



INSECTICIDE SUSCEPTIBILITY OF HOUSE FLIES
(*MUSCA DOMESTICA*) FROM A LIVESTOCK FARM
IN TYUMEN REGION, RUSSIA

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Summary

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The susceptibility of the field populations of the house fly *Musca domestica* L. (Diptera: Muscidae) from a livestock farm of the Tyumen region, Russia, to six insecticides (deltamethrin, cypermethrin, thiamethoxam, permethrin, fipronil, chlorfenapyr) is reported. After a forced contact of adult flies of susceptible strains and first generation of field populations with the residues of insecticides at the bottom of glass cups, the median lethal dose of each insecticide was determined by probit analysis. According to the resistance ratio, the susceptibility of the evaluated field population to insecticides increased in the order: cypermethrin < deltamethrin < permethrin ≤ chlorfenapyr < thiamethoxam < fipronil.

Key words: field population, insecticides, *Musca domestica* L., resistance ratio, susceptibility

The house fly *Musca domestica* L. (Diptera: Muscidae) is a typical representative of zoophilous flies that cause significant harm to the animals as ectoparasites and interfere with the work of the maintenance staff. Flies disturb the animals, reduce the sanitary condition of farms and animal feed, may be carriers of infectious and parasitic diseases of animals (Förster *et al.*, 2007; Wang *et al.*, 2011; Scott *et al.*, 2014). The main method of house flies control worldwide is chemical control

using insecticides. In Russia, pyrethroid and neonicotinoid insecticides are most commonly used to combat ectoparasites of animals. Fipronil and chlorfenapyr are among the new classes of insecticides in Russia. Fipronil is used in crop production, medical pest control (mainly in the form of gels and baits to control cockroaches), in veterinary medicine for combating ectoparasites on small domestic animals. Chlorfenapyr is a promising pyrethroid insecticide to control the insect popu-

lation. Chlorfenapyr, a pro-insecticide, has a unique mechanism of insecticidal activity connected to the ability of its metabolites to block the ATP synthesis in insect body cells (US EPA, 2001). In world practice, chlorfenapyr is mainly used in crop production and termite control, it is promising as a means to control the malaria vectors and other synanthrope insects (N'Guessan *et al.*, 2007; Romero *et al.*, 2010). It is not introduced widely in Russia and is mostly used for plant protection.

It is known that *M. domestica* quickly develops resistance to insecticides used against it (Scott *et al.*, 2013; Khan *et al.*, 2015). Previously, researchers reported on the formation of house fly populations resistant to pyrethroids and neonicotinoids on livestock and poultry farms around the world (Marçon *et al.*, 2003; Kaufman *et al.*, 2010; Khan *et al.*, 2013; Scott *et al.*, 2013; Abbas *et al.*, 2015). Development of cross-resistance to insecticides of other groups is also possible. For example, it is known that house fly population resistant to permethrin develops resistance to fipronil (Liu & Yue, 2000).

Information on the susceptibility of *M. domestica* field populations to the insecticides used on the territory of Russia is sporadic and mainly refers to flies of towns (inhabited localities). Thus, in the 1990s, a study of 19 populations of flies from central (Moscow, Kaluga, Tver regions) and southern (Astrakhan) regions of Russia, demonstrated their high resistance to organophosphorus insecticides (chlorophos, carbofos) and tolerance or sensitivity to such pyrethroids as permethrin, neopinamine, fenvalerate (Polyakova, 1995). It has also been reported that the flies of the Moscow population were highly resistant to pyrethroids (Roslavtseva, 2001). Such studies of flies on livestock complexes are rare and have

not been carried out for more than 10 years (Pavlov, 2006).

For effective control of the number of pests, including the housefly, it is necessary to carry out monitoring of the level of susceptibility in field populations to the used insecticides. The aim of this study was to assess the susceptibility to insecticides of different groups in the *M. domestica* population from a livestock farm in the Tyumen region, where pyrethroid insecticides had been used for a long time.

For the experiment, 3–5-day old laboratory susceptible and field populations adult flies were used. Field population of flies was collected in the facilities (piggeries, cowsheds) of a livestock farm in the Tyumen region (Chervishevo village, 56°56'08" N. 65°25'34" E.). Then insects were transferred in an insectarium and kept in cages made of metal frame 25 × 25 × 25 cm of size, covered with fine mesh, with a special sleeve to run insects. The cages also had food (milk powder: glucose 1:1) in Petri dishes and cups with water with a cotton wicks in them. The first generation of adult flies (F1) has been obtained in the laboratory out of the collected population of flies; it has been tested for susceptibility to insecticides. Susceptible insect culture was obtained from the laboratory of the Novosibirsk Agrarian University in 2009 and was reared in the insectarium without contact with insecticides for more than 50 generations.

The testing was carried out using commercial insecticides containing deltamethrin (Deltsid 4% EC, Agrovetzashita, Russia; FAS 1% tablets, Agrovit on scientific and technological orders of Gigiena-Bio, Russia), cypermethrin (Samarovka insecticide 25% EC, Samarovo, Russia), thiamethoxam (Agita 10% WG, NOVARTIS ANIMAL HEALTH Inc.,

Switzerland), as well as industrial permethrin (97%, crystals, China), fipronil (97%, powder, China), chlorfenapyr (97%, crystals, China).

