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# EVALUATION OF LARVIVOROUS PROPERTY OF *PUNITUS AMBASSIS, PUNITUS SARANA, CLARIAS BATRICHUS* AND *WALLAGO ATTU* NATIVE FISH SPECIES OF GWALIOR (M.P): A LABORATORY INVESTIGATION

Mohammad Ishfaq Wani\*, Sarita Shrivastava

Department of Zoology, Government Science Model College Gwalior, Madhya Pradesh, India \*Corresponding author: ishfaqzoology12@gmail.com, dBrs1956@gmail.com

### ABSTRACT

All tropical countries across the globe have to suffer mosquito-borne diseases every year like malaria, dengue, and filariasis. So, to reduce the carriers of these horrible diseases at the main root level, many chemical methods have been practiced which is quite impressive step, but it simultaneously causes ecological imbalance and hence pollutes and disturbs the environmental cycles. Biological control of mosquito vectors at the source level seems to be the best cheap price, eco-friendly, socially acceptable, highly impressive, stunning, and effective method than that of other methods employed. The present study is focused on using Punitus ambassis, Punitus sarana, Clarias batrichus, and wallago attu, native fish species of Gwalior (M.P), as a predator of mosquito larvae at the source level. All the predation experiment tests were conducted against mosquito eggs, larvae, and alternative food (wingless houseflies) at dissent pray densities in separated glass aquariums containing analyzed pond waters of Jal-Vihar and Fish farm of Gwalior. The time duration of each predation experiment is one week and fishes are fed with an equal number of mosquito eggs, larvae, and wingless houseflies at different densities at different time intervals on each experimental day. The study divulges that Punitus ambassis, Punitus sarana, Clarias batrichus, and Wallago attu are engaged with Larvicidal property with differences in their feeding consumption efficacies. Larvae feeding consumption was positively related to the body size of fishes. Thus, small the size of larvivorous fish, the more will be its power of larvicidal activity. According to the results of our present study, it is prophecy, that the endemic fish species of Gwalior Madhya Pradesh namely Punitus ambassis and Punitus sarana are likely to be impressive and effective in consuming mosquito eggs and larvae in breeding places.

Keywords: Larvae, Laboratory, Native fish, Temperate, Punitus ambassis.

## 1. INTRODUCTION

Mosquitoes are small flying nuisance insects, belongs to the family Culicidae. As they feed on the blood of living vertebrates, they are considered harmful insects. While feeding on blood, they may become carriers for many animal diseases, transmitting harmful zoonotic and other important diseases [1, 2]. For oogenesis female mosquitoes feed on host blood and find their way towards transitory water bodies. Since mosquitoes are regarded as insecure and are involved in direct and indirect disturbances of humans and other animals, thus their management, as well as control, has always been focused in the community scientific research studies.

For the reduction of these nuisance insects like mosquitoes, many methods have been operated so for include manufactured chemicals, genetic managerial approaches; however, they all bear some drawbacks related to environmental issues like aquatic pollution, bio-magnification, etc. Across the globe, Scientists have negated significant and serious ecological results upon the entire elimination of mosquitoes, creating a suitable room for developing the security and an excellent and ecological friendlier control method [3, 4]. Among them, the bio-control method of mosquito larvae control is the safest and is wilful use of natural foes to lower the population of reproducing pest organisms, which have attained undertaking for controlling bother insects like mosquitoes at the source level [5-8]. Bio-larvicides are the safest, excellent, attractive, and socially acceptable methods in mosquito control, and this has the deliberate use of biological agents that eat or demolish the mosquito larvae and other pests.

Diseases arising because of mosquitoes have been a prime issue of problems in all most all temperate countries like tropical and subtropical countries across the globe. Presently there is no successful vaccine against most such diseases to cure patients within a shorter period. These life-threatening diseases not only become a burden for our day to day life but are also responsible for killing 3 million people across the world per year [7, 9-11]. So, to get rid of these dreadful diseases, the minimization in the populace size of mosquitoes at the source level is very important. Various types of synthetic chemicals called insecticides are extensively used for combating the adult and larval mosquito populations in the aquatic habitat, which is hazardous not only for human health but for other fauna and flora too in the aquatic water body. Hence it is proved as an important cause of environmental pollution and deterioration. Chemical control approaches were practiced to eradicate the pests but due to the persistent use of these harmful chemicals, mosquitoes become less effective because of the development of resistance property in their genome [12-15].

