# ASTRONOTUS OCELLATUS (CICHLIDAE: PISCES) AND MACROPODUS OPERCULARIS (ANABATIDAE: PISCES) AS PREDATORS OF IMMATURE AEDES FLUVIATILIS (DIPTERA: CULICIDAE) AND BIOMPHALARIA GLABRATA (MOLLUSCA: PLANORBIDAE)

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Two fish species, Astronotus ocellatus (Cichlidae) and Macropodus opercularis (Anabatidae) were tested for predacious behaviour toward immature mosquitoes (Aedes fluviatilis, Diptera: Culicidae) and schistosomiasis snail hosts (Biomphalaria glabrata, Mollusca: Planorbidae), in the presence or absence of non-living food and in laboratory conditions. A. ocellatus, a species indigenous to Brazil, was a very efficient predator of both organisms ( $\alpha = 0.05$ ); M. opercularis, an exotic species, preyed well on immature mosquitoes, but small snails and snail egg-masses were ingested only irregularly. Both fish species seemed to prefer live to non-living food.

Key words: biological control - Brazil - fishes - Astronotus ocellatus - Macropodus opercularis - mosquitoes - snails

Residual insecticides continue to be the mainstay of many mosquito control operations. However as a result of increasing resistance among vector species, high cost of chemical products and operational procedures as well as concern for environmental quality, alternatives to such pesticides are being increasingly sought. Among these, the biological control of immature mosquitoes using fishes has received much attention in recent years (WHO, 1982, 1984; Alio et al., 1985) and has been placed by the World Health Organization (WHO) as "Priority II" on a list for the development of biological control agents (WHO, 1984).

On the other hand, though resistance to molluscicides usually is not a problem in the control of intermediate hosts of schistosomiasis, several instances exist where high cost and operational difficulties are factors limiting the success of such programmes. Fishes, as biological control agents of snail vectors, have been studied several times (Milward-de-Andrade & Antunes, 1969; Motta & Gouvea, 1971; WHO,

1982; Feitosa & Milward-de-Andrade, 1986). Usually, in laboratory experiments, as a first step to assess their predacious behaviour, fishes have been tested against one vector organism at a time, in constant food conditions. Because immature mosquitoes and snails can be found together in many breeding places, fishes that are able to control both would be specially useful. Also it seems important to assess the influence of alternative non-living food on predatory activity.

In the present work, the predactious behaviour of two fish species — Astronotus ocellatus (Cichlidae) and Macropodus opercularis (Anabatidae) against mosquito larvae or pupae Aedes fluviatilis: Culicidae) and egg-masses and snails (Biomphalaria glabrata: Planorbidae) has been tested. A. ocellatus is a species indigenous to Brazil, a native of Amazonas but now found throughout the country. Its biology and mass reproduction is well known (Fontenele, 1951; Braga, 1953). M. opercularis is of Asian origin and is mainly bred as an ornamental fish, but it seems to have been used, empirically, as a mosquito control agent around Belo Horizonte, Brazil. Costa (1990) reported its introduction in the "Billings" water reservoir in São Paulo, Brazil, for mosquito control.

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## MATERIALS AND METHODS

## **Fishes**

Astronotus ocellatus — Young specimens, 9 to 11 cm in length were purchased in fish shops and maintained in aquaria containing 35 l of dechlorinated tap water (up to 5 fishes/aquarium). Though described as omnivorous (Menezes, 1962) this species is usually fed at on meat, or chopped fish or shrimps in captivity (Braga, 1953; Feitosa & Milward-de-Andrade, 1986). Because of our somewhat artificial conditions, these foods were not readly consumed, thereby causing water pollution, so crushed B. glabrata and B. taenagophila snails were used in their place with good results.

Macropodus opercularis — Adult specimens of both sexes, also bought in fish shops, 5 to 7 cm in length were also maintained in 351 aquaria (up to 7 fishes/aquarium) and fed on commercial fish-food, composed of desiccated and ground shrimps.

