Onderstepoort J. vet. Res. 45, 209–212 (1978)

NUTRITIONAL VALUES OF DIFFERENT BLOOD DIETS EXPRESSED AS REPRODUCTIVE POTENTIALS IN ADULT *STOMOXYS CALCITRANS* L. (DIPTERA: MUSCIDAE)

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ABSTRACT

SUTHERLAND, B., 1978. Nutritional values of different blood diets expressed as reproductive potentials in adult *Stomoxys calcitrans* L. (Diptera: Muscidae). *Onderstepoort Journal of Veterinary Research* 45, 209–212 1978.

Experiments with *Stomoxys calcitrans* adults showed that different blood diets markedly affect the lifespan and reproductive potential of this species. When fed on blood from herbivores (cattle, sheep, goat, horse and donkey) the adults lay more eggs than they do when fed on blood from omnivores (pig). Blood from carnivores (dog) is even less suitable than that from omnivores, and no eggs are laid when the flies are fed on chicken blood.

Résumé

VALEURS NUTRITIVES DE SANGS DIFFÉRENTS, EXPRIMÉES EN TANT QUE POTENTIEL REPRODUCTEUR DES ADULTES DE STOMOXYS CALCITRANS *L. (DIPTERA: MUSCIDAE)*

Des expériences faites sur des Stomoxys calcitrans *adultes ont montré que la diversité des sangs constituant leur régime alimentaire a une nette influence sur la longévité et le potentiel reproducteur de cette espéce. Quand on les nourrit de sang d'herbivores (gros bétail, moutons, chèvres, chevaux et ânes) les adultes pondent plus d'oeufs que lorsqu'ils ont reçu du sang d'omnivores (porcs). Le sang des carnivores (chiens) est encore moins approprié que celui des omnivores, et les mouches ne pondent pas d'oeufs quand on les a nourries de sang de poulet.*

INTRODUCTION

In the adult stage, *Stomoxys calcitrans* is an obligatory ectoparasite, both sexes of which feed on blood, and extensive host lists for different parts of the world have been compiled by various research workers (Newstead 1906; Bishopp, 1918; Hafez & Gamal-Eddin, 1959b; Anderson & Tempelis, 1970; La Brecque, Meifert & Weidhaas, 1972; Monty, 1972; Howell, Walker & Nevill, 1978). Du Toit (1975) states that the stable fly, under South African conditions, will parasitize 'practically all domestic animals'. It can also become a pest in zoos. In the Pretoria Zoological Gardens, for example, the bites of stable flies have caused allergic reactions, with resultant epidermal sores, on polar bears (*Thalarctos maritimus*) (Sutherland, unpublished data, 1976), and at the Johannesburg Zoological Gardens a young eland (*Taurotragus oryx*) died after persistent attacks by large numbers of this fly (L. P. Colley, personal communication, 1976). Stable flies can also act as vectors of numerous pathogenic organisms (Zumpt, 1973).

There is little evidence in the literature that this fly feeds on birds under natural conditions. Fuller (1913) records it from chickens, but Anderson & Tempelis (1970), who worked on Californian poultry ranches, rarely found it parasitizing hosts other than mammals.

Stable flies apparently cannot live and reproduce normally unless they feed on blood. Hafez & Gamal-Eddin (1959b) found that flies, fed on a 10% sugar solution, had a 50% mean survival time of only 3 days at 25 °C and, moreover, the ovarian follicles of the females did not develop beyond Stage II, as classified by Detinova (1962). Smit (1964) noted that a female fly could lay her first batch of eggs after 3 blood meals, but Anderson & Tempelis (1970) claimed that some females took up to 7 blood meals before starting to oviposit. Du Toit (1974) found that adult stable flies, when fed simply on a manure filtrate, lived for only 2 days but, when given a 5% sugar solution, their mean survival time increased slightly to 2,5 days.