Transitory aquatic ecosystems (pools, puddles, ponds, etc.) become suitable and flawless reproducing places for mosquitoes to breed freely. Thus, mosquitoes are emerging from such water bodies hence we can say these ephemeral water bodies become the actual sources of adult mosquitoes. Some species of fishes consume mosquito larvae as food because of their carnivorous property. Thus, the fish species that feed on immature mosquito larvae called wigglers are referred to as larvivorous fish. Biological control of mosquitoes refers to the pioneering or manipulation of fauna to put down the population of carriers or vectors. A broad range of organisms helps us to synchronize the mosquito populations naturally through the process of many ecological interactions like predation, competition, and parasitism. From all the biological control agents, larvivorous fishes are used most ordinary and extensively in vector control management. In various regions of India, a foreign larvicidal fish namely Gambusia affinis is used as a predator of immature wigglers or mosquito larvae, which is currently running in diverse rivers, ponds, and other water bodies. This foreign or exotic mosquito fish species is quite potent and successful in the lowering of mosquito larvae populations at breeding places, but it bears also certain environmental issues related to ecological imbalance and its deterioration, possible impacts on the native or original fish species, and other effects on biodiversity of the water body in which it is introduced. Thus, this research will stress the use of native larvivorous fish species of India with special reference to the Gwalior region of Madhya Pradesh to analyze their larvivorous property, so that we can use them for malaria bio-control management programs.

Thus, the research envisages evaluating and contributes an inventory biological control tool. It is accepted as not only excellent, little expenditure, ecologically sound, non-polluting, socially acceptable method but is the highly successful and potent method in vector management as compared to other methods applied.

## 2. MATERIAL AND METHODS

In this study, efforts were made to assess the larvivorous property of four native fish species of the Gwalior region of Madhya Pradesh under laboratory conditions.

# 2.1. Collection of mosquito eggs

As we know, female mosquitoes lay eggs on the surface of the water body in the form of egg rafts. The eggs were collected either in the form of egg rafts or individual form with the help of mosquito larvae collecting nets, spoons and O type brush from local in an around the Gwalior city ( $26^{\circ}22$ ' N 78^{\circ}18' E) like from shady ponds, flower pots and drains and ditches adjacent to the Jiwaji University Gwalior. The eggs were brought to the laboratory in small buckets or polythene bags and were shifted to  $18 \text{cm} \times 13 \text{cm} \times 4 \text{cm}$  size enamel trays containing 500 ml analyzed pond water.

# 2.2. Collection and maintenance of mosquito larvae

After laying the egg rafts by a female mosquito on the surface of the water, eggs are hatched into larvae after a few days depend upon the temperature of the water body in which they are laid. The mosquito larvae are collected by netting and dipping methods from the local shady ponds, water stored tiers, containers, ditches, and flower pots containing water and other stagnant water bodies adjacent to the Gwalior city (26°22' N 78°18' E). Mosquito larvae were then transferred in 5 L glass containers or buckets and were then brought to the laboratory. In the laboratory water containing larvae and other constituents were filtered to remove the phytoplanktons and zooplanktons and other dried leaves. After the sieving process, mosquito larvae were transferred to 7 L glass containers and were reared under laboratory conditions by giving them supplementary fish feed (namely Tubifex Worms). The entire collection of mosquito larvae and eggs was done by dipping, netting, and pipetting methods in October, November, and September.

# 2.3. Collection of Houseflies

Housefly swarms (*Musca domestica*) are mainly found near the municipal garbage sites, household dustbins, etc. As

the houseflies are attracted to garbage, they are collected from municipal garbage sites of Gwalior ( $26^{\circ}22'$  N  $78^{\circ}18'$  E) by using a plastic jar, inside sprayed with sugar solution and filled with the small pieces of mixed dates, sugar, honey, and apple squashes. The jar with the sweet sugar solution was placed near the garbage site for trapping maximum flies. The lid of the jar was kept open which allowed the flies to enter inside the jar. Lid of the jar was covered to trap the flies. In the laboratory, the wings of flies were removed with the help of scissor to make them wingless, so that they couldn't fly inside the aquarium when used as food for mosquitoes. The size of the houseflies was measured by the geometry scale which is recorded as 1.5 cm.

