# Sex attraction in *Cruznema lambdiense* (Nematoda: Rhabditidae)

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#### SUMMARY

Cruznema lambdiense males did not attract males, and females did not attract females. Young virgin males responded to young virgin females but not to old virgin females. Young virgin females, however, responded to both young and old virgin males. Virgin males were not attracted to non-virgin females, but non-virgin males were attracted to virgin females. Non-virgin females showed a positive response to virgin males, and virgin females also responded to non-virgin males. Attraction of males to females and females to males increased when the number of attractant worms increased from ten to 50, but greater numbers did not produce a corresponding increase in attraction. When both males and females were put together in varying ratios at the attractant source, the attraction of females increased slightly to ratio of female: male of 1:50 to 20:50 but declined thereafter to increasing ratios. Males, however, did not show any similar increase to male: female ratios and attraction gradually decreased from 1:50 to 50:50 male: female ratio.

#### Résumé

L'attraction sexuelle chez Cruznema lambdiense (Nematoda: Rhabditidae)

Les mâles de *Cruznema lambdiense* ne sont pas attirés par les mâles, ni les femelles par les femelles. Les jeunes mâles vierges répondent à l'attraction des jeunes femelles vierges mais pas à l'attraction des vieilles femelles vierges. Par contre, les jeunes femelles vierges sont attirées par les mâles vierges, jeunes ou vieux. Les mâles vierges ne sont pas attirés par les femelles non-vierges mais les mâles non-vierges sont attirés par les femelles vierges. Les femelles non-vierges répondent positivement à l'attraction des mâles vierges et les femelles vierges répondent également à celle des mâles non-vierges. L'attraction des mâles par les femelles et des femelles par les mâles augmente quand le nombre des individus attractifs augmente de dix à 50 mais au-delà l'attraction n'augmente plus. Quand le groupe d'individus attractifs est composé d'une proportion variable de mâles et de femelles, l'attraction des femelles augmente quand la proportion femelle : mâle augmente de 1 : 50 à 20 : 50 et diminue ensuite quand cette proportion devient plus importante. Par contre, cette augmentation de l'attraction n'est pas observée chez les mâles, dont l'attraction diminue graduellement quand le rapport mâle : femelle varie de 1 : 50 jusqu'à 50 : 50.

The effects of age and reproductive state of Panagrellus redivivus on sex attraction has been studied by Duggal (1978). Adult male P. redivivus showed no significant attraction towards copulated or gravid females although virgin females were attracted to males throughout their life span. Chiloplacus symmetricus females, however, did not respond to males but the males showed a positive response towards females (Ahmad & Jairajpuri, 1980 a). In the following

work, the sex attraction behaviour of *Cruznema lambdiense* (Maupas, 1900) Thorne, 1961 is discussed.

#### Materials and methods

The nematodes were cultured xenically in peptone agar supplemented with wheat flour. Sex attraction was studied in modified "mickey mouse" chambers (Ahmad & Jairajpuri, 1980 b). Significance tests were made by comparing the number of worms in the test and control chambers

All experiments were replicated five times, except one involving attraction to varying number of worms which was replicated only three times. Always 100 worms were placed in the inoculation chamber.

#### Homosexual attraction

Males and females were separated during the fourth moulting stage and were reared in separate Petri dishes. 50 males to be tested for attractants were placed in a straw pipe containing agar in the centre of the test chamber for 6 hr, to allow gradients to form; 100 worms were then released in the inoculation chamber and their distribution was recorded after 3 hr. A similar experiment was done to study the attraction of females to females.

#### HETEROSEXUAL ATTRACTION

Sex attraction was also tested with the same method, between young and old virgin males and females, virgin males and non-virgin females and between non-virgin males and virgin females. The mean life span of virgin worms was 6.5 days and ten days for non-virgins (Ahmad & Jairajpuri, to be published). Virgin or non-virgin "young" worms were one to three day-old and "old" worms were six to seven day-old.

#### Effect of the number of worms

Attraction to virgin males or virgin females was tested by using 10, 50, 100 and 200 worms. The final distribution of the worms was recorded after 3 hr.

#### EFFECT OF MALE AND FEMALE RATIO

Attraction of virgin males and virgin females to varying ratios of 1:50, 10:50, 20:50, 30:50, 40:50 and 50:50 was studied in "mickey mouse" chambers. The two sexes were placed in the straw pipe in the test chamber, then allowed to mix.

#### Results

#### HOMOSEXUAL ATTRACTION

Males did not show any significant attraction (P > 0.1) towards virgin males (Fig. 1 A), and virgin females did not respond to virgin females (P > 0.1; Fig. 1 B).

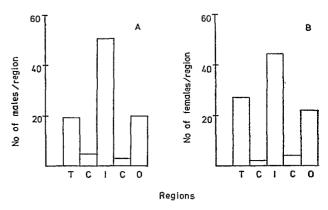


Fig. 1. A: Response of males to males; B: Response of females to females. (Abbrev. T = test chamber; C = connecting channel; I = inoculation chamber; O = control chamber).

# ATTRACTION BETWEEN VIRGIN MALES AND VIRGIN FEMALES

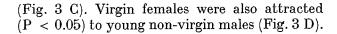
Young (one to two day-old) virgin males were attracted (P < 0.001) to young virgin females of the same age (Fig. 2 A) but did not respond (P > 0.05) to 6-7 day old virgin females (Fig. 2C). Young virgin females were also attracted (P < 0.001) to young virgin males (Fig. 2 B), and (P < 0.01) to old virgin males (Fig. 2 D).

# Attraction between virgin males and non-virgin females

Virgin males were not attracted (P > 0.1) to two to three day-old non-virgin females (Fig. 3 A), but non-virgin females of the same age were attracted (P < 0.01) towards virgin males (Fig. 3 B).

