

Scientific and technical work

6. Flies

6.1 Insecticide resistance in *Musca domestica*

6.1.1 Laboratory strains kept in 2002

At the end of 2002, DPIL kept 19 strains representing a wide variety of resistance mechanisms and origins for use in testing and research work. A list of the strains and their origins is given in Table 6a. In all these strains, the resistance originated in the field. In several strains, selection with one (or two) insecticide(s) is carried out between one and four times a year in order to maintain the particular resistance type. As has been the case since the beginning of our investigation of resistance in houseflies in 1948, all our strains are available to laboratories that wish to use them for research, development of new insecticides, etc. This has assisted international research on insecticide resistance and given us useful feedback on our resistance problems.

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Table 6a. Laboratory strains of *Musca domestica* maintained during 2002

Strain	Origin	Year	Remarks	Lab pressure
<i>1. Strains subjected to periodic insecticidal pressure (adult dipping, exposure to vapour, or feeding with treated sugar) from a compound to which at least part of the population showed clear resistance at the time of collection</i>				
17 e	DK	1950		lindane
39 m ₂ b	DK	1969		tetrachlorvinphos*
49 r ₂ b	DK	1970		dimethoate*
381 zb	DK	1978		permethrin and dimethoate*
690 ab	DK	1984		methomyl feeding*
594 vb	DK	1988		azamethiphos feeding*
571 ab	Japan	1980	High OP-R	fenitrothion
698 ab	Burma	1985	(not kdr)	DDT
790 bb	DK	1997		diflubenzuron
802 ab	DK	1997		cyromazine
807 ab	DK	1997		diflubenzuron
<i>2. Originally resistant field strains kept without insecticidal pressure</i>				
7	DK	1948	Reverted DDT-R	None
772 a	DK	1989	Common lab. test strain	None
791 a	DK	1997	Multi-R	None
<i>3. Susceptible strains</i>				
BPM	Leiden	1955		None
WHO Ij ₂	Pavia	1988		None
NAIDM	Texas	1991		None
<i>4. Strains with resistance mechanisms isolated</i>				
A ₂ bb	DK	1982	Super-kdr Chr. 1, 2 and 3 with marker genes	None
LPR	USA	1995	Pyr-R kdr, P450 monooxygenase	None

Some resistance to various (other) OP compounds and to DDT

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6.2 Biological and physical control of *Musca domestica* and *Stomoxys calcitrans*

6.2.1 Biological control of houseflies and stable flies with focus on indoor confinements housing animals on straw bedding

Based on previous studies (see Annual Report 2000) the parasitoid, *Spalangia cameroni* Perkins (2-3 mm in size) was identified as a suitable candidate for biological control of houseflies and stable flies.

The effect on parasitism of releasing 50, 100 and 200 female *S. cameroni* per m² in straw-bedded areas of six organic farms in the early period of the fly season (April-May) and middle part of the fly period (June-August) was evaluated. The aim was to examine if a relationship exists between release density and the number of fly pupae parasitized, and to evaluate the effect of the season on parasitoid activity. As control for activity of naturally occurring parasitoids, sentinel pupal bags were exposed in the bedding a week before the release of *S. cameroni*. Parasitism was measured by means of sentinel pupal bags from the day of release and weekly for four weeks.

Early in the fly season, the overall parasitism of *S. cameroni* was low ($10.5 \pm 1.6\%$ SE) and not related to release rates. In June, the average parasitism increased significantly across treatments to $19.7 \pm 2.9\%$ SE, mainly due to an increase in ambient temperature, ($F_{1,59}=4.24$; $P<0.04$). The highest release rate, 200 female parasitoids per m² led to the highest level of parasitism although not significantly different from 50 and 100 females per m² ($F_{2,23}=0.38$; $P<0.6888$). Further, a significant decline in parasitism was found following the second week of the release of parasitoids. Thus, massive releases of *S. cameroni* early in the fly season (April-May) may have little overall effect on the fly development, since most of the released individuals will be inactive because of relatively low outdoor temperatures (12-15°C). Later in the season, parasitoid activity will increase significantly, due to a raise in temperatures, though a high release rate of parasitoids per m² in the present study did not result in a correspondingly higher percentage of fly pupae killed. Approximately 200 female *S. cameroni* per m² of straw bedding seem to be the maximum when related to the cost of producing parasitoid. Furthermore, as suggested previously (see Annual Report 2001) releases of *S. cameroni* should be made biweekly due to a high death rate of released individuals.