# 2.4. Collection, maintenance, and identification of fishes

Twelve fishes of four native fish species namely *Punitus ambassis*, *Punitus sarana*, *Clarias batrichus*, and *Wallago attu* were brought alive in fish shipping boxes from the Tigra Dam which is located about 23kms away from the Gwalior city (M.P), India. In the laboratory, before experiments, fish species were segregated and were maintained separately species-wise and were acclimatized for seven days under laboratory conditions and were kept in the glass containers containing aerated tap water. Fishes were fed with small pieces of earthworm (*Eisenia fetida*) and commercial fish feed (Tubifex Worms). Identification of fishes was done with the help of standard taxonomic keys of Rringuelet et al, and Ghedotti [16, 17].

# 2.5. Predation experiments

As this work is laboratory-based, the purpose of the experimental design is to stimulate the natural conditions inside all the fish glass aquariums (Aqueon having dimensions 25cm×20cm×20cm) by filling half of the glass aquariums with analyzed pond waters of Jal-Vihar and Fish farm of Gwalior, having the capacity of holding 10 litres of water. Fish lengths and weights of acclimatized fish species were measured by using a geometry scale and electronic weighing machine apparatus. The recorded lengths of the four native fishes are Punitus embassies 8.7 cm, Punitus sarana 14.6 cm, Clarias batrichus 20.5 cm, and Wallago attu 26 cm, and the fishes after measurement of lengths and weights were allowed to release in experimental aquariums in the morning hours. All the predation experiments were carried out in 10 L glass fish aquariums (Aqueon  $25 \text{cm} \times$ 20cm×20cm) half-filled with analyzed pond waters. Each

experiment was carried out separately in triplicates. On each predation experimental day, an equal number of 150 mosquito larvae, 200 mosquito eggs, and 70 wingless insects were given to each fish species on daily basis. The time duration of each experiment was one week and all the four fish species were fed with an equal total number of mosquito eggs 1450, 1050 larvae, and 490 wingless houseflies at different intervals of time in the entire experiment. Each food item was given separately to the fishes but not in a combined form so that counting of unconsumed food was easy. The predation starting time of each experimental day begins from morning 9 am to evening 6 pm. After the end of each experimental day, the number of unconsumed foods left by the fishes was removed and counted daily. Fishes were then transferred, from experimental glass aquariums to normal water tanks and were fed with small pieces of earthworm (Eisenia fetida) and commercial fish feed (Tubifex Worms) during the night. The data obtained during the days of each experiment was analyzed statically.

# 2.6. Statistical analysis

The data obtained during the entire experimental days were analyzed by using MS excel 2007 (Average  $\pm$  Standard Error).

# 3. RESULTS AND DISCUSSION

Mitigation or reduction of nuisance mosquitoes have been tried by diverse types of methods, however, due to certain limitations, related to ecological imbalance and environmental problems; there is a shift in approach to control and manage mosquito populations scientifically. Due to the ban or restrictions by environmental protection agencies (EPAs), there are now fewer synthetic chemicals available than there have been for the last 20 years [18-20]. Thus, bio-control of nuisance insects like mosquitoes have become more practical [21, 22]. In this contemplate, larvivorous fishes have been used worldwide for combating mosquito larvae [1, 2]. Researchers across the globe have evaluated indigenous fish species to find out appropriate local biological control agents [23, 25]. Various researches have recommended the introduction of mosquitofish species, Gambusia affinis, and Poecilia reticulata, and indigenous species to be successful to demolish nuisance mosquito population multiplying places [24, 25]. Concern has also been felt about the manipulation of foreign mosquito fish for mosquito larvae control due to their possible serious consequences on endemic fish species. The introduction

of mosquito fish *Gambusia affinis* in Greece (Europe) led to a decline of the native fish species Valencia letourneuxi [26, 27] and the related findings were reported in the United States, Spain & Australia [28-30]. Keeping the mentioned concerns in mind, scrutinized, endemic or native fishes were tested for their larvivorous potential and predatory potential properties so that we can use them in mosquito bio-control programs to reduce the malarial agents in ruler areas of a country.

