./m^2 in contrast to 1.0 spec./m^2 on the plot treated with actarophyte mixed with extrasol and 1.2 spec./m^2 with the use of green gold 0.3% together with extrasol. The efficiency of colonization was rather low, which owes to the small population of pests, meaning that the lack of nutrition impaired the reproduction of the bioagent.

Colonization of entomophages against the cotton bollworm and the European corn borer proves to be ineffective. Specifically, the use of *Trichogramma* at a rate of 300 g/ha reduced the pest population by 62% (Table 6). In turn, the release of the green lacewing against the European corn borer and cruciferous fleas at the rate of 500 eggs/ha provided only a 48% decrease in their population.

The greatest efficiency is demonstrated by the colonization of the *Habrobracon* against the cotton bollworm. The release of the *Habrobracon* as an additional suppressor for the pest at the rate of 500 spec./ha resulted in a 70% reduction in their number.

Discussion

The conducted studies testing biological preparations and entomophages on rape, wheat, and corn crops testify that with the comprehensive application of safe plant protection products against pests, it is quite feasible to organize organic farming in crop agrocenoses.

One of the fundamental methods of ecologization of protective measures, providing ecologically clean (organic) products at the very beginning of crop cultivation, is the phytoexpertise of seeds. Herewith, the introduction of protective and stimulating compositions made up of safe preparations during this period simultaneously contributes to seed health improvement, increases their laboratory germination, and enhances the energy of growth processes (Dosmanbetov *et al*., 2020).

In our experiment, the treatment of seeds with extrasol and bisolbisan yielded an increase of 92.5-100% in their laboratory germination. Meanwhile, the percentage of disease infestation is only 0.8-1.7% compared to 97.5% in the control case. The relevance of using protective and stimulating compositions is supported by the data obtained in the experiment when establishing the contamination of seeds with diseases before sowing (Tables 2-4).

Entomophagy testing reveals that some species have relatively poor efficacy. Suffice it to say that the parasite *trichogramma* at the rate of 300 g/ha against the cotton bollworm and the green lacewing against the European corn borer at the rate of 500 eggs/ha suppressed the development of pests only by 62-48.0%, respectively (Table 6). Therefore, we consider the use of these bioagents against cotton bollworm and the European corn borer, respectively, to be inexpedient. A rather high efficiency against the cotton bollworm is demonstrated by the bioagent *Habrobracon*, whose colonization at the rate of 500 spec./ha reduced the population of the pest by 70% (Table 6).

Thus, our findings show that changes in agro technologies of plant protection measures, while ensuring the safety of crops from damage, create conditions for obtaining ecologically clean (organic) products. The introduction of such a technique in plant protection technology as phytoexpertise of seeds with the use of protective and stimulating compositions made of safe agents to improve their health can enhance the germination and growth processes of plants. Additional incorporation of entomophages into the comprehensive protection of plants provides ecologization of agricultural technologies and ultimately creates conditions for reducing pesticide loads on agrocenoses, which eliminates the negative effects on the environment and provides environmentally friendly products for the population and animals. The conducted research is consistent with the general trend of ecologization of the agro-industrial complex around the world, with the need for food becoming a greater concern for the population of the planet every year (Varghese and Sharma, 2018; Manne and Kantheti, 2021).

In this regard, the authors of many studies stress the extent to which food production must be scaled up to feed the ever-increasing population of the earth (Bachu *et al*., 2006; Thorp *et al*., 2008; Waga and Rabah, 2014).

Conclusion

The first known investigation related to the treatment of rape crops against cruciferous fleas using an unmanned aerial vehicle shows that the biological effectiveness of this method allows for suppressing the development of the pest up to 95.8-96.2%. The use of the *Habrobracon* bioagent against the cotton bollworm suppressed the development of the pest by 70%. The results of colonization by the parasitic *Trichogramma* and lacewings were somewhat weaker. In further studies, other more efficient types of entomophages could be used.

Research on the mass breeding and use of entomophages in agrocenoses will continue in our future works to develop and implement a comprehensive system of biological protection of corn, rape, and other crops against pests.

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Author’s Contributions

All authors equally contributed to this study.

Ethics

This article is original and contains unpublished material. The corresponding author confirms that all of the other authors have read and approved the manuscript and no ethical issues are involved.

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