

Effects of a Juvenile Hormone Analogue on the Biology of the Stable fly

(*Stomoxys calcitrans* (L.) Diptera : Muscidae)

Chitapa Ketavan and Pichitra Kanjanamungsuk¹

ABSTRACT

The biological studies of *Stomoxys calcitrans* (L.) were conducted under laboratory conditions those of $25 \pm 1^\circ \text{C}$, $88 \pm 1\% \text{RH}$. and $31 \pm 3^\circ \text{C}$, $76 \pm 4\% \text{RH}$. The adults were fed on cow blood while the larvae were reared on cow blood and cow dung. The mean duration of the life cycle under the two conditions were slightly different, 27 days and 28 days respectively. Larval development was not significantly different under the two laboratory conditions.

Upon feeding on cow dung mixed with various concentrations of JHa (2-methoxy-9-(4-isopropyl phenyl) -2, 6-dimethylnonane, (MV-678)), all the larvae were found dead at a concentration of 100 ppm while a certain number of larvae were able to enter pupal stage at 10, 1 and 0.1 ppm but no adult emergence. However, after dissecting these puparia, the pupal adult intermediate forms were found. The 0-hour-old puparia treated with MV-678 at different concentrations revealed the same result. The LC_{50} of the compound for the adult male and female when subjected to topical application were 0.589 and 0.708 microgram respectively.

INTRODUCTION

The juvenile hormone analogue (JHa) or insect growth regulator is considered as the "third generation insecticide". Up till now, very few insects have developed resistance to this kind of insecticide, for instance, certain strains of houseflies (Cerf and Georghiou, 1972). However, each JHa is intended to affect specifically a certain species of insect for example Stauffer R-20458, (E)-6, 7-epoxy-1-(p-ethyl-phenoxy)-3, 7-dimethyl-2-octene was active against stable fly but not the house fly (Wright 1972, Wright & Spates 1972 a & b, Wright *et al.* 1974). The methoprene, isopropyl (E, E)-11-methoxy-3, 7, 11-trimethyl-2, 4-dodecadienoate) was active against both stable fly and house fly in the laboratory but not applicable in the field. (Wright *et al.* 1973).

According to its specificities to target pests, safety for fish and wildlife, non-persistence in the environment and biodegradation, the JHa has been recently developed for agricultural uses. However, the advance development in effective

control by JHa in Thailand has been retarded mainly by the limited knowledge of both in insect biology and activity of the compounds. This investigation was mainly to fulfill and develop the method of stable fly control since they are one of the most serious pest of livestock in this region.

MATERIALS AND METHODS

The biological studies of stable fly (*Stomoxys calcitrans* (L.)) were conducted under two conditions, at $25 \pm 1^\circ \text{C}$, $88 \pm 1\% \text{RH}$. in the incubator and under laboratory conditions at $31 \pm 3^\circ \text{C}$, $76 \pm 4\% \text{RH}$. The adults were fed on cow blood² while the larvae were reared on cow blood and cow dung. The oviposition period, total number of eggs laid and the duration of development were recorded.

The juvenile hormone analogue (JHa), 2-methoxy-9-(4-isopropyl-phenyl)-2, 6-dimethylnonane : MV-678, was supplied by Stauffer Chemical Co. San Francisco as a technical grade of

¹ Dept. of Entomology, Faculty of Agriculture, Kasetsart Univ .

² To avoid blood coagulation, 3 gm of sodium citrate was added to 1,000 ml of blood, when stored at 4°C the blood could be usable up to 7 days.