Investigation results pursued that the endemic or native fish species namely *Punitus ambassis, Punitussarana, Clarias batrichus*, and *Wallago attu* possess excellent larvivorous potential property, to be used as biological control agents for mosquito larvae and other pests. Thus, in the mosquito bio-control programs, vectors can be controlled by the introduction of native fish species. Mosquito bio-control by using endemic larvivorous fish species proves less harmful than that of foreign larvivorous fish species [24, 31]. In the first experiment, our data ( average  $\pm$  SE) on larval feeding consumption efficacy sequence(LFCES) is PA 79.047 $\pm$ 0.259 > PS  $58.95 \pm 0.258 > CB \ 46.47 \pm 0.404 > WA \ 35.14 \pm 0.305$ (table 1 & 2) revealed that the above mentioned four native four species used in this examination, demonstrated larvicidal potential property so can be used as larvicidal agents, like earlier studies [32]. Thus, according to the LFCES, Punitus ambassis and Punitus sarana are more stunning and magnificent in combating mosquito larvae than the other two species namely Clarias batrichus and Wallagoattu. Punitus species are more successful and powerful in controlling mosquito larvae because of having a small-sized body. Smaller the body size of the larvivorous fish, the more it will possess larvivorous property because small body-sized fishes prefer small prey as food for their survival than that of large-sized food items.

Table 1: Larval feeding consumption efficacy of native fish species at different intervals of time of per day

Species name & body size	Day	No. of larvae supplied at 9 am	No. of larvae consumed from 9 am to 1 pm	No. of unconsumed larvae at 1 pm	No. of larvae supplied at 2 pm	No. of larvae consumed at 6 pm	Total no. of unconsumed larvae at 6 pm	Total no. of larvae supplied in a day	Total no. of larvae consumed in a day (Average ± SE)
	1	100	83	17	50	45	5	150	$128\pm0.272$
	2	100	75	25	50	38	12	150	$113 \pm 0.272$
Pontius	3	100	88	12	50	42	8	150	$130\pm0.471$
ambassis	4	100	72	28	50	40	10	150	$112 \pm 0.272$
(8.7cm)	5	100	78	22	50	36	14	150	$114\pm0.272$
(8.9g)	6	100	90	10	50	34	16	150	$124\pm0.471$
	7	100	66	34	50	43	7	150	$109 \pm 0.471$
	1	100	69	31	50	30	20	150	99±0.272
D :	2	100	60	40	50	32	18	150	92±0.272
Punitus	3	100	54	46	50	31	19	150	85±0.272
sarana (14.6cm)	4	100	59	41	50	35	15	150	94±0.272
(14.0 <i>cm</i> ) (10.12g)	5	100	55	45	50	27	23	150	82±0. 272
(10.12g)	6	100	58	42	50	22	28	150	80±0.272
	7	100	62	38	50	25	25	150	87±0.548
	1	100	59	41	50	28	22	150	87±0.272
<i>C</i> 1 ·	2	100	50	50	50	31	19	150	81±0.27
Clarias batrichus	3	100	44	56	50	25	25	150	69±0.272
(20.5cm)	4	100	39	61	50	23	27	150	$62 \pm 0.272$
(185g)	5	100	44	56	50	22	28	150	66±0.272
(1059)	6	100	38	62	50	24	26	150	62±0.272
	7	100	35	65	50	26	24	150	$61 \pm 0.272$
	1	100	28	72	50	18	32	150	46±0.272
.11	2	100	35	65	50	22	28	150	$57 \pm 0.471$
wallago	3	100	30	70	50	20	30	150	50±0.471
attu (26 cm)	4	100	33	67	50	21	29	150	54±0.471
(26cm) (456g)	5	100	40	60	50	26	24	150	66±0.272
(+509)	6	100	25	75	50	22	28	150	$47\pm0.272$
	7	100	28	72	50	21	29	150	49±0.272

In the second experiment, our data (average  $\pm$  SE) on egg feeding consumption efficacy sequence (EFCES) based on our observation is PA 66.35 $\pm$ 0.116 > PS 50.64 $\pm$ 2.493 > CB 34.71 $\pm$ 0.107 > WA 25.21 $\pm$ 0.091 given in the (table 3 & 4) shows that small-sized native fish species namely *Punitus ambassis* and *Punitus sarana* 

consumes more eggs than that of other two big sized native fish species namely *Clarias batrichus* and *Wallago attu*. Thus, it shows again small-sized fish species prefer small food like eggs as food while *Clarias batrichus* and *Wallago attu* shows less interest towards small food items like mosquito eggs.