# Mosquitoes

Aedes fluviatilis is a Neotropical and semidomestic species, frequent throughout Brazil. Fourth instar larvae and recently formed pupae were obtained from the colony existing for several years at "Centro de Pesquisas René Rachou" (CPRR), FIOCRUZ, Belo Horizonte, originally started with mosquitoes collected around Belo Horizonte (Consoli & Williams, 1981). It had previously been found that neither of the fish species employed feed on Ae. fluviatilis eggs.

# Snails

Large (15 to 27 mm/diameter), medium sized (8 to 10 mm/diameter) and small (0.2 to 0.5 mm/diameter) melanic *B. glabrata* snails and egg-masses were obtained from the colony that has been maintained at CPRR/FIOCRUZ for nearly 20 years.

## Experiments

All experiments took place in 351 aquaria containing dechlorinated tap water and lasted for 24 h. Natural illumination was kept throughout the tests, temperature ranging between 22 and 30 °C (air) and from 19 to 26 °C (water). Similar experiments were performed with both fish species. Table I shows the numbers of fishes, immature mosquitoes, snails and alternative non-living food present in each aquarium in all experiments.

Predation on immature mosquitoes in the presence or absence of snails and non-living food — Separate experiments were conducted offering mosquito larvae (Experiment 1) and pupae (Experiment 2) together or not with small snails and alternative non-living food, which consisted of smashed snails (2 g/aquarium) for A. ocellatus and of 0.2 g of commercial fish-food for M. opercularis. Previously, it had been calculated that an individual fish of each species would be able to consume the amounts indicated in 24 h. Each experiment was repeated 3 times for A. ocellatus and owing to more irregular predating pattern, 6 times for M. opercularis.

Predation on differently sized snails, in the presence or absence of non-living food — For each fish species, snails and alternative food were distributed as shown in Table I (Experiment 3) and repeated 3 times. Amounts of non-living food remained as in experiments 1 and 2.

Predation on egg-masses and eggs of B. glabrata — Recent egg-masses were carefully detached from pieces of transparent plastic sheets on which they had been laid, the eggs were counted and distributed in aquaria as shown in Table I (Experiment 4). Non-living food was offered in the amounts described before.

#### Statistical evaluations

Means were compared using the "Duncan's test" (Edwards, 1960) and a level of  $\alpha = 0.05$  (5%) was adopted.

### RESULTS AND DISCUSSION

Experiments 1 and 2 - Table II shows the means and standard deviations of the numbers of predated larvae, pupae and snails in exper. 1 and 2, for both fish species. All fishes survived the experiments. A. ocellatus ate all larvae both when these were offered as the sole food source (A) or together with crushed snails (B). When offered together with live molluscs, larvae were significantly predated and snails almost completely consumed (C). Nearly all B. glabrata were eaten when offered alone. The same fish species ate all pupae and snails in whatever combination they were offered. Non-living food was consumed whenever available, but never completely. Braga (1953) describes a carnivorous diet for this species and Motta

TABLE I

Numbers of fishes, mosquito larvae or pupae, snails (L = large; M = medium size; S = small), snails egg-masses in the presence (+) and absence (-) of non-living alternative fish-food

·	Experiment no.											
Aquaria	1/2				3				4			
	Larvae or pupae	Fish	Snails	Non- living food	Fish	L	Snails M	s	Non- living food	Fish	Egg- masses	Non- living food
A	300	1	_	_	1	10	_	_	_	1	10	+
В	300	1	-	+	1	10	_	_	+	1	10	+
C	300	1	30	_	1	_	10	_	_	1	10	+
D	_	1	30	_	1	_	10	_	+	1	10	_
${f E}$	_	_	30		1	_	_	10	_	1	10	_
F	300	_	_	_	1	_	_	10	+	1	10	_
G	_	_	<del></del>	-	_	10	10	10	_		10	-

TABLE II Means and standard deviations ( $\bar{x} \pm s$ ) of predated larvae, pupae and snails in experiments 1 and 2 and mortality in controls (E and F)