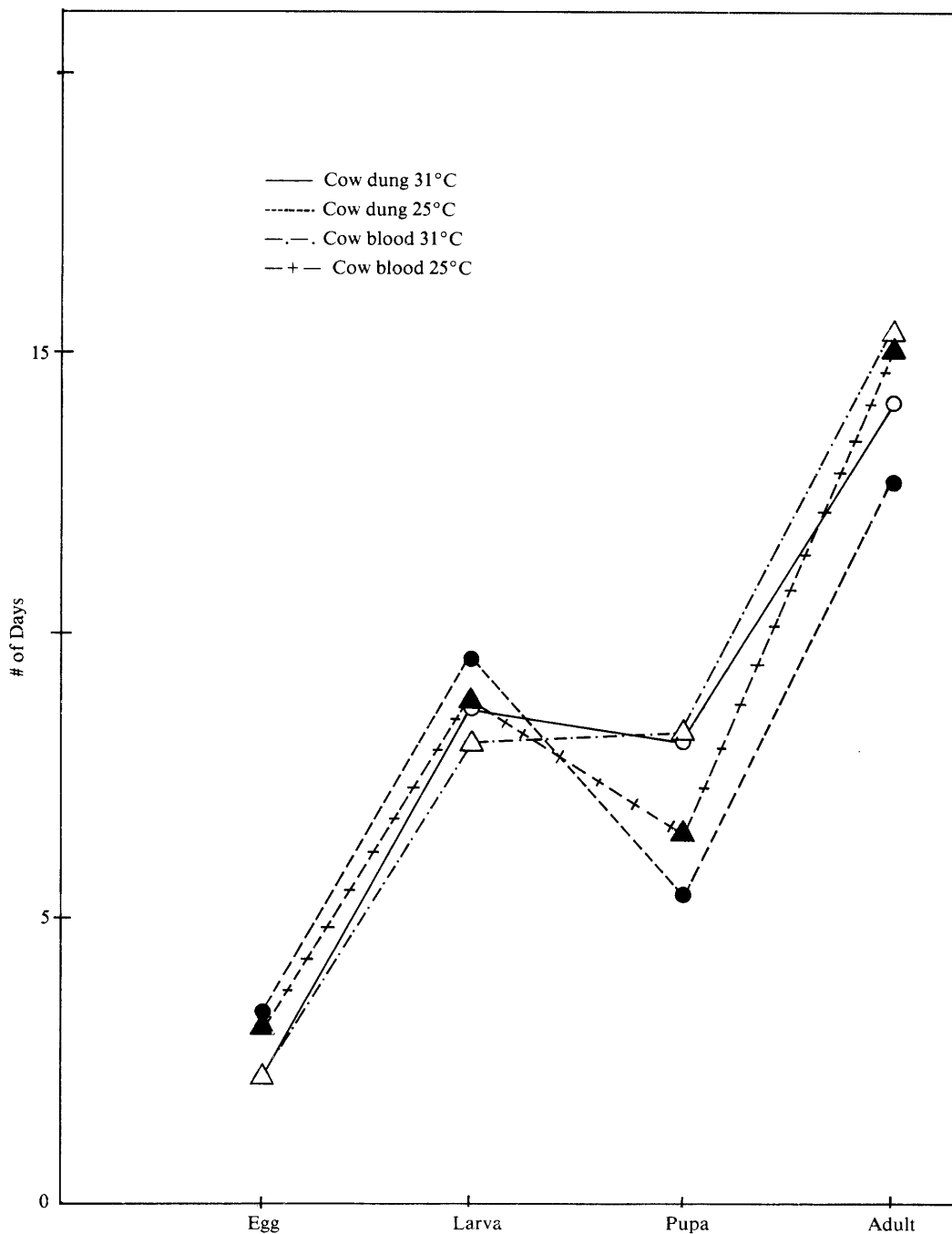


Figure 1 : Development time for each stage of the male *S. calcitrans*.

93.3 wt % (80.2% para, 13.3% ortho). For evaluation of this analogue it was placed in the larval rearing medium at different concentration (in a solution of acetone). Also it was applied topically to both puparia and adults under laboratory conditions.

The MV-678 was incorporated into the larval diet, 20 gm of dry cow dung, at the rates of 100, 10, 1 and 0.1 ppm. After 24 hours, the newly-hatched larvae were placed in these media, using 25 larvae for each concentration. The percentage of inhibition of adult eclosion was observed 2 weeks later. In addition, the 0-hour-old

