General toxicity study of a pyrethroid preparation with permethrin

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Abstract. This article presents the results of the general toxicity assessment of a pyrethroid preparation containing permethrin. The acute toxicity assessment of the preparation showed that the dose of 2060 mg/kg and 1545 mg/kg was the lowest dose that caused lethal outcome in rats and guinea pigs, respectively. A single administration of the studied preparation to rats and guinea pigs at doses of 1373 mg/kg and 1030 mg/kg 9, respectively, did not lead to death of the experimental animals. According to the OECD classification, the pyrethroid preparation with permethrin belongs to hazard class 4. Experiments on mice indicate that the studied preparation is not characterized by a cumulative effect. A single treatment of calves with a pyrethroid preparation containing permethrin at a concentration of 0.05% of the active substance did not cause signs of toxicosis, changes in the general condition and behavior of the animals. We also did not note any deaths of calves with a single treatment with the studied preparation at the above concentration. It was concluded that the pyrethroid preparation with permethrin does not have a toxic effect when used and can be classified as a safe drug.

1 Introduction

By now, synthetic pyrethroids have found wide application in animal husbandry to protect cattle from ectoparasites [1. 2]. These broad-spectrum insecticides are characterized by significant selective toxicity and a period of residual effect on the animal's hair [3. 4]. Synthetic pyrethroids do not accumulate in the body of the treated animal and are excreted in low quantities in milk [5. 6]. The above-mentioned properties of pyrethroids are significant due to the fact that new classes of insecticides have recently appeared, which have different mechanisms of action and anthropogenic impact [7-9]. Some researchers have shown that after treating cattle with a pyrethroid-based preparation, the incidence of attacks by pasture flies and horseflies is significantly reduced, and a decrease in the number of mosquito detections is noted [10. 11].

The Type I group of pyrethroids includes permethrin, which is closely related to natural compounds found in the flowers of various species of chrysanthemums [12. 13]. Permethrin

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is systemically active against a range of pests [3. 14]. It is able to act on the nerve cell membrane to disrupt sodium channel current, resulting in delayed repolarization and paralysis of the pest [3. 15].

Permethrin was originally registered in 1979 for use by the U.S. Environmental Protection Agency [12]. According to Agency members, about 2 million pounds of this pyrethroid are used annually in agriculture and medicine [16].

Permethrin preparations are widely used in modern animal husbandry to combat ectoparasites [14. 17]. This pyrethroid in the form of solutions and ointments is used to treat the skin and fur of animals, as well as the buildings of livestock enterprises [6. 18].

The safety assessment of veterinary drugs is aimed at preserving and protecting the health of farm animals by eliminating a potential drug whose predicted risk from clinical use exceeds its possible benefit [19. 20]. In the process of establishing safety, information is obtained on the toxicity, pharmacological properties, pharmacokinetic parameters and metabolism of the studied drug [21]. Preclinical safety assessment of a veterinary drug includes general toxicity studies [20].

The aim of this work was to evaluate the general toxicity of a pyrethroid drug with permethrin.

2 Materials and methods

A pyrethroid preparation with permethrin was used in the experiments. The preparation is a yellowish solution with a specific odor. The composition of the preparation includes: permethrin - 2%, nephraz - 44%, neonol - 10%, dioxanol - 44%. In appearance, this is a yellow-brown liquid that mixes with water in all proportions. The preparation is recommended for use on animals to combat ectoparasites, as well as to destroy flies in livestock and farm buildings.

The LD_{50} of the test drug was determined using the method of Deichmann and Le Blanc (1943) [22]. The studies were conducted on laboratory mongrel guinea pigs and rats. The drug was administered intragastrically. When determining LD_{50} for intragastric administration, the test doses were selected so that each subsequent dose differed from the previous one by 1.5 times. The lowest dose causing a lethal outcome was taken as LD_{50}. The control group of animals was administered drinking water in the same volume and according to the same scheme as the drug under study. Nutrition and conditions of keeping the animals met the established requirements.

The cumulative properties of the drug were determined on 10 outbred mice using the method we described earlier [4], which takes into account the procedure for calculating the cumulative coefficient (Kcum) with daily administration of a dose of 1/5 LD_{50} (established taking into account the death of animals by day 14).

To assess the effect of a pyrethroid preparation with permethrin on hematological indices of calves' blood, two groups of 2-3-month-old black-and-white bull calves were selected, 5 heads in each. The calves of the first group were sprayed with the preparation at a dose of 0.05% (by active substance), the second group (control) - with water. Blood for the study was taken from animals before spraying and 1, 5, 15 and 30 days after it.

To study the effect of the drug on hematological blood parameters, the following parameters were selected: hemoglobin content, erythrocytes, leukocytes and erythrocyte sedimentation rate. Determination of hematological parameters was carried out using generally accepted methods [4].

When working with animals, the requirements of the European Convention for the Protection of Vertebrate Animals (Strasbourg, 18 March 1986) and the ethical principles were observed [23]. The ethical standards of the Institutional Animal Ethics Committee were followed during the study.

The maintenance and care of the experimental calves was carried out in accordance with the "Veterinary rules for keeping cattle for the purposes of their reproduction, growing and sale" [Order of the Ministry of Agriculture of the Russian Federation of October 21, 2020 N 622 "On approval of the Veterinary rules for keeping cattle for the purposes of their reproduction, growing and sale"].

The data of the experimental studies were processed using the variation statistics method. For this purpose, the Statistica 13.3 application program was used. The statistical significance of the differences was determined by the value of the Student criterion.