### ATTRACTION BETWEEN NON-VIRGIN MALES AND VIRGIN FEMALES

Non-virgin males showed significant attraction (P < 0.001) towards young virgin females



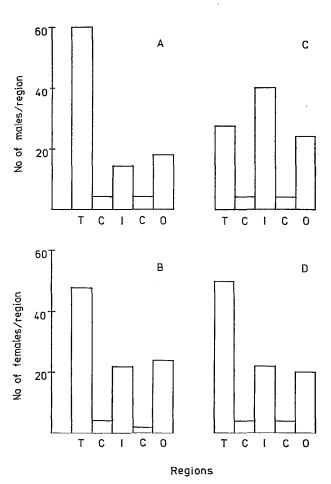


Fig. 2. A: Response of young virgin males to young virgin females; B: Response of young virgin females to young virgin males; C: Response of young virgin males to old virgin females; D: Response of young virgin females to old virgin males; (Abbrev. same as in Fig. 1).

### EFFECT OF THE NUMBER OF WORMS ON SEX ATTRACTION

Attraction of virgin males to varying numbers of virgin females and vice-vera, showed a similar pattern (Fig. 4). In both cases, attraction was relatively low with ten individuals, and increased significantly (P < 0.01) for numbers up to 50, then remained constant when the number of individuals increased up to 200.

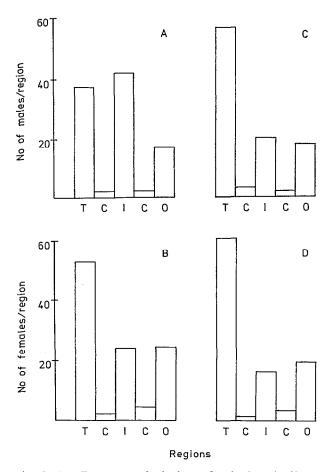


Fig. 3. A: Response of virgin males to two to three day-old non-virgin females; B: Response of two to three day-old non-virgin females to virgin males; C: Response of non-virgin males to young virgin females; D: Response of virgin females to 1-2 day-old non-virgin males; (Abbrev. same as in Fig. 1).

## EFFECT OF MALE AND FEMALE RATIO ON SEX ATTRACTION

Up to a female: male ratio of 20:50, the attractiveness of virgin females was not hindered and although no significant difference have been observed, a synergistic effect is probable for there was an increase in attractiveness when the ratio increased from 1:50 to 20:50. Female: male ratios of more than 20:50 resulted in a sudden decrease in attraction and the female response was not significant at ratios of 40:50 and 50:50 (P > 0.1).

In contrast, virgin males did not show any increase in attraction to any of the male:

female ratios (Fig. 5). There was a gradual decline in attraction to increasing ratios, and at 30:50 male: female ratio the males were not attracted (P > 0.1).

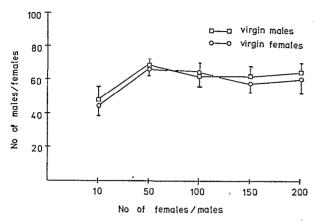


Fig. 4. The effect of the number of attractant worms on sex attraction.

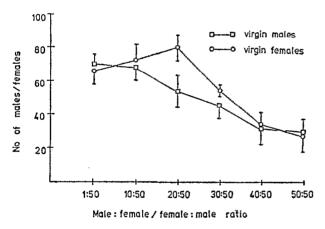


Fig. 5. The effect of male and female ratio on sex attraction.

#### Discussion

Like Panagrolaimus rigidus (Greet, 1964), Panagrellus silusiae (Cheng & Samoiloff, 1971) and P. redivivus (Duggal, 1978), both sexes of Cruznema lambdiense produced sex attractants which resulted in a mutual response towards each other. Virgin females lost their attractiveness as they grew old, and they apparently became unattractive after copulation. These observations are similar to those of Duggal

(1978) for P. redivivus except that he did not test females to virgin and non-virgin males separately. As he took males directly from the culture, presumably, they were non-virgin, and since virgin females were attracted to them throughout their life span, it may be concluded that non-virgin males produced attractants throughout their life span as in the present study with C. lambdiense. Duggal concluded that P. redividus females were attractive or attracted to males only when they had no sperm and large number of oocytes in their oviduct. As the female response is dependant on the presence of male attractants and as males produced attractants throughout their life span, it may be apparent that a "no response" by the females is brought about by a blockage in the chemosen sory receptors. For such a feedback system to operate, the reproductive system would be connected to the central nervous system. However, Yuen (1971) failed to observe nerve tissues in the reproductive system of Aphelenchoides blastophthorus. After the above experiments with C. lambdiense it is suggested that perhaps copulation initiated a change in the reproductive tract of the female that ultimately inhibited production of sex-attractants. This suggestion is based on the study of Cheng & Samoiloff (1972) who showed that inhibition of gonad development also inhibited sex attraction in P. silusiae, and hence the gonads were the source of the attractants. Even in C. symmetricus, young (four to five day-old) copulated females were attractive to males (Ahmad & Jairajpuri, 1980 a). As six to seven day-old virgin females also became unattractive, there is a possibility that degenerative changes in the gonad might also inhibit production of sex attractants.

In C. lambdiense like in P. silusiae (Balakanich & Samoiloff, 1974) males and females showed a maximum response at 50 worms, and did not increase thereafter. The cause of the slight, though not significant, indication of synergy in female attraction to 20:50 female: male ratio is uncertain as like sexes did not attract each other. But it may be that this result was either due to a mixing of the male and female pheromones, or to some other secretion that might have been procuced by one or both sexes upon physical contact.

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