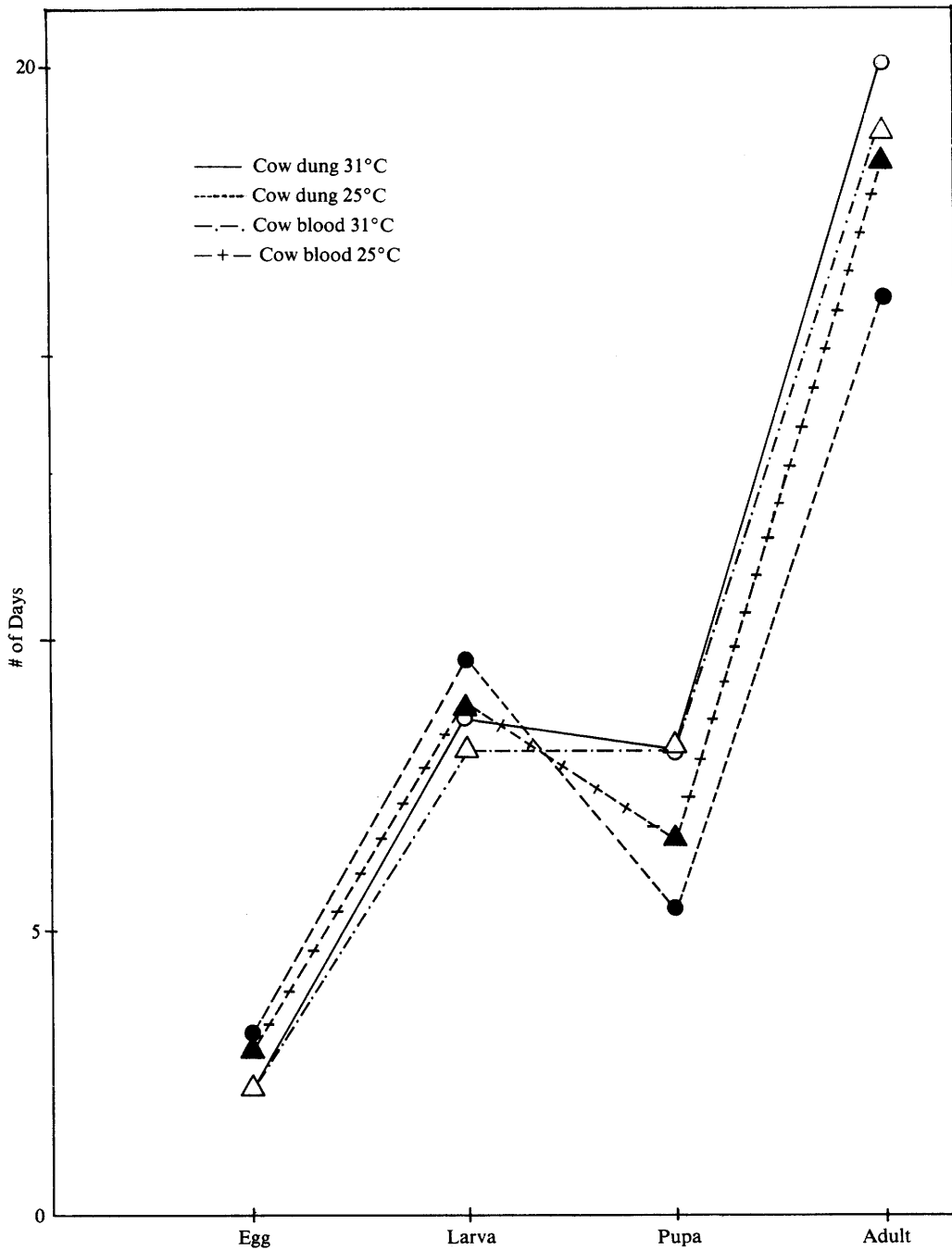


Figure 2 : Development time for each stage of the femela *S. calcitrans*.

puparia were treated topically with 1 μ l of concentration of 10, 1, 0.1, 0.01 and 0.001 ppm, the eclosion was checked after 10 days. Puparia from which no adult eclosed were dissected to determine whether pupal-adult intermediates were present. The determination of percentage

of inhibition was based on the number of pupal-adult intermediates relative to the total samples.

In the 3rd type of test, the MV-678 was applied topically to the 6-hour-old adult with 1 μ l concentration of 10, 1, 0.01 and 0.001 ppm. The mortality was checked every 24 hours for 7 days and up to 20 days for the adult longevity.

Table 1. Average size of *S. calcitrans* (L.)

