EXPONENTIAL GROWTH IN CULTURE OF THE PLANARIAN MOSQUITO PREDATOR *DUGESIA DOROTOCEPHALA* (WOODWORTH)[1]

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ABSTRACT. Three living and 7 frozen food substances were used to culture the planarian, *Dugesia dorotocephala* (Woodworth), an effective biological control agent against *Culex* spp. Mosquito larvae were slightly superior to the brine shrimp, *Artemia salina* (L.), while red *Tubifex* sp. was the least satisfactory for asexual multiplication among living foods. All frozen foods except oysters were generally inferior to living foods. Among the viscera, pork and beef livers were superior to beef kidney and chicken liver. Planarians showed exponential growth by doubling their population ca. every 10 days on live foods and oysters.

The fresh water planarian, *Dugesia dorotocephala* (Woodworth), a useful biological control agent against mosquito larvae (Legner and Medved 1974, Legner et al. 1975, Medved and Legner 1974, Yu and Legner 1976), has not been mass produced at practical levels required for large scale field applications. This study focuses on the effectiveness of various food substances to stimulate asexual multiplication.

In the field planarians are reported to feed on an array of substances including oligochaetes, arthropods, and molluscs (Hyman 1951, Jennings 1957, Mitchell 1974, Reynoldson and Young 1963). In the laboratory, *Daphnia* spp. and the nauplii stage of brine shrimp, *Artemia salina* (L.) are commonly used (McConnell 1967). Nonliving foods are less desirable because they tend to contaminate the medium. Some foods such as animal muscle and egg yolk, fats, starches and sugar, are not very suitable for planarian growth (Wulzen 1923). Also, the component in animal tissue which promotes planarian growth is heat labile (Wulzen 1924, 1927).

This study compared the asexual multiplication rates of planarians fed on 3 living and 7 frozen food substances in plant-filtered culture systems.

MATERIALS AND METHODS. The culture of *D. dorotocephala* originally collected from Lytle Creek, San Bernardino Co., California had been maintained in the laboratory on *Culex* spp. larvae for a year prior to the experiment.

Three living and 7 frozen foods were used. Larvae of *Cx. quinquefasciatus* Say were cultured in a greenhouse with caged chickens provided for adult blood meals. Adult brine shrimp, *A. salina*, were produced in a 10 x 4 x 0.3 m earthen pond containing 12,000 L of ASP-2 culture media (Provasoli et al. 1957, van Baalen 1962). The shrimp fed on phytoplankton supplemented with trout food (Purina Trout Chow, Ralston Purina Co., St. Louis, MO.), which was soaked, homogenized, and then filtered through a 50-mesh dip net into the pond. Red *Tubifex* sp. were purchased locally and maintained in running tap water for use. Frozen foods included beef, pork, and chicken livers, beef kidney, shrimp, squid and oysters, which were also purchased locally. Only the muscles of shrimp and squid were used. All 7 foods were frozen before use.

The planarians were reared in plastic pans, 36 x 30 x 13 cm. containing 5 ca. 400-cc granite stones, 8 L tap water and 3 water hyacinth, *Eichornia crassipes* (Martius) Solms, or 30 g of filamentous green algae *Rhizoclonium hierglyphicum* (Agarah) Kützing when frozen food was used, a system patterned after one described by McConnell (1967). Air temperature was controlled at 26° ± 2° C. and light intensity

