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Comparative study of gender based larvicidal potency of *Punitus ambassis, Ompak bimaculatus, Chana murulius and, Heteropneustes fossalis* native fish species of Gwalior (M.P): A laboratory examination

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Abstract
From the 1900 centaury, mosquito control has become a point of focus in scientific communities epically health-providing units. All most all, tropical and sub-tropical countries have to face mosquito-borne ailments and infectious diseases every year like dengue fever, malaria, filariasis, Japanese encephalitis, skin irritation, etc. To overcome the vectors of these dreadful diseases at the source level various chemical methods have been employed so for but have certain environmental issues. Biological control is the best alternative, cost-effective, and eco-friendly method for mosquito control at the source level. The present study is focused on using some common native fish species of Gwalior (M.P) as a predator of mosquito larvae. All Predation experiments were conducted against mosquito larvae, pupae, and alternative food (aquatic insects) at varying pray densities in separate aquariums containing analyzed pond waters of Jal-vihar and fish form of Gwalior (M.P). The time duration of each predation experiment is three days, and in each day, fishes were fed with mosquito larvae, pupae, and alternative food at varying densities at different time intervals of the experimental day. The study revealed that Larvae consummation was positively related to the small fish species having female gender and prefers mostly small prey as food. Thus, small-sized female fish species possess greater predation efficacy on mosquito larvae and the sequence of larvicidal potency of female fish species was noted in the present study is *Punitus ambassis*>*Ompak bimaculatus*> *Chana murulius*> *Heteropneustes fossalis*. As per the results of our study, it is predicted that females have a greater value of larvicidal potency than that of males. Thus, female gender plays an important and positive role in mosquito biocontrol at the source level.

Keywords: zooplankton, gender, biocontrol, potency, eco-friendly

1. Introduction
Mosquitoes are blood-sucking dipteran insects that act as a vector of several important life-threatening diseases, including Malaria, Lymphatic Filariasis, chikungunya, dengue fever, and Japanese encephalitis [1, 2, 3, 19]. While feeding on blood, they may become carriers for many animal diseases, transmitting harmful zoonotic and other important diseases [4-7]. For oogenesis female mosquitoes feed on host blood and find their way to swamps, ponds, and marshy areas where females lay eggs. Ephemeral aquatic ecosystems (pools, puddles, ponds, floodplains, etc.) become ideal breeding sites for mosquitoes. Thus, mosquitoes are originating from such water bodies, hence we can say these water bodies are the actual sources of mosquitoes. Among the live feeds that fish consumes, mosquito larvae are one of the most favourite food items for the larvivorous fish. Fish that are predators of the immature stages of mosquitoes are referred to as larvivorous fish. A biological control refers to the introduction or manipulation of animals to suppress the population of a vector. A wide range of organisms helps to regulate mosquito populations naturally through predation, parasitism, and competition.
Among all the biological control agents, larvivorous fish are the most common and widely used in vector control. In India, an exotic larvicidal fish (*Gambusia affinis*) is used as a predator of mosquitoes which is currently running in many rivers and other water bodies. This exotic mosquitofish is quite effective in minimizing mosquito larvae, but it has also certain environmental issues related to ecological imbalance, possible impacts on native fish species,

and other effects on the biodiversity of the water bodies. Since mosquitoes are considered dangerous and are responsible for direct and indirect damages, their control has always been focused in community scientific studies. Mosquito-borne diseases have been a major problem in almost all tropical and subtropical countries and currently, there are no successful vaccines against most such diseases. These diseases not only become a burden in our daily life but are also responsible for more than 3 million deaths per year [8, 9, 38]. To overcome these diseases, the reduction in the population size of mosquitoes is essential. Many synthetic insecticides are widely used for controlling adult and larval mosquito populations, which is harmful to human health and non-target animals and arthropod populations and an important cause of environmental pollution. Chemical control methods were used to eradicate them but due to the intense use of these chemicals, mosquitoes become less effective because of the development of resistance property in their genetic material [10-14].

Scientists have negated serious ecological consequences upon complete eradication of mosquitoes, creating room for developing the safest and environmentally friendly control methods [8, 9, 15-18]. Among them, biological control is the deliberate use of natural enemies to reduce the number of pest organisms, which have gained acceptance for controlling nuisance [20, 21, 22]. Biological larvicides are the safest and attractive methods in mosquito control and this is by use of biological agents (e.g., mosquito fish) that eat or destroy the mosquito larvae.

In India, an exotic larvicidal fish (*Gambusia affinis*) is used as a predator of mosquitoes which is currently running in many rivers and other water bodies. This exotic mosquitofish is quite effective in minimizing mosquito larvae, but it has also certain environmental issues related to ecological imbalance, possible impacts on native fish species, and other effects on the biodiversity of the water bodies. This study will emphasize on the use of native larvivorous fish species of India with special reference to Gwalior (M.P) to evaluate their larvivorous potential property so that we can use them for malaria biocontrol programs. Thus, the study envisages assaying and providing an inventory of biological control. It is considered to be not only the best low price, eco-friendly method but highly effective in vector management as compared to the other chemical methods.