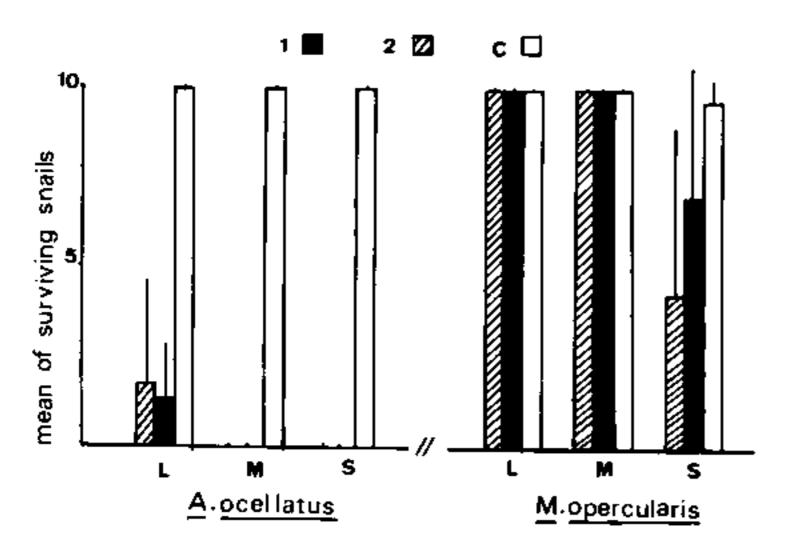
Aquaria		Astronotu	s ocellatus		Macropodus opercularis					
	Larvae	Snails	Pupae	Snails	Larvae	Snails	Pupae	Snails		
A	300.0± 0.0		300.0±0.0		290.0±24.4		221.0±93.9			
В	$300.0 \pm 0.0$		300.0±0.0		265.1±38.7		254.0±53.0			
Ċ	263.3±53.1	29.6±0.5	300.0±0.0	30.0±0.0	269.1±55.2	$3.3 \pm 3.6$	246,0±57.2	8.6±12.7		
D		26.3±5.1		30.0±0.0		14.3±12.9		18.1±13.6		
E				0.6±0.5		$0.3 \pm 0.8$		0.5± 0.8		
F	$3.3 \pm 3.0$		14.6±3.0		$3.8 \pm 3.1$		10.5± 5.6			

& Gouvêa (1971) refer to it as omnivorous and malacophagous; Feitosa & Milward-de-Andrade (1986) described it as malacophagous and mention the inclusion of mosquito larvae in its habitual laboratory diet. In our conditions immature mosquitoes and accompanying snails were clearly preferred to non-living food.

Macropodus opercularis ate most larvae in any combination offered, always in significant amounts (A, B and C). Snails were predated significantly only when they were the sole food available (D); pupae were always consumed in significant amounts (A, B and C). In these experiments M. opercularis preyed significantly on snails only in absence of other food and non-living food was never completely consumed. It seems therefore, that living prey is also preferred by this species, the maximal number

of larvae ingested in 24 h being at least more than twice the number referred to by Costa (1988). Mentioned by Jenkins (1964) as a potential predator of mosquito larvae, this species is usually bred as an ornamental fish in Brazil, and is referred by some fish sellers in Belo Horizonte as having been empirically introduced in ponds, wells and other water reservoirs in this area with the aim of controlling mosquito larvae. In São Paulo, the easy environmental adaptability also in polluted water, its larvivorous behaviour and its introduction in the "Billings" reservoir (Costa et al., 1985, 1987; Costa, 1988, 1990) have been described. Though all available data seem to support the mosquito controlling ability of this exotic species, additional studies as those recomended by WHO (1981a, b, 1984), are highly and urgently desirable.

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Means and standard deviations referent to numbers of large (L), median (M) and small (S) Biomphalaria glabrata surviving predation of Astronotus ocellatus and Macropodus opercularis in presence (1) and absence (2) of alternative food and in control (C).

Experiment 3 — Figure shows the means and standard deviations of the numbers of large, medium sized and small B. glabrata surviving predation of A. ocellatus and M. opercularis. The former species preyed on all small and medium sized molluscs, and significantly on large ones, whether or not in the presence of crushed snails. Almost all surviving large snails were attacked and presented many torn of shell parts, being unable to survive. All crushed snails were also consumed. Feitosa & Milward-de-Andrade (1986) report the preference of A. ocellatus toward smaller molluscs; Gouvea & Motta (1971) described the preference for molluscs and the resemblance of its natural habitat to that of B. glabrata; the same authors (Motta & Gouvea, 1971) described a measurable diminution in a B. glabrata population after the introduction of this species in the "Amaralina" lake in Salvador, Bahia.