Table 2: percentage (Average	SE) of larvae consumed by	y each fish species in a week

	Total no. of	Total no. of	Total no.	% age of larvae
Species name	larvae supplied	unconsumed	consumed larvae	consumed in a week
-	in a week	larvae in a week	in a week	(Average±SE)
Punitus ambassis (8.7cm) (9.9g)	1050	220	830	79.047±0.259
Punitus sarana (14.6cm) (12.90g)	1050	431	619	$58.95 \pm 0.258$
Clarias batrichus (20.5cm) (245g)	1050	562	488	46.47±0.404
wallago attu (26cm) (750g)	1050	681	369	35.14±0.305

PA= Punitus ambassis PS= Punitus sarana CB= Clarias batrichus WA= Wallago attu

### Table 3: Egg feeding consumption efficacy of native fish species at different intervals of time per day

Species name		No. of	No. of eggs	No. of eggs	No. of	No. of	Total no. of	Total no.	Total no. of
	Day	eggs	consumed	unconsumed	eggs	eggs	unconsume	of eggs	eggs consumed
	Day	supplied	from 9 am	at 1 pm	supplied	consumed	d eggs at 6	supplied	in a day
		at 9 am	to 1 pm	at i pin	at 2 pm	at 6 pm	pm	in a day	(Average $\pm$ SE)
	1	120	90	30	80	55	25	200	$145 \pm 0.471$
Dermiters	2	120	75	45	80	48	32	200	$123 \pm 0.471$
Punitus ambassis	3	120	88	32	80	52	28	200	$140 \pm 0.272$
(8.7cm)	4	120	93	27	80	44	36	200	137±0.272
(8.9g)	5	120	78	42	80	37	43	200	115±0.272
(0. <i>9g)</i>	6	120	90	30	80	47	33	200	137±0.272
	7	120	70	50	80	62	18	200	$132 \pm 0.272$
	1	120	69	51	80	50	30	200	119±0.272
D	2	120	60	51	80	42	38	200	$102 \pm 0.272$
Punitus	3	120	54	66	80	47	33	200	$101 \pm 0.272$
sarana (14.6cm)	4	120	59	61	80	38	42	200	97±0.272
(14.0cm) (10.12g)	5	120	55	65	80	37	43	200	92±0.272
(10.12g)	6	120	58	62	80	32	48	200	90±0.272
	7	120	62	58	80	46	34	200	$108 \pm 0.272$
	1	120	44	76	80	30	50	200	74±0.272
<i>C</i> 1	2	120	50	66	80	26	54	200	76±0.272
Clarias	3	120	59	61	80	24	56	200	83±0.272
batrichus (20.5cm)	4	120	38	82	80	25	55	200	63±0.272
	5	120	44	76	80	20	60	200	64±0.272
(185g	6	120	39	81	80	25	55	200	64±0.272
	7	120	35	85	80	27	53	200	62±0.272
	1	120	28	92	80	23	57	200	51±0.272
117 11	2	120	25	95	80	20	60	200	45±0.272
Wallago	3	120	40	80	80	18	62	200	58±0.272
attu	4	120	33	87	80	25	55	200	58±0.272
(26cm) (456 a	5	120	30	90	80	18	62	200	48±0.272
(456g	6	120	35	85	80	14	66	200	49±0.272
	7	120	28	92	80	16	64	200	44±0.272

In the third experiment, our data (average  $\pm$  SE) on housefly's feeding consumption efficacy sequence (HFCES) on the basis of observation is WA 95.51 0.372 > CB 77.75 $\pm$ 0.061 > PS 36.53 $\pm$  0.402 > PA 20.20 $\pm$ 0.167 given in table (Table 5 & 6). This sequence is quite different from that of the first two feeding sequences, here in this sequence Wallago attu consumes more wingless houseflies followed by Clarias batrichus, while as the other two small fish species namely Punitus ambassis and Punitus sarana shows less interest towards wingless insects because the size of the food item is big. It again shows that larger fishes like Wallago attu and *Clarias batrichus* prefer large food items for their survival and small fish species prefer small prey as food like larvae and eggs of mosquito for their survival as shown in (Fig.1). Thus, the sequence of the larvivorous feeding consumption efficacy consumption sequence in this study is Punitus ambassis > Punitus sarana > Clarias batrichus> Wallagu attu. The predatory feeding consumption efficacy sequence (PFCES) of native fish species based on wingless houseflies' consumption is Wallagu attu> Clarias batrichus> Punitus sarana> Punitus ambassis. This (PFCES) shows the greater predatory nature of Wallagu attu and Clarias batrichus while Punitus