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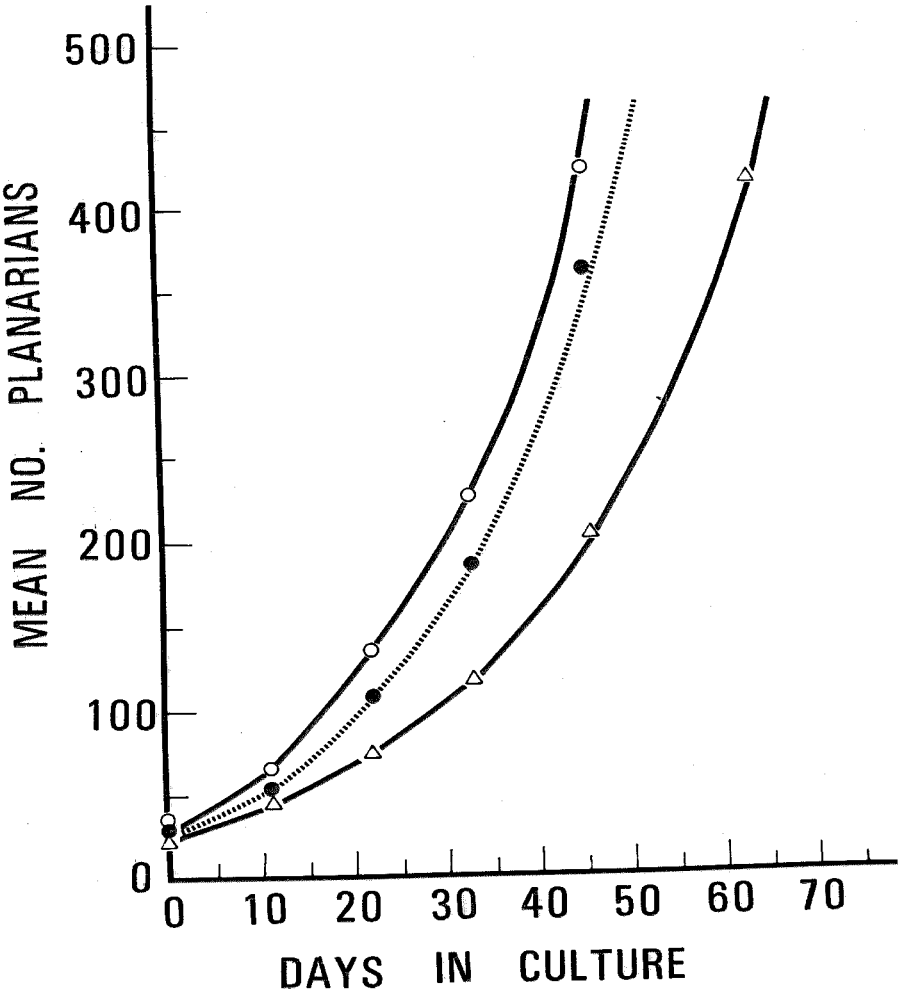

Fig. 1. Growth curves for *Dugesia dorotocephala* fed on 3 live baits: mosquito larvae (open circle), brine shrimp (solid circle), and red *Tubifex* sp. (triangle). The planarians were reared in 8 L water with 3 water hyacinth plants at 26°±2° C. without harvest.

was kept under 1,000 ft-c. on clear days by whitewashing the greenhouse in which the experiment was performed. Photoperiod was extended to 15 hr. with fluorescent lights giving ca. 200 ft-c. illumination on the water surface before dawn. Under these conditions, dissolved 0_2 in the culture pans fluctuated optimally between 25 ppm in the afternoon to 3 ppm at night (Legner et al. 1976). Planarian population growth rates were determined from an initial 30, 10–15 mm long individuals in each replicate; 4 replicates were made of each kind of food which was supplied every 48 hr. The total number of planaria was determined every 10 days. Three size groups