Macropodus opercularis did not prey on any large or medium sized mollusc, though sometimes seemed to attack them. Predation of small snails was irregular and not significant. The malacophagous competence of this species seems not to be very dependable, though Gouvea & Motta (1971) report the ingestion of up to 25 B. glabrata (1 cm/diameter) per day.

Experiment 4 – Astronotus ocellatus preyed on all egg-masses when no other food was present, and a significant number of them in the presence of alternative food. In this case a reduction of 68.6% in the number of eggs

occurred (Table III). Ingestion of crushed snails was complete in some instances. It was observed that sometimes swallowed egg-masses were completely or partially expelled resulting in empty or half empty gelatinous masses and many scattered eggs. Isolated eggs were recovered from the bottom of the aquaria, but torn empty gelatinous masses were not considered as egg-masses. Feitosa & Milward-de-Andrade (1986) did not observe the predation by this species when egg-masses were attached to the glass walls of aquaria. None of the tested fish species are surface scratchers but since in nature egg-masses may be attatched to soft materials like leaves or even on loose leave fragments (Souza & Lima, 1990; Souza, personal communication) their predacious behaviour on B. glabrata eggs in natural conditions remains to assess.

Macropodus opercularis did not ingest egg-masses significantly, either in presence or absence of alternative food, but it always attacked some of them and ingested some eggs. There was a reduction of 38.7% of eggs in the aquaria without other food and 41.1% in those containing it.

The Scientific Working Group on Biological Control of Vectors (WHO, 1981) established the following criteria for the evaluation of fish species as possible biological control agents for mosquitoes: readiness in attacking and feeding on immature mosquitoes, adaptability to local environmental conditions, potential for mass production, assessment of the ecological impact of the introduced fish and its tolerance to insecticides.

In the experimental conditions described, A. ocellatus seems to be a very effective predator of immature mosquitoes, having also a good potential for mass production (Braga, 1953). Being an indigenous species of wide geographical distribution, its ecological impact and adaptability should not represent difficulties in many instances. Large environments should be specially considered, because adults of this species can reach 1500 g in weight, being also suitable as human food (Motta & Gouveia, 1971). Its tolerance to insecticides remains to be assessed.

Predation on immature mosquitoes by *M*. opercularis also seems to be effective, its hardiness and adaptability having been described

TABLE III
Egg-masses $(\bar{x} \pm s)$ and total eggs of Biomphalaria glabrata before and after predation of Astronotus ocellatus
and Macropodus opercularis, in the presence or absence of alternative food

		$A.\ oce$	ellatus	M. opercularis					
	0 h		24 h		0 h		24 h		
	egg-masses $(\bar{x} \pm s)$	eggs total	egg-masses ( $\overline{x} \pm s$ )	eggs total	egg-masses $(\overline{x} \pm s)$	eggs total	egg-masses $(\overline{x} \pm s)$	eggs total	
No alternative food	10.0 ± 0.0	623	<u> </u>	_	10.0 ± 0.0	1182	7.0 ± 4.4	725	
With alternative food	10.0 ± 0.0	440	2.7 ± 2.3	138	10.0 ± 0.0	1320	7.3 ± 2.5	778	
Control	$10.0 \pm 0.0$	206	$10.0\pm0.0$	206	$10.0 \pm 0.0$	478	10.0 ± 0.0	454	

(Abreu, 1968; Costa et al., 1985, 1987). Having been commercially produced for a long time, its mass production should be easily feasible. Being an exotic species in Brazil, its ecological impact in several kinds of habitats and on numerous organisms should be carefully evaluated, as well as its tolerance to insecticides.

In respect to its malacophagous habits, our results on A. ocellatus mainly agree with earlier works. M. opercularis, on the other hand, seems not to be a very promising snail predator.

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