	Size (mm)			
	31°C 76% RH		25°C 88% RH	
	Cow Dung	Cow Blood	Cow Dung	Cow Blood
Full grown larva	10.27 ± 0.408	10.20 ± 0.438	10.264 ± 0.466	10.233 ± 0.476
Puparium	5.26 ± 0.107	5.23 ± 0.095	5.31 ± 0.137	5.29 ± 0.137
Male	6.64 ± 0.279		6.53 ± 0.286	
Female	7.01 ± 0.296		7.09 ± 0.357	

Table 2. Developmental period in 1 generation of *S. calcitrans* (L.)

Larval Media	Temperatures (°C)	Mean Development (Days)
Cow Dung	25 ± 1	27.9
	31 ± 3	28.6
Cow Blood	25 ± 1	27.6
	31 ± 3	28

Table 3. Percent survival of *S. calcitrans* (L.) in different diest and conditions.

Larval diests	% survival	
	25 ± 1°C, 88 ± 1% RH	31 ± 3°C, 76 ± 4% RH
Cow blood	44.75	16.25
Cow dung	57.25	20.25

RESULTS AND DISCUSSION

Growth and Development

The mean duration of stable fly development at 25 ± 1°C, 88 ± 1% RH. and 31 ± 3°C, 76 ± 4% RH. using two different larval media are presented in fig. 1 and 2. Female longevity was slightly longer than those of the male, i.e., 1-31 days for the female and 2-27 days for the male. The size of egg, larva, pupa and adult is shown in Table 1. The developmental period in 1 generation under the two conditions were about the same, 28 days (Table 2).

The total number of eggs oviposited per female obtained in this investigation was only 204, when using cow dung as the larval media. Parr (1962) found that one female oviposited 376 eggs in the same media mixed with some

blood and sugar. Moreover, the data obtained (Figs. 3 & 4) indicates that the media and temperature played an important role in the percentage of hatching and survival. It was observed that the percentage of survival from egg to adult was significantly higher in the larvae reared on cow dung at 25 ± 1°C, 88 ± 1% RH. (Table 3). This result was very similar to those reported by Bailey *et al.* (1975).

The pre-oviposition and the oviposition period of female in two different diests and conditions were not significantly different.

To obtain the maximum quantity of larvae, it was suggested to use the cow dung as the larval medium and the optimal conditions for rearing stable fly should be 25°C with approximately 88% RH. During the oviposition period in this