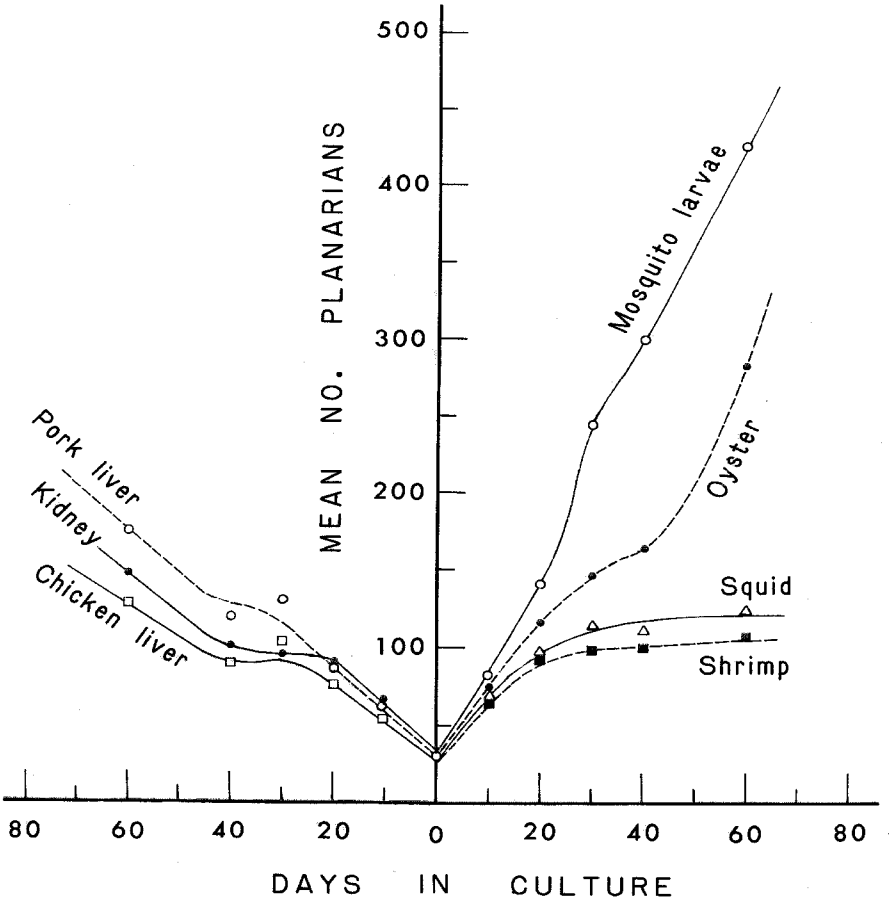

Fig. 2. Growth curves for *Dugesia dorotocephala* fed on 7 kinds of thawed frozen foods, with live mosquito larvae as a control. The planarians were reared in 8 L water with 30 g filamentous green algae under 26° ± 2° C. without harvest.

determined were (1) large, 15 mm +, (2) medium 10–15 mm, and (3) small, 10 mm −.

An excess of food was provided at each feeding. Three 3rd instar *Cx. quinquefasciatus* larvae were furnished for each planarian so that ca. 1 larva was left for each planarian 2 hr after feeding. In the case of brine shrimp and *Tubifex* sp., each planarian was given ca. 2 prey or the amount that could be consumed in 2 hr. Frozen food was first thawed and cut into pieces of ca. 1 cc. Five pieces of the same food were then affixed to a 25 cm bamboo stake that was placed horizontally on the bottom of the culture pan. The remaining food was removed from the water with the stake 5 hr later.

Planaria were counted ca. every 10 days followed by filtering the water through a dip net. Excess plants, due to growth, were removed, and the stones and the pan cleaned. Planarians were then returned with the water to the rearing unit for the next 10-day period.

RESULTS AND DISCUSSION. The 3 live baits fed for 2 months from November 13, 1975 to January 16, 1976, were all satisfactory for multiplication (Fig. 1). Brine shrimp were superior to red *Tubifex* sp., but mosquito larvae were the most effective. These planarians multiplied from 30 to ca. 400 in 46 days, after which their growth slowed.

Oysters were the best in the frozen food class for multiplication (Fig. 2). Animal viscera were satisfactory, while shrimp and squid muscle were poor. The multiplication curves leveled after planarians had fed on the muscles for 1 month (Fig. 2). This was in agreement with earlier findings that muscle was inferior to animal viscera as planarian food (Wulzen 1923). There was no difference between pork and beef livers (the latter not shown in Fig. 2), while chicken liver was definitely inferior.

All foods were adequate for body length gain. A typical size distribution during the course of an experiment was demonstrated on a diet of *Tubifex* sp. for 63 days (Table 1). Planaria progeny required 46 and 63 days to reach the medium and large size, respectively.

Brine shrimp appeared to be the most practical live bait for planarian mass production by causing exponential growth (Fig. 1). Brine shrimp are commercially available and can be easily maintained in sea water, although they are slightly less effective than mosquito larvae in promoting planarian multiplication. Also, being marine organisms, brine shrimp bear no diseases to fresh water planarians. By comparison, "holing" disease (McConnell 1967) was found once among planarians fed with red *Tubifex* sp. Brown *Tubifex* sp. that are more easily cultured, were found to be too large for the planarians to attack.

Oysters were most suitable in the frozen food class, producing an almost exponential growth in the planaria culture. Beef and pork liver did not induce comparable multiplication rates, but may be suitable alternatives to oysters where costs and availability are primary considerations.

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Table 1. Average number of *Dugesia dorotocephala* in 3 size groups during the course of a feeding experiment on *Tubifex* sp.

Size	Days in Culture					
	0	11	22	33	46	63
Large (15 mm+)	10	10.5	16.0	18.8	30.0	74.5
Medium (10–15 mm)	20	9.0	8.5	25.5	29.0	87.0
Smal (10 mm—)	0	28.0	50.8	64.5	129.5	253.5
Total	30	47.5	74.3	108.8	188.5	415.0

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