Received 9 May 1978—Editor

Little is known about the comparative nutritional value of blood from different hosts. Schoof (1964) and Jones (1966) listed guinea-pigs, mice and sheep as having 'unsuitable' blood. They quoted Pospisil (1961), however, as saying that the blood of cattle, horses, sheep and pigs is 'suitable', but unfortunately they did not give the parameters on which they based their classification. Du Toit (1975), who referred to longevity in terms of 'fly days', used the total egg production per female as one of his parameters. He found that bovine blood was more suitable than that from ovines, and this in turn was better than pig blood.

In the following investigation, the suitability of a given blood diet is expressed in terms of its *reproductive potential*. This can be defined as the expected total number of eggs produced under optimum conditions by 100 *S. calcitrans* females, taking into account variables such as pre-oviposition mortality and the number of viable eggs produced per female during her lifespan.

MATERIALS AND METHODS

The stock colony was maintained and all experiments were conducted in an incubator room at 27 °C (±1°), with a relative humidity between 60%–80% and a photoperiod with an 8-hour light and 16-hour dark cycle. All experiments commenced with teneral flies less than 18 h old. Each group of flies was kept separately in a 0,027 m^3 gauze cage and was provided daily with a specific blood diet and an oviposition dish. The latter was a flatbottomed glass vial, 30,0 mm in diameter and 50,0 mm high, 75% filled with a well-fermented mixture of chopped lucerne 800 g, wheat bran 200 g and water 2 000 mℓ. This medium was tightly wrapped in a square of black cotton material to prevent the females from burying their eggs in it and was kept damp to prevent the eggs from drying out. At least 200 individuals of both sexes, divided into 3 more or less equal groups, were used when evaluating each of the diets.

Dead flies were removed from the cages daily and sexed. The onset of the oviposition period was noted and subsequently the eggs were removed and counted daily. Egg viability was determined by regularly transferring newly laid eggs onto wet filter paper in

Petri-dishes, then putting them in the incubator room to hatch. At least 300 eggs were used to evaluate the viability of eggs laid by females on a given blood diet. If the experimental flies did not produce enough eggs, the entire procedure was repeated with a large batch of flies whose eggs were used only to determine hatchability. The ovaries of some of the females in the latter groups were dissected and examined for gonotrophic development.

The blood used in all the different diets was collected at random from slaughtered animals at abattoirs or by venepuncture from robust donor animals that had not been exposed to any chemical treatment that could affect the flies. Because of possible variations in the blood composition of animals belonging to the same species, blood from several individuals was used to make up each diet. Coagulation was prevented by adding 3,0 g of sodium citrate per litre. Since this amount of sodium citrate was insufficient to prevent minor clots from forming in chicken blood, which is fairly rich in calcium, the blood was blended in a standard homogenizer for 5 min at 10 000 rpm. The blood used in the diets was stored in an ordinary refrigerator at $\pm 4°$ C, agitated gently before use and discarded if not used within 10 days.

Cumulative daily mortality rates were plotted on graph paper and the MT_{50} and MT_{100} values determined. The *MT values* refer to the mean time lapse (days) required to achieve a natural mortality of either 50% or 100%, whichever the case might be. *Pre-oviposition mortality* was taken as the natural mortality amongst females in a given test cage up to the time when egg laying started. Since these flies become very excitable on handling, the gravid females were not caged separately and *the egg production per female* was calculated by dividing the total egg harvest from a given cage by the number of live females present during the oviposition period. The oviposition period itself was taken as the time-interval from the beginning of egg laying to the death of the last female (Du Toit, 1974).

An ordinary analysis of variance was done on all the experimental data for a one-way classification model in which all the observations were independent of each other and an F-value was calculated for significance or non-significance between the various treatments. For the least significant differences between possible pairs of treatments, Bonferroni's multiple comparison procedure was used (Miller, 1966).