2. Materials and Methods

During this study, attempts were made to evaluate the comparative larvicidal potency on the gender basis of some common native fish species against mosquito pupae, larvae, and aquatic insects as alternative food, under laboratory conditions.

2.1 Collection, maintenance, and identification of fishes

Twelve fishes of four native fish species namely *Punitus ambassis*, *Ompak bimaculatus*, *Chana murulius*, *Heteropneustes fossalis* of the Gwalior region have been brought alive in fish shipping boxes from the Tigra Dam which is located about 23kms away from Gwalior (M.P), India. Before experiments in the laboratory, the fish species are segregated and are maintained separately species-wise, acclimatize them for seven days under laboratory conditions, and are kept in glass containers of aerated tap water and are fed with commercial fish feed (tubifex worms) and pieces of earthworms. Fishes are further identified with the help of standard taxonomic keys of Rringuelet *et al*, (1967) and Ghedotti (1998).

2.2 Collection of mosquito eggs

The mosquito eggs have been collected from local (in and around Gwalior city (26.22˚ N 78.18 ˚E) shady pond, water stored containers, and stagnant drains and ditches with the help of mosquito egg collecting net and O type brush. The eggs have been brought to the laboratory and have been transferred to 18 cm × 13 cm × 4 cm size enamel trays containing 500 ml of pond water.

2.3 Collection and maintenance of mosquito larvae

The mosquito larvae have been collected in September to get maximum larvae by dipping, netting, and pipetting methods from shallow ponds, water stored containers, ditches, drains and other stagnant water bodies adjacent to the Gwalior city (26.22˚ N 78.18˚ E) and have been brought to the laboratory in plastic buckets and they were sieved to remove the phytoplankton's, zooplanktons and other dried leaves and are then transferred to glass containers and are reared with supplementary food consisting of protein biscuit (60%) and dried yeast powder (40%).

2.4 Collection of aquatic insects

Water insects are simply collected from local water bodies adjacent to the Jiwaji University Gwalior (M.P) as per S.C. Goswami (2004) procedure of insect collection.

2.5 Predation experiments

As this work is laboratory-based, the experimental design intends to stimulate natural conditions in fish glass aquariums (25cm×20cm×20cm) by filling the laboratory aquariums with the analyzed pond water. Acclimatized fish species are weighed and allowed to release into separate aquariums in the morning hours. All the predation experiments carried in 10 L glass aquariums (25cm×20cm×20cm) half-filled with analyzed pond waters. Each experiment is carried out separately in triplicates. The duration of each predation experiment is one week, and all the four species namely *Punitus ambassis*, *Ompak bimaculatus*, *Chana murulius and*, *Heteropneustes fossalis* are fed with an equal number of eggs, larvae, and alternative food in different experiments at different intervals of time in each day of a week. After the feeding duration of each experiment, the number of foods left in the experimental aquariums was counted and removed on daily basis at the end of the experiment and the native larvivorous fish species are removed from experimental aquariums to plastic buckets (5L) and are fed with commercial feed (tubifex worms) and small pieces of Earthworm (*Eisenia fetida*) during the night. The data obtained from each of the experiments was analyzed statistically.

3. Statistical analysis

MS excel 2007 (Average ± Standard Error) is the tool that was used to analyze the data, obtained during the experimental days of our research.

4. Result and Discussion

Reduction of mosquito has been tried by various methods, however, due to certain drawbacks, related to environmental

issues; there is a shift in approach to control the mosquito population. Due to the ban or restrictions by environmental protection agencies, there are now fewer chemicals available than there have been for the last 20 years. [23-25] Thus, the biological control of mosquitoes has become more practical [26-30]. In this regard, larvivorous fishes have been used worldwide for controlling mosquito larvae [31-33]. Researchers all around the world have evaluated indigenous fish species to identify appropriate local biological control agents [15]. Several studies have suggested the introduction of fish mosquito fish species, *Gambusia affinis, and Poecilia reticulata*, and indigenous species to be effective in minimizing mosquito populations breeding places [15, 34-36]. Concern has also been felt about introducing exotic fish for mosquito control due to their possible impacts on native fish species. The introduction *of Gambusia affinis* in Greece (Europe) led to the decline of the endemic fish species *Valencia letourneuxi* [34, 37] and similar findings were reported in United States, Spain, and Australia [39-41] Keeping above mentioned concerns in mind, in this study, native fishes was tested for its larvicidal potential and predatory potential.

Our three predation experiments under laboratory conditions convinced us that the native fishes have also greater potential to be used as biological control agents for mosquito larvae and other pests. The first experiment was performed on mosquito larvae as a predation experiment on three separate days. Equal numbers of larvae were fed to both genders of different species of native fishes on daily basis at different intervals of time in a day. Larvae consumption data of both females and male fishes are given in (Table 1.)

Table 1: Larvae consumption by males and females at different intervals of time on separate days.