Another field experiment with biocontrol of housefly and stable fly populations was conducted on six organic dairy cattle farms in the period August-November where approximately 200 female *S. cameroni* per m² of straw bedding were released biweekly. The aims of the study were:

- i) To examine whether *S. cameroni* could suppress the fly populations below a nuisance level given that the onset of control occurred when the flies were abundant in the stables.
- ii) To examine the relative impact of temperature, pupal parasitism and infection of adult flies with Entomophthorales (see section 6.3.4 for *Entomophthora*) on the population dynamics of housefly populations in the period August and until primo December.

Six organic dairy cattle farms with a history of high housefly and stable fly densities were selected for the study and split up into one group of three farms receiving parasitoids (200 females per m²) and the other group was kept as controls. Activity of released *S. cameroni* including naturally occurring parasitoids was measured by exposing laboratory-reared housefly pupae in sentinel bags in the stables. Furthermore, the direct effect of *S. cameroni* on the fly populations was measured using a pseudologarithmic scale defined by the DPII where index=0 denotes 0-3 flies per animal and index=7 denotes 200-400 flies per animal.

The final outcome of the control part (i) still has to be thoroughly analysed, but it seems that the suppressive effect of *S. cameroni* is rather limited when the populations of houseflies and stable flies have already reached high levels. However, in a previous study albeit covering one dairy cattle farm only, especially the

housefly population was brought down from index=7 to index=2 within one month by *S. cameroni*. In that study about 800-1000 female *S. cameroni* were released per m², a release rate four- to five-fold higher than in the 2002 study.

If *S. cameroni* is released early in the fly season where the populations of houseflies and stable flies are still low, less than 200 female *S. cameroni* per m² seem sufficient to maintain the populations of especially the housefly at acceptable, low levels throughout the fly season (see annual Report 2000). In contrast if control of the flies is initiated late (July-August) when the flies build up high population levels, release rates of approx. 1000 females *S. cameroni* per m² seem more appropriate, but not commercially feasible.

H. Skovgård

6.2.2 Life-table studies of a pupal parasitoid *Urolepis rufipes*

In August 2002, Lena Steenseng defended her M.Sc. entitled “Life-table studies of the pupal parasitoid *Urolepis rufipes* (Hymenoptera: Pteromalidae) on the housefly *Musca domestica* (Diptera: Muscidae)”. The manuscript included in the report has been accepted for publication in a refereed journal.

H. Skovgård and L. Steenseng

6.2.3 Hyphomyceteous fungi

Hyphomyceteous fungi are considered as potential biocontrol agents against adult and immature stages of houseflies and the compatibility of the entomopathogenic fungus *Metarhizium anisopliae* and the pupal parasitoid *S. cameroni* was evaluated in the laboratory (see Annual report 2000 for further description). The susceptibility of female parasitoids to fungus infection as well as the effect on female reproduction, survival, and population dynamics was determined.

The isolate of *M. anisopliae* was moderately virulent at high conidia concentrations (1×10^8 conidia ml⁻¹) to the parasitoid, causing 68% mortality after 6 days. When *S. cameroni* was provided with optimal reproductive conditions, no overall effect of fungal treatment on reproduction was found ($F_{3,73} = 0.684$, $P = 0.508$), although treatment with 1×10^8 conidia ml⁻¹ caused 50% infected females. Furthermore, the physiological susceptibility of the parasitoids to the fungal treatment was not expressed in the lifetable parameter r_m , compared with a control group, which seems to indicate that the isolate of *M. anisopliae* would also be compatible with *S. cameroni* under field conditions.

T. Steenberg and C. Nielsen

6.2.4 *Entomophthora muscae* and *E. schizophorae* in houseflies and stable flies

The prevalence of *E. muscae* and *E. schizophorae* in houseflies was determined on six farms on a weekly basis from August to November as part of the study mentioned in 6.2.1. Maximum infection rates varied from 38% to 78% with no general trend of increasing prevalences with increasing fly populations. Males were infected more often than females. *E. schizophorae* was recorded on three farms in late July, and on all six farms later in the season; this species infected up to 25% of the specimens killed by Entomophthoralean fungi. Cadaver counts were made on vertical surfaces on all farms also on a weekly basis. Despite the finding of a few specimens of fungus-infected cadavers of *Drosophila* sp. and *Stomoxys calcitrans*, subsequent sampling of live flies of the two species did not reveal fungus infection in these populations.

In September, an unusually high number of sporulating cadavers of *Stomoxys calcitrans* was noticed on a dairy farm in Jutland. Incubation of live stable flies and houseflies showed that both species were infected by *E. muscae*, with prevalences of approx. 30%. The fungi have been isolated *in vitro* in liquid medium and will be screened for their ability to produce infective spores from cultures transferred to solid substrate (egg-

yolk medium). Transmission experiments using *in vivo* strains from the two fly species are under way to study the host specificity not only on species level but also in different fly strains.

Tove Steenberg