RESULTS

The effects of 8 different blood diets on adult *S. calcitrans* are listed in Table 1. In the following discussion, differences at the 99% probability level (P=0,01) will be referred to as 'very significant' and at the 95% probability level (P=0,05) as 'significant'.

MT_{50} and MT_{100} mortality levels

The different blood diets had very little effect on the MT_{50} mortality level in adult *S. calcitrans*, the only significant difference being that males lived longer on a diet of cattle blood than on dog blood.

At the MT_{100} mortality level the males again lived significantly longer on cattle blood than on dog blood, and on goat than on dog blood. They had a very significantly longer lifespan on a diet of donkey blood than on dog or chicken blood, and on goat than on chicken blood.

Very significant differences occurred between the MT_{100} levels of both female and adult (i.e., males and females) flies fed on cattle, donkey and goat blood and those fed on dog or chicken blood. The females also lived significantly longer on pig or horse blood than on chicken blood. A diet of chicken blood gave a significantly shorter lifespan than horse, sheep or pig blood.

Pre-oviposition mortality and pre-oviposition period

Pre-oviposition mortality was very significantly lower in flies fed on cattle or sheep blood than on goat or dog blood, while on horse blood it was significantly lower than on dog blood.

The pre-oviposition periods of females fed on donkey and goat blood were very significantly longer than that of females fed on cattle blood. The former 2 diets also gave significantly longer pre-oviposition periods than sheep blood.

TABLE 1 The effect of different blood diets on the mortality and reproduction of adult *Stomoxys calcitrans*. The data listed are the means of the parameters in each treatment

Blood diet	Mortality (MT)						Reproduction				
	MT_{50} (Days)			MT_{100} (Days)			Pre-oviposition		Eggs		M.R.P.*= $\frac{100-(P\times E\times V)}{100}$
	Male	Female	Adult	Male	Female	Adult	Mortality (%) (P)	Period (Days)	Number (E)	Viability (%) (V)	
Avian:											
Chicken.....	4,4	3,9	4,1	11,0	9,7	10,3	100	—	—	—	0
Carnivore:											
Dog.....	3,0	3,3	3,2	11,3	10,7	11,0	90	7,5	4,2	91	23
Herbivore:											
Cattle.....	8,7	8,2	8,5	15,7	15,3	15,5	32	5,3	58,4	98	3 614
Donkey.....	7,7	6,2	7,5	16,7	16,0	16,3	66	9,3	31,6	95	977
Goat.....	4,9	4,1	4,5	16,3	15,3	15,8	78	9,3	24,8	94	496
Horse.....	5,9	6,4	6,1	14,3	13,7	14,0	63	8,0	9,8	91	327
Sheep.....	6,6	6,6	6,6	14,7	13,3	14,0	46	6,0	25,2	85	1 132
Omnivore:											
Pig.....	6,1	5,7	5,9	15,0	14,0	14,5	65	7,7	4,8	98	153

* M.R.P.=Mean reproductive potential

Number of eggs

The females produced few eggs when fed on pig or dog blood and none on a diet of chicken blood. Examination of the ovaries of the females fed on chicken blood showed that the most mature ovarian follicles had only developed to Stage III, as classified by Detinova (1962). At this stage the follicles apparently resorb the yolk and thus prevent egg production. De Wilde (1964) defined this type of sterility as *nutrimentary castration,* 'which is known to occur in many insects as a consquence of lack of food in the adult (Wigglesworth, 1948) or of deficiency of a specific nutritional element (Grison, 1957)'.

The egg production of *S. calcitrans* females was very significantly the greatest when they were fed on cattle blood. Very significantly more eggs were produced on a diet of donkey blood, and significant y more so on diets of sheep or goat blood than on dog blood.

Viability of the eggs

At the 95% probability level, the viability of the eggs produced by *S. calcitrans* on diets of cattle and pig blood differed significantly from that of eggs produced by flies fed on sheep blood. There were no significant differences in egg viability when flies were fed on dog, donkey, goat or horse blood.