sarana and Punitus ambassis show less predatory nature as they consume a fever number of houseflies. Out of the four native fish species, the most destructive for mosquito larvae populace is *Punitus ambassis* followed by Punitus sarana and the least destructive is Wallagu attu followed by Clarias batrichus as shown in (fig.1 & 2). Rupp et al and Chandra et al, suggested that, only native or endemic larvivorous fish species should be used as biological control of mosquito larvae to avoid the invasive nature of foreign or exotic species such as Gambusia and Poecilia. The indigenous native fish species namely Punitus ambassis and Punitus sarana showed excellent and stunning results with high larvivorous feeding consumption efficiency and good survival ability in small volumes of water containers. Besides this, they don't cause any harm to other native or endemic fish species and also breed naturally [31, 32]. Biological control of mosquito larvae by fishes especially with the introduction of native fish species is considered socially, scientifically accepted, eco-friendly, low-cost, and safe to humans and other non-target populations in the aquatic habitats [33-36]. Hence native larvivorous fishes must be used for mosquito bio-control programs as they are less harmful to other endemic species [37-40].

Table 4: percentage	(Average ± SE) of eggs consu	med by each fish species in a week

	Total no. of eggs	Total no. of	Total no.	% age of eggs						
Species name	supplied in a	unconsumed eggs	consumed eggs in	consumed in a week						
-	week	in a week	a week	(Average $\pm$ SE)						
Punitus ambassis (8.7cm) (98.9g)	1400	471	929	66.35±0.116						
Punitus sarana (14.6cm) (10.90g)	1400	691	709	50.64±2.493						
Clarias batrichus (20.5cm) (200g)	1400	914	486	34.71±0.107						
wallago attu (26cm) (556g)	1400	1047	353	25.21±0.091						

Table 5: Wingless houseflies feeding consumption efficacy of native fish species at different intervals of time of per day

Species name & body size	Day	No. of wingless houseflies supplied at 9 am	No. of wingless houseflies consumed from 9 am to 1 pm	No. of unconsumed wingless houseflies at 1 pm	No. of wingless houseflies supplied at 2 pm	No. of wingless houseflies consumed from 2 pm at 6 pm	No. of unconsumed wingless houseflies at 6 pm	Total no. of wingless houseflies supplied in a day	Total no. of wingless houseflies consumed in a day (Average ± SE)
	1	40	10	30	30	12	18	70	$32\pm0.272$
	2	40	5	35	30	7	23	70	$12\pm0.272$
Punitus	3	40	6	34	30	4	26	70	10±0.272
ambassis	4	40	4	36	30	3	27	70	$7\pm0.272$
(8.7cm)	5	40	5	35	30	6	24	70	11±0.272
(8.9g)	6	40	8	32	30	7	23	70	15±0.272
	7	40	4	6	30	8	22	70	$12\pm0.272$
Punitus	1	40	12	28	30	15	15	70	27±0.272
sarana	2	40	12	28	30	14	16	70	26±0.272
(14.6cm)	3	40	14	26	30	12	18	70	26±0.272

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(10.12g)	4	40	10	30	30	10	20	70	$20\pm0.272$
· · · · ·	5	40	16	24	30	15	15	70	31±0.272
-	6	40	12	28	30	16	14	70	28±0.272
-	7	40	11	29	30	10	20	70	21±0.272
	1	40	32	8	30	25	5	70	57±0.272
Clasica	2	40	33	7	30	26	4	70	59±0.272
Clarias - batrichus -	3	40	35	5	30	24	6	70	59±0.272
(20.5 cm) -	4	40	28	12	30	26	4	70	54±0.272
(20.3 cm) - (185g) -	5	40	26	14	30	23	7	70	49±0.272
(1859) -	6	40	35	5	30	28	2	70	49±0.272
-	7	40	32	8	30	22	8	70	54±0.272
	1	40	39	1	30	28	2	70	67±0.272
-	2	40	36	4	30	26	4	70	62±0.272
wallago -	3	40	37	3	30	28	2	70	65±0.272
attu (26cm) (456 a)	4	40	48	2	30	29	1	70	$77\pm0.272$
	5	40	38	2	30	27	3	70	65±0.272
(456g) -	6	40	40	0	30	27	3	70	67±0.272
-	7	40	39	1	30	26	4	70	65±0.272