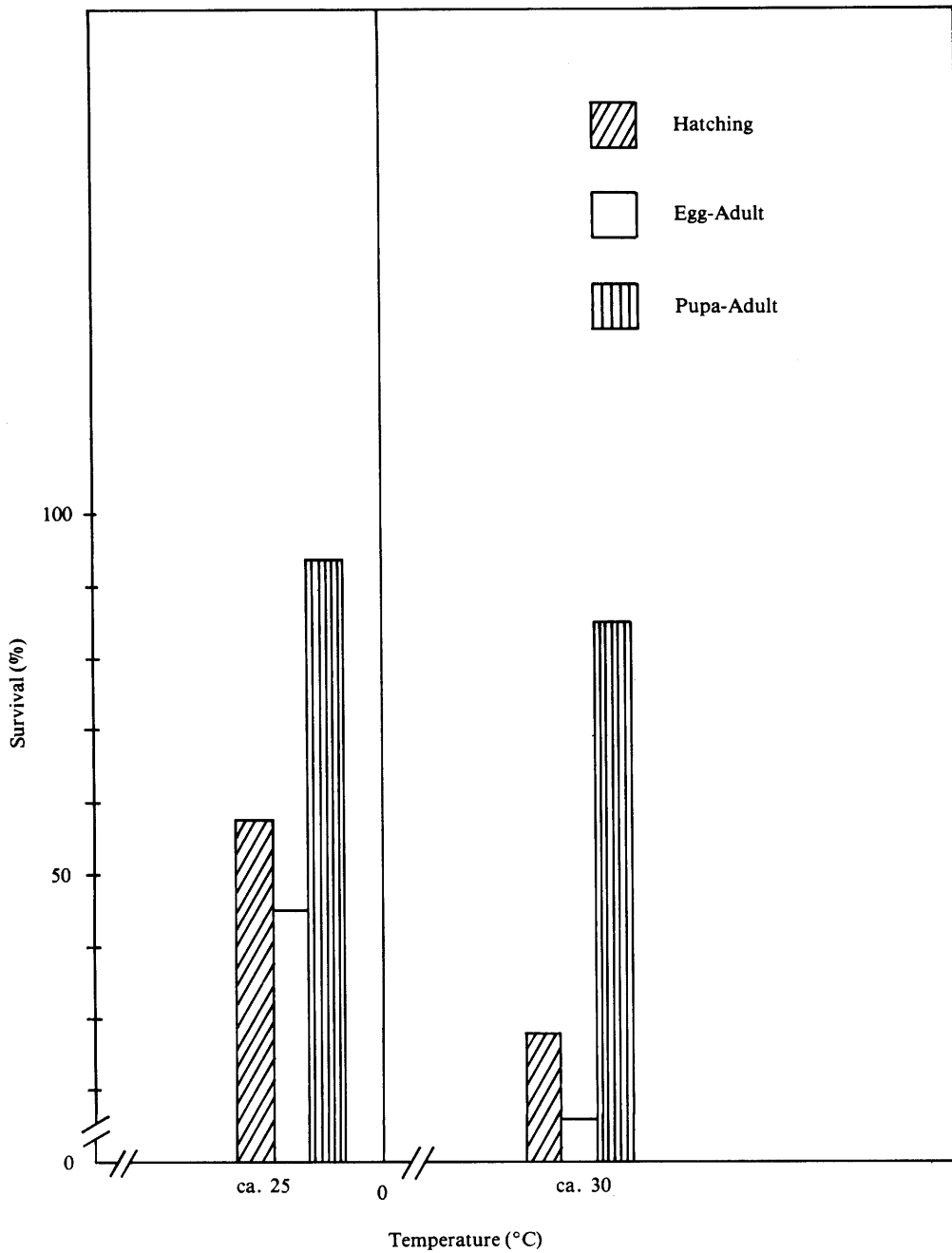


Figure 3 : Survival of *S. calcitrans* reared on cow dung.

experiment, cotton soaked with NH_4OH was placed in the rearing cage to act as an oviposition stimulant because in nature the female flies were very attractive to the animal urea.

Two hymenopteran pupal parasites were obtained from the stable fly colony reared in the laboratory, i.e. *Spalangia endius* Walker and *Pachycrepoideus vindemiae* (Rondani) as well

as predator mite : *Macrocheles muscaedomesticae* (Scop.). This mite was reported as the egg and larval predator (Kinn, 1966 ; Williams and Roger, 1977).

The evaluation of MV-678 :

Percent inhibition of the pupal stage when newly hatched larvae are treated with MV-678 is shown in Table 4.