3 Results and discussion

Compared with the most commonly used organophosphate, organochlorinated and methylcarbamate insecticides, the synthetic pyrethroid permethrin has been shown to be a less toxic insecticide to mammals [24]. Permethrin is also less toxic than cypermethrin and fenvalerate [24], which are type II pyrethroids [25]. According to the literature, permethrin causes a tremor-type syndrome characterized by tremors throughout the body, aggressive behavior, and ataxia [13].

A mandatory condition for the use of new veterinary drugs is the preliminary conduct of toxicological studies on laboratory animals [20. 26].

An acute toxicity assessment of the pyrethroid preparation permethrin showed that a dose of 2060 mg/kg was the lowest dose that caused lethality in laboratory rats. An intermediate dose of 1716 mg/kg body weight did not cause death in the experimental animals. Single doses of 1373 mg/kg and 916 mg/kg were not lethal to rats.

When studying the acute toxicity of a pyrethroid preparation with permethrin on guinea pigs, we found that a dose of 1545 mg/kg was the lowest dose that caused a lethal outcome in animals. The introduction of the preparation at an intermediate dose of 1288 mg/kg of body weight did not lead to the death of the experimental animals. A single administration of the studied preparation at doses of 1030 mg/kg and 687 mg/kg to guinea pigs did not cause the death of the latter.

After the introduction of the pyrethroid preparation, the animals showed signs of agitation, which passed after 20-30 minutes. The general condition of the animals remained satisfactory, no changes in behavior were observed, appetite and thirst were normal, there were no convulsions, reactions to tactile, pain, sound and light stimuli were adequate. During the experimental period (14 days), signs of intoxication and death of animals were also not noted. The results of the acute toxicity assessment of the pyrethroid preparation with permethrin are presented in Table 1.

Table 1. Acute toxicity parameters of a pyrethroid preparation with permethrin when administered intragastrically to laboratory animals.

Animal	LD_{50}, mg/kg	Therapeutic dose, mg/kg	Therapeutic breadth index or LD_{50}/therapeutic dose
Rats	2060	7.5	274.7
Guinea pigs	1545	7.5	206.0

It has been established that according to the OECD classification, the drug belongs to hazard class 4.

When studying the accumulation, a dose of 0.2 LD_{50} (0.2 LD_{50} = 412 mg/kg) was used, which was administered to white mongrel rats for 20 days. During the experiment, 3 out of 10 animals died (i.e. 30%). At the end of the study (18-20 days after the administration of the drug), the animals showed depression, decreased appetite, rapid breathing, shortness of

breath, and thirst. As a result of the experiments, we found that the accumulation coefficient is 6.7 (Table 2).

Table 2. Cumulative effect of pyrethroid preparation with permethrin.

LD_{50}, mg/kg	Total dose Dk, mg/kg	Kcum
2060	82400	6.7

The results obtained in accordance with the guidelines for establishing the toxic properties of veterinary drugs allow us to classify the studied pyrethroid drug as a group of drugs with no cumulative properties.

To establish the effect of a particular drug on the animal's body and its safety, a comprehensive toxicological assessment should be carried out, which includes a hematological analysis [4]. Without these studies, it is difficult to judge the possibility of widespread use of a veterinary drug in practice.

A single treatment of calves with a pyrethroid preparation containing permethrin at a concentration of 0.05% of the active substance did not cause signs of toxicosis, changes in the general condition and behavior of the animals. We also did not note any deaths of calves with a single treatment with the studied preparation at the above concentration.

In Table 3 we present data on the hematological parameters of calves after the use of a pyrethroid drug.

Table 3. Morphological parameters of calf blood after spraying with a pyrethroid preparation with permethrin at a concentration of 0.05%.

Indicators	Units of measurement	Before spraying	After spraying			
			1 day	5 days	15 days	30 days
Erythrocytes	10^{12}/l	6.3±0.7 5.9±0.8	5.8±0.6 6.2±0.4	6.3±0.8 6.6±0.5	5.7±0.2 6.1±0.3	5.9±0.1 6.0±0.2
Hemoglobin	g/l	96.4±8.8 97.5±8.8	93.9±5,4 96.8±5.3	103.6±5.5 100.7±5.1	97.2±3.7 96.4±3.4	100.0±8.0 99.4±6.5
Leukocytes	10^{9}/l	9.5±1.7 9.8±1.4	10.2±1.4 9.7±1.3	6.7±0.2 6.8±0.4	7.6±0.5 8.2±0.4	6.5±0.7 6.3±0.8
Color index	-	0.5±0.04 0.4±0.03	0.5±0.05 0.6±0.05	0.6±0.04 0.7±0.03	0.4±0.02 0.5±0.06	0.6±0.07 0.7±0.07

Note: The numerator shows the data of the experimental group of animals, and the denominator shows the data of the control group

According to the data in Table 3, it is evident that the treatment of calves with the studied preparation had virtually no effect on the morphological parameters of their blood. The number of erythrocytes, leukocytes, hemoglobin and color index did not differ from the parameters of the control group ($p > 0.05$).

Based on the results of this study, it can be reasonably concluded that the pyrethroid preparation with permethrin does not provoke the development of pathological reactions and does not have a toxic effect when used by animals.

4 Conclusions

Thus, the results of toxicological studies show that the pyrethroid preparation with permethrin does not have a toxic effect when used and can be classified as a safe drug.

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