To determine the toxicity of the insecticides for flies the method of dosed contacting of insects was used (Pavlov & Pavlova, 2005). The insect groups were exposed to the test drug without anaesthetic by forced contact with an insecticide residues on the bottom of a glass cup for 30 min. Acetone solutions of insecticide were prepared in 5–6 concentrations. Then, 1 mL of each solution was added to glass cups with diameter 35–40 mm and 40–45 mm height. After evaporation of acetone a certain amount of active substance – the dose – was left in each cup. Then 10 flies were placed into each cup by a piston, consisting of a mesh cloth and a spring spacer ring, achieving close contact of the insects with the insecticide residues on the bottom of a cup. After a 30-min exposure, pistons in cups were raised to supply insects with drinking bowls with 5% glucose solution. The death of the insects was registered during one day. Experiments with each dose of the insecticide were performed at least 3 times.

The results of insect mortality depending on the dose were analysed by probit analysis (Finney, 1971) using Free LD50/LC50 Calculator (July 7, 2016; Dr. M. Alpha Raj by Calculating LD50/LC50 using Probit Analysis in Excel Blog) for calculating LD₅₀ for 95% confidence intervals (CI), slopes, standard errors, and χ^2 . Median lethal dose was expressed as a microgram of the active substance per gram of tested insects' weight. To determine the susceptibility of the field population to the insecticides the resistance ratio (RR) was calculated as the ratio of LD₅₀ of the insecticide for field population flies

to LD₅₀ for laboratory strain of flies. Susceptibility of the field population to the insecticides was determined based on the following criteria: RR<1 – high susceptibility to the insecticide, RR=1 – absence of resistance (the population is sensitive to the insecticide), RR=2–10 – very low resistance (the insects are tolerant to the insecticide), RR=11–20 – low resistance (the insects are moderately resistant), RR=21–50 – medium resistance, RR=51–100 – high resistance (the insects are resistant to the insecticide), RR>100 – very high resistance (the insects are highly resistant to the insecticide) (Dremova *et al.*, 1999; Abbas *et al.*, 2015).

Laboratory tests have shown that for flies of both laboratory and field populations the pyrethroid deltamethrin had the highest toxicity. Next came the pyrethroid cypermethrin, pyrazole fipronil and neonicotinoid thiamethoxam. The permethrin toxicity was significantly lower. The lowest toxicity against adults *M. domestica* of both laboratory and field populations was that of pyrrole chlorfenapyr (Table 1).

According to our results, the field populations of flies did not have a high resistance to deltamethrin, cypermethrin and permethrin. A very low resistance to cypermethrin (RR = 3.66) and deltamethrin in the composition of the FAS preparation (RR = 3.07) should be noted. A higher sensitivity to permethrin in the studied population of flies is probably due to the fact that insecticides containing this pyrethroid are quite rare.

House fly populations with varying degrees of resistance to pyrethroids were found around the world on livestock and poultry farms. For example, in the USA, *Musca domestica* caught in pens for cattle in the southeast Nebraska showed reasonable resistance to permethrin: resistance ratio of 7.3 when contacting with insecti-

Table 1. Toxicity of insecticides to adult house flies of field population (Chervishevo village, Tyumen municipal region, Russia) compared to susceptible population

| Insecticide | Population | N | LD ₅₀ (95% CI) | Slope (±SE) | χ ² | RR (95% CI) |
|--------------|-------------|-----|---------------------------|-------------|----------------|------------------|
| Deltamethrin | Susceptible | 265 | 0.443 (0.210–0.938) | 0.911±0.166 | 0.544 | |
| | Chervishevo | 532 | 0.407 (0.249–0.666) | 1.701±0.109 | 0.010 | 1.97 (2.06–1.88) |
| Cypermethrin | Susceptible | 328 | 0.873 (0.432–1.767) | 1.017±0.156 | 0.794 | |
| | Chervishevo | 200 | 1.251 (0.834–1.877) | 2.218±0.090 | 0.909 | 3.07 (3.35–2.82) |
| Permethrin | Susceptible | 298 | 1.561 (0.727–3.328) | 1.116±0.168 | 0.016 | |
| | Chervishevo | 980 | 5.707 (3.565–9.137) | 1.444±0.104 | 0.007 | 3.66 (4.90–2.75) |
| Thiamethoxam | Susceptible | 210 | 16.11 (10.61–24.45) | 2.550±0.093 | 0.809 | |
| | Chervishevo | 210 | 20.11 (12.78–31.66) | 2.118±0.101 | 0.913 | 1.25 (1.20–1.29) |
| Fipronil | Susceptible | 397 | 4.073 (2.771–5.988) | 2.399±0.085 | 0.894 | |
| | Chervishevo | 240 | 4.555 (2.933–7.074) | 1.965±0.098 | 0.826 | 1.12 (1.06–1.18) |
| Chlorfenapyr | Susceptible | 457 | 4.308 (2.048–9.060) | 0.891±0.165 | 0.190 | |
| | Chervishevo | 539 | 3.449 (1.983–6.001) | 1.369±0.123 | 0.361 | 0.80 (0.97–0.66) |
| | Susceptible | 440 | 28.29 (17.42–45.96) | 1.565±0.107 | 0.112 | |
| | Chervishevo | 420 | 36.32 (25.32–52.12) | 2.569±0.080 | 0.197 | 1.28 (1.45–1.13) |