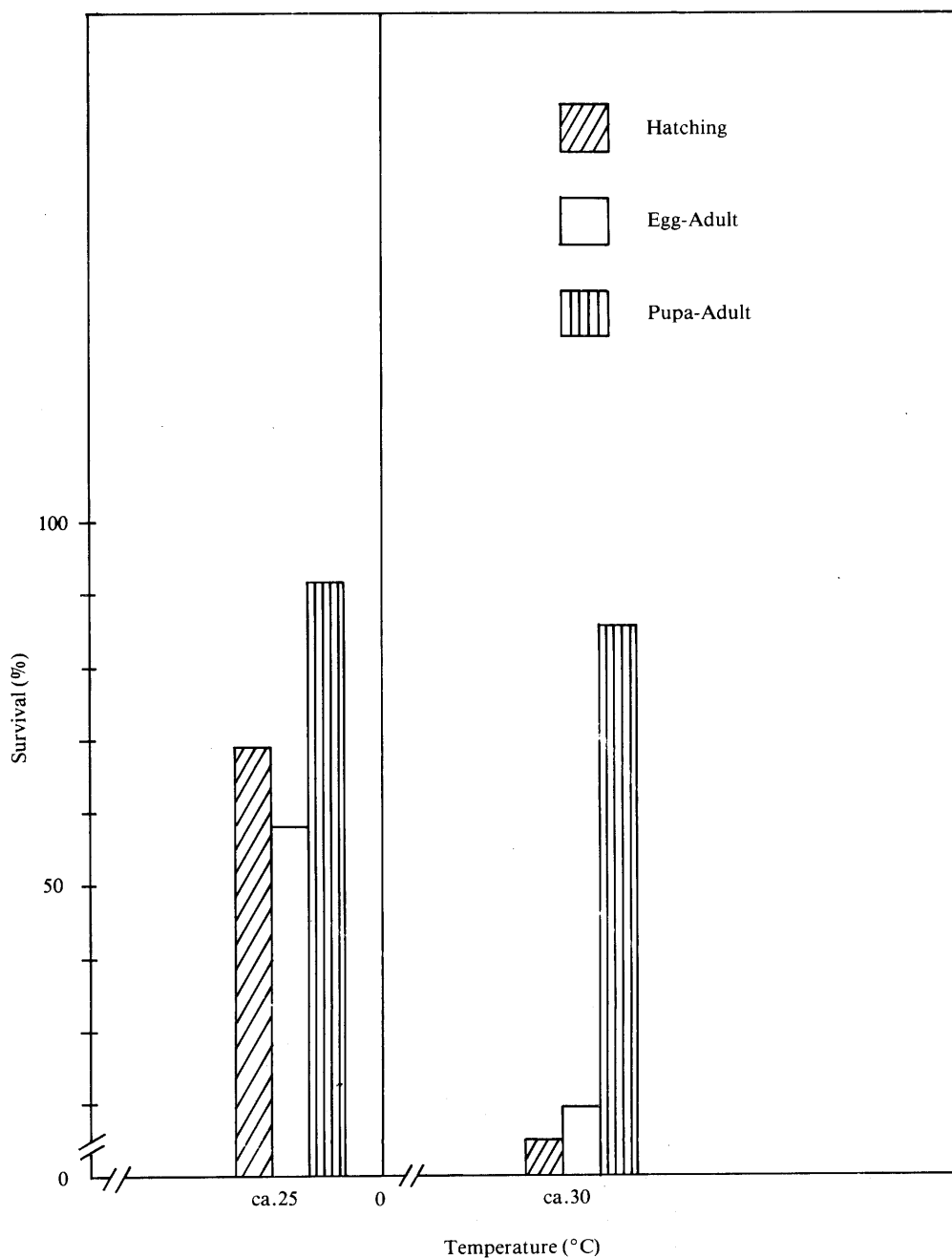


Figure 4 : Survival of *S. calcitrans* reared on cow blood.

These data have indicated that the JHa (MV-678) was highly active against *S. calcitrans* (L.) since there were no adult eclosion from this experiment. All larvae were found dead at the concentration of 100 ppm. At the lowest concentration of 0.1 ppm., the percentage of pupae obtained seemed to be relatively high, 91.67% but there was no adult eclosion. The results obtained from these experiments are similar to those

reported by Schwarz *et al.* (1974). The weight of the newly-formed puparia (white pupal stage) varied directly with the concentration of JHa, those subjected to the concentration of 0.1 ppm were the highest. (Table 4).

Dissection of the puparia from which no adult emerged revealed that pupal-adult intermediates had developed. This morphological stage consists of the head and thorax with adult

Table 4. *S. calcitrans* (L.) percentage of pupation of treated larvae.

Concentration (ppm.)	Pupation (%)	Ave. Weight of Puparia (mg.)
100	0	0
10	21.67	9.17
1	83.33	10.40
0.1	91.67	11.15
Acetone	100	8.90

Table 5. Mortality of *S. calcitrans* (L.) treated with MV-678, and observed after 7 days of exposure.

Concentrations (ppm)	Mortality (%)*	
	Male	Female
10	89.65**	81.48
1	51.72	48.41
0.1	41.38	33.33
0.01	34.49	29.63
0.001	13.80	11.11
Acetone	0	0

* Corrected by Abbott's Formula

** Highly significance

characteristics such as compound eyes, ocelli and antennae as well as the setae. However, the thorax and abdomen were morphologically that of a pharate pupa, light brown in color and no longitudinal line on the thorax. The abdomen was twisted and unsegmented.