### Table 6: Percentage (Average $\pm$ SE) of wingless houseflies consumed by each fish species in a week

	Total no. of	Total no. of unconsumed	Total no. consumed wingless	% age of wingless houseflies	
Species name	wingless houseflies supplied in a week	wingless houseflies	houseflies in a	consumed in a week $(A + CE)$	
	11	in a week	week	(Average $\pm$ SE)	
Punitus ambassis (8.7cm) (98.9g)	490	391	99	20.20±0.167	
Punitus sarana (14.6cm) (10.90g)	490	311	179	36.53±0.402	
Clarias batrichus (20.5cm) (200g)	490	109	381	77.75±0.601	
wallago attu (26cm) (556g)	490	22	468	95.51±0.372	

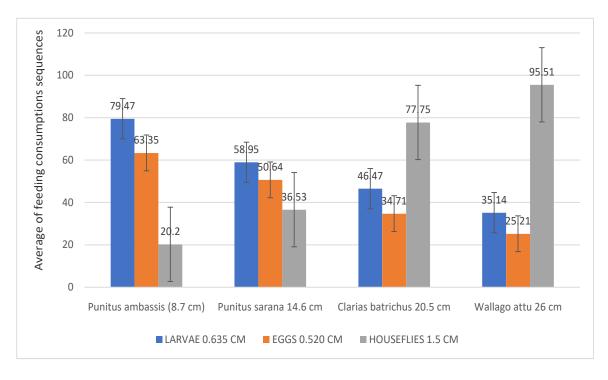


Fig. 1: Average  $\pm$  SE of larvae, eggs, and housefly's feeding efficacy consumption sequences by native fish species during the entire three experiments

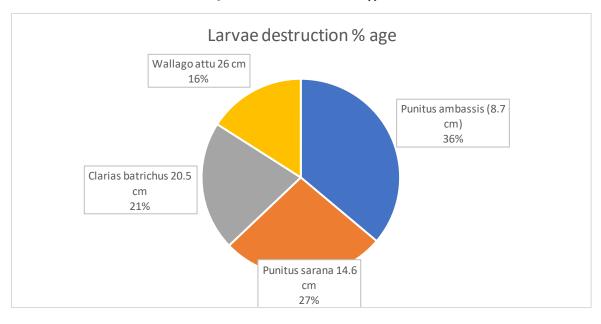


Fig. 2: Larvae feeding consumption (destruction) percentages by four native larvivorous fish species

## 4. CONCLUSION

Larvae feeding consumption was positively related to the body size of fishes. Small fishes mostly prefer small prey as food while large-sized fishes prefer large prey as food like wingless houseflies for their survival. Thus, the small-sized fish species bears excellent power of predation efficacy on mosquito larvae and eggs, while large-sized fishes possess less ability of predation efficacy on mosquito larvae and eggs but high predation consumption on wingless houseflies. Wallagu attu and Clarias batrichus showed the predatory nature inside the experimental water aquariums, as they showed vigorous behaviour of carnivory towards the wingless houseflies. Thus, according to the body size of fishes and ability to feed consumption efficacies averages, the sequence of the larval consumption feeding efficacy sequence (LCFES) was noted in this present study is Punitus ambassis 79.047±0.259 >Punitus sarana 58.95±0.258 >Clarias batrichus 46.47±0.404 >Wallago attu 35.14± 0.305. Out of the four native fish species, the most destructive for mosquito larvae populace is Punitus ambassis followed by Punitus sarana and the least destructive is *Wallagu attu* followed by *Clarias batrichus*.

According to the results of our present study, it is prophecy, that the endemic fish species of Gwalior Madhya Pradesh namely *Punitus ambassis* and *Punitus sarana* are likely to be impressive and effective in consuming mosquito eggs and larvae in breeding places. As the size of the mosquito eggs and larvae are small, hence are easily trapped and eaten by the small selected larvivorous fish species namely *Punitus ambassis* and *Punitus sarana*. It has been further scrutinized that, these two small natural fish species possess the ability to survive easily in small household fish tanks containing a low depth of water (5L), making them ideal for introduction in small water bodies and household coolers as mosquito larvae consumer.Hence native larvivorous fish species must be used for mosquito biocontrol programs as they are less harmful to other endemic species in their local ponds than that of using exotic fish species.

#### Abbreviations:

LFCES: Larval feeding consumption efficacy sequence. EFCES: Egg feeding consumption efficacy sequence.

HFCES: Houseflies feeding consumption efficacy sequence.

PA: Punitus ambassis, PS: Punitus sarana, CB: Clarias batrichus, WA: Wallago attu

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