N – the number of adult flies used in bioassay including control; Resistance ratio (RR) = LD₅₀ of field-collected population/LD₅₀ of susceptible population. Criteria of susceptibility: RR<1 – high susceptibility to the insecticide, RR=1 – the absence of resistance (the population is sensitive to the insecticide), RR=2-10 – very low resistance (the insects are tolerant to the insecticide), RR=11-20 – low resistance (the insects are moderately resistant), RR=21-50 – medium resistance, RR=51-100 – high resistance (the insects are resistant to the insecticide), RR>100 – very high resistance (the insects are highly resistant to the insecticide) (Drenova *et al.*, 1999; Abbas *et al.*, 2015); LD₅₀ units – µg of active substance per one g of insects' weight; for deltamethrin – in the numerator as FAS, in the denominator as Delcid.

cide residues on glass surfaces (Marçon *et al.*, 2003). In Florida dairy farms, population of flies resistant to pyrethroids have been also found: more than 20-fold resistant to permethrin and 10-fold to the beta-cyfluthrin (Kaufman *et al.*, 2010). In Pakistan, population of flies resistant to pyrethroids have been also found on dairy farms: resistance ratio (at LC₅₀) 30.22–70.02 for cypermethrin and 5.73–18.31 for deltamethrin (Khan *et al.*, 2013).

In Russia, in the Tyumen region, an exceptionally high resistance to pyrethroids during their long-term use in pig and calf barns in Nizhnaya Tavda village was found in the local population of house flies, and a higher resistance was observed to the more effective preparations. In 1999, RR to Butox (deltamethrin 5% EC) was 1600, Fastak (alpha-cypermethrin 10% EC) – 1000, Arrivo (cypermethrin 25% EC) – 800, Stomasanum (permethrin 20% EC) – 200, Baversan (fenvalerate EC) – 50, mixtures of malathion and cypermethrin – 200, permethrin with diazinon – 61 and chlorfenvinphos with alphasmethrin – 45 (Pavlov *et al.*, 2006).

The studied field house fly population did not reveal resistance to thiamethoxam (RR = 1.12), despite the fact that neonicotinoid insecticides are quite common. Researchers from different countries noted the formation of resistance to neonicotinoids in natural populations of the house fly. For example, Kaufman *et al.* (2010) reported the emergence of tolerant and resistant to imidacloprid house fly populations on Florida State dairy farms. According to Khan *et al.* (2015), the flies collected in different areas of Pakistan had different resistance to thiamethoxam: RR (at LC₅₀ values) ranged from 7.66 to 20.13.

Another insecticide, to which the field population of flies caught by us was also

susceptible was chlorfenapyr (RR = 1.28). As can be seen from the results, chlorfenapyr is inferior to fipronil and pyrethroids in toxicity for flies of both lab-susceptible and field populations. In the available literature, there is no information about the formation of house fly populations resistant to chlorfenapyr. This is probably due to the absence of chlorfenapyr containing preparations for combating flies, and its predominant use for plant protection.

Our results show a high susceptibility of the field population to fipronil (RR = 0.80). In Russia insecticides containing fipronil are allowed in the form of gels and poisoned baits (for example, to control cockroaches), and therefore such preparations are not applied to control the fly populations. This is probably the reason why field populations of flies on farms retain susceptibility to fipronil. Given the high toxicity of fipronil for flies and a high susceptibility of the field population of flies to it, exploration and development of a suitable method of fipronil use to reduce the number of flies on livestock breeding complexes is considered promising. It is important to consider the world experience, indicating the possibility of forming resistance to fipronil in house flies after its intensive use (Kristensen *et al.*, 2004; Abbas *et al.*, 2016).

Overall, the results suggest that the population of *M. domestica* on the livestock farm in Chervishevo village, Tyumen municipal region, after many years of usage of pyrethroid insecticides to control flies, retained susceptibility to permethrin and acquired tolerance or very low resistance to deltamethrin and cypermethrin. It is necessary to restrict the use of pyrethroids to fly control. The monitoring of the susceptibility of field *M. domestica* populations to the conventional

and new insecticides is important for more effective of houseflies control on livestock complexes.

From practical point of view, in order to prevent the development of resistance, it is important to use the rotation of insecticides with different modes of action, taking into account the level of susceptibility of the local population to the insecticides used. The rotation scheme can be as follows: organophosphates – neonicotinoids – carbamates – neonicotinoids (Roslavtseva, 2011). According to our results, the flies of field population are susceptible to pyrroles (fipronil) and pyrazoles (chlorfenapyr), therefore, the design of fipronil- and chlorphenapyr-containing insecticidal formulations for flies control and their inclusion in a system of integrated pest management seems promising.

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