None of the adults emerged when puparia were treated topically with MV-678 at every concentration. At 10 ppm all pupae were found dead in the puparia and showed no sign of growth. At the other 4 lower concentrations, the pupal-adult intermediates were observed with the same characteristics as mentioned above.

The mortality of treated adults were significantly high in the male at the concentration of 10 ppm. (Table 5). The LC_{50} for adult male and female calculated from Read and Muench method were 0.589 and 0.708 μg respectively. No treated adults were found alive after 7 days.

The MV-678 seemed to be highly effective against growth and development of stable fly

since no adults eclosed from both treated larvae and pupae in this investigation. However, the pupal-adult intermediates were observed when exposed to the lower concentrations.

According to Wright & Schwarz (1972) the results confirmed that MV-678 could inhibit the growth activity of newly-hatched larvae and the 0-hour old puparia or white pupal stage of stable fly. Since the effective dose of MV-678 used in this experiment was not too high, only 0.1 ppm, and due to its selective activity, this experiment strongly demonstrated that the efficiency of JHa was capable in controlling the livestock pest as described by Wright and Spates (1972a and b). However, the application of JHa in Thailand is quite new and this is only the preliminary laboratory study and field trials should be conducted in order to find the best applicable measure in the future.

LITERATURE CITED

- Bailey, D.L., T.L. Whitfield and G.C. LaBrecque. 1975. Laboratory biology and technique for mass producing the stable fly, *Stomoxys calcitrans* (L.) (Diptera : Muscidae). J. Med. Entomol. 12 (2) : 189-193.
- Cerf, D.C. and G.P. Georghiou. 1972. Evidence of cross-resistance to a juvenile hormone analogue in some insecticide resistant house flies. Nature 239 : 401-402.
- Kinn, D.N. 1966. Predation by the mite, *Macrocheles muscaedomesticae*. (Acarine : Macrochelidae), on three species of flies. J. Med. Entomol. 3 (2) : 155-158.
- Parr, H.C.M. 1962. Studies on *Stomoxys calcitrans* (L.) in Uganda, East Africa: II Notes on life-history and behavior. Bull. Entomol. Res. 53 (2): 437-443.
- Schwarz, M., R.W. Miller, J.E. Wright, W.F. Chamberlain and D.E. Hopkins. 1974. Compounds related to juvenile hormone exceptional activity of arylterpenoid compounds in four species of flies. J. Econ. Entomol. 57 (5): 598-601.
- Williams, D.F. and A.J. Rogers. 1977. Stable flies infested with the mite *Macrocheles muscaedomesticae*. Rev. Appl. Entomol. Ser. B. 65: 501.
- Wright, J.E. 1972. Hormones for control of livestock arthropods : Effectiveness of three juvenile hormone analogues for control of stable flies. J. Econ. Entomol. 65 : 1361-1364
- Wright, J.E., J.B. Campbell and P. Hester. 1973. Hormones for control of livestock arthropods. Evaluation of two juvenile hormone analogues applied to breeding materials in small plot tests in Nebraska and Florida for control of the stable fly. Environ. Entomol. 2: 69-72.
- Wright, J.E., T.P. McGovern, R. Sarmiento and M. Beroza. 1974. Juvenile hormone activity of substituted arly 3, 7-dimethyl-6-octenyl ethers in the stable fly and house fly. J. Insect Physiol. 20: 423- 427
- Wright, J.E. and M. Schwarz. 1972. Juvenili- zing activity of compounds related to the juvenile hormone against pupae of the stable fly. J. Econ. Entomol. 65 (6): 1644-1647.
- Wright, J.E. and G.E. Spates. 1972a. A new approach in integrated control : insect juvenile hormone plus a hymenoptera a parasite against the stable fly. Science. 178: 1292- 1293.
- Wright, J.E. and G.E. Spates. 1972b. Labora- tory evaluation of compounds to determine juvenile hormone activity against the stable fly. J. Econ. Entomol. 65: 1346- 1349.
- Wright, J.E. and P.E. Sonnet. 1973. Juvenile hormone activity of citronellylamine and citronellol derivatives against pupae of the stable fly and the house fly. J. Med. Entomol. 10: 477- 480.