Reproductive potential

A diet of cattle blood gave a very significantly higher reproductive potential than that of goat, horse, pig or dog. The same significant difference occurred between diets of donkey or sheep and pig or dog blood. A very significant difference exists between diets of goat, horse and dog blood, and a significant difference between diets of pig and dog blood.

Thus the reproductive capacity of *S. calcitrans* females was clearly higher on diets of blood f o n herbivores than from omnivores. Hardly any reproduction occurred when flies were fed on carnivore blood, and none when they were fed on chicken blood.

Discussion

Sodium citrate merely removes calcium ions from the blood (Goodman & Gilman, 1970) and thus its use as an anti-coagulant is the standard method in stable fly research followed by various authors (Du Toit, 1974; Du Toit, 1975; Hafez & Gamal-Eddin, 1959a; Parr, 1959; Parr, 1962; Wang & Gill, 1970; Bailey, Whitfield & La Brecque, 1975; La Brecque, Weidhaas & Whitfield, 1975). Jones (1966) found that blood containing 5,0 g of sodium citrate per litre was still acceptable to *S. calcitrans,* while Hafez & Gamal-Eddin (1959a) proved that this chemical had no harmful effect on the adult flies or their progeny.

When rearing stable flies on citrated buffalo calf blood at 3 temperatures varying between 25 °C and 34 °C Hafez & Gamal-Eddin (1959a) found that males lived longer than females (15,1 days at 25 °C and 3,2 days at 34 °C.). Bailey *et al.* (1975) and Killough & McKinstry (1965) found the total lifespan (adult MT_{100}) of *S. calcitrans* fed on citrated bovine blood to be 21 days and 4,5 weeks respectively at 24 °C. Du Toit (1975) found the lifespan (adult MT_{100}) to be 43 days on a bovine blood diet, but did not state the temperature at which the flies were held, nor did he distinguish between the sexes. The differences between the present and previously published results may be attributable to differences in temperature, as Hafez & Gamal-Eddin (1959a) showed that longevity (adult MT_{100}) increased with a decrease in temperature.

In this study the shortest pre-oviposition period was 5,3 days in flies on a diet of cattle blood and the longest was 9,3 days in flies on both donkey and goat blood. Du Toit (1975) gave a pre-oviposition period of 6,5 days for bovine and pig blood and 7,5 days for ovine blood. Hafez & Gamal-Eddin (1959a) had shown a reduction of the pre-oviposition period from 7,9 days at 22 °C to 5,5 days at 30 °C in flies fed on citrated buffalo calf blood. Various other workers also reported variations in the lengths of pre-oviposition periods in flies fed on different blood diets and kept at different temperatures (Glaser, 1923; Simmons, 1944; Ashrafi, 1964; Schoof, 1964; Killough & McKinstry, 1965; Suenaga, 1965; Bailey *et al.,* 1975; La Brecque *et al.,* 1975).

On diets of dog and pig blood a very high percentage of the females died before they could reproduce and this population limiting factor should be taken into account in considering the overall reproductive capacity of these females.

Females fed on cattle and pig blood produced eggs with higher viability rates than those on sheep blood. Du Toit (1975) found that females fed on bovine blood produced larger numbers of viable eggs than those fed on pig blood, and more on pig blood than they did on ovine blood.

Acknowledgements

The author is indebted to the Faculty of Veterinary Science, University of Pretoria, for supplying the dog blood, and also to Prof. H. E. Paterson, Drs I. G. Horak, C. J. Howell and K. R. Solomon and Mr E. M. Nevill for their constructive criticism of the manuscript. A sincere word of thanks is due to Dr K. R. Solomon for helping with the statistical analyses and to Miss J. B. Walker for editing the manuscript.

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Printed by and obtainable from The Government Printer, Private Bag X85, Pretoria, 0001