Species name and body size	Gender	Experimental days	No. of larvae supplied at 9 am	No. of larvae supplied at 2 pm	Total no. of larvae supplied in a day	Total no. of larvae consumed in a day	Total no. of unconsumed larvae in a day
Punitus ambassis (8.5cm)	Female	1	30	40	70	70	0
		2	30	40	70	68	2
		3	30	40	70	70	0
Punitus ambassis (8.2cm)	Male	1	30	40	70	65	5
		2	30	40	70	67	3
		3	30	40	70	63	7
Ompak bimaculatus (12.3 cm)	Female	1	30	40	70	65	5
		2	30	40	70	62	8
		3	30	40	70	58	12
Ompak bimaculatus (12.cm)	Male	1	30	40	70	63	8
		2	30	40	70	60	10
		3	30	40	70	55	15
Chana murulius (16.3 cm)	Female	1	30	40	70	52	18
		2	30	40	70	50	20
		3	30	40	70	53	17
Chana murulius (16 cm)	Male	1	30	40	70	42	28
		2	30	40	70	50	20
		3	30	40	70	45	25
Hetreopneustes fossalis (18.5 cm)	Female	1	30	40	70	42	27
		2	30	40	70	40	30
		3	30	40	70	39	31
Hetreopneustes fossalis (18cm)	Male	1	30	40	70	40	30
		2	30	40	70	35	35
		3	30	40	70	30	40

The second experiment was performed on mosquito eggs as a predation experiment on three separate days. An equal number of eggs were fed to both genders of different native fish species on daily basis at different intervals of time in a day. Egg consumption data of both females and male fishes are given in (Table 2.)

Table 2: Egg consumption by males and female fish species at different intervals of time on separate days.

Species name and body size	Gender	Experimental days	No. of eggs supplied at 9 am	No. of eggs supplied at 2 pm	Total no. of eggs supplied in a day	Total no. of eggs consumed larvae in a day	Total no. of unconsumed-d eggs in a day
Punitus ambassis (8.7cm)	Female	1	25	35	60	58	2
		2	25	35	60	60	0
		3	25	35	60	59	1
Punitus ambassis (8.4cm)	Male	1	25	35	60	55	5
		2	25	35	60	56	4
		3	25	35	60	52	8
Ompak bimaculatus (12.5 cm)	Female	1	25	35	60	54	6
		2	25	35	60	51	9
		3	25	35	60	51	9
Ompak bimaculatus (13.cm)	Male	1	25	35	60	50	10
		2	25	35	60	51	9
		3	25	35	60	48	12

Chana murulius (16.5cm)	Female	1	25	35	60	50	10
		2	25	35	60	43	17
		3	25	35	60	42	18
Chana murulius (16.2 cm)	Male	1	25	35	60	41	19
		2	25	35	60	41	19
		3	25	35	60	38	22

The third experiment was performed on aquatic insects as a predation experiment on three separate days to determine the carnivorous nature of the native fish species. Equal numbers of aquatic insects were fed to both genders of different native fish species on daily basis at different intervals of time in a day. Aquatic insect consumption data of both females and male fishes are given in (Table. 3) & the average value of all consumptions is given in (Table. 4).

All three predation experiments were performed to analyze the comparative larvicidal potency and carnivorous nature of the native fish species based on gender. The total number of unconsumed food material was counted every day in the end, to get the appropriate food consumption rates on each day of the experiment.

Table 3: Aquatic insects' consumption by males and female fish species at different intervals of time on separate days.

Species name and body size	Gender	Experimental days	No. of aquatic insects supplied at 9 am	No. of aquatic insects supplied at 2 pm	Total no. of aquatic insects supplied in a day	Total no. of consumed aquatic insects in a day	Total no. of unconsumed aquatic insects in a day
Punitus ambassis (8.5cm)	Female	1	10	5	15	2	13
		2	10	5	15	3	12
		3	10	5	15	1	14
Punitus ambassis (8.2cm)	Male	1	10	5	15	2	13
		2	10	5	15	2	13
		3	10	5	15	1	14
Ompak bimaculatus (12.3 cm)	Female	1	10	5	15	10	5
		2	10	5	15	10	5
		3	10	5	15	11	4
Ompak bimaculatus (12.cm)	Male	1	10	5	15	11	4
		2	10	5	15	12	3
		3	10	5	15	10	5
Chana murulius (16.3 cm)	Female	1	10	5	15	15	5
		2	10	5	15	13	7
		3	10	5	15	13	7
Chana murulius (16 cm)	Male	1	10	5	15	11	4
		2	10	5	15	13	2
		3	10	5	15	12	3
Hetreopneustes fossalis (18.5 cm)	Female	1	10	5	15	15	0
		2	10	5	15	15	0
		3	10	5	15	15	0
Hetreopneustes fossalis (18cm)	Male	1	10	5	15	14	1
		2	10	5	15	14	1
		3	10	5	15	13	2

Table 4: Average no. of eggs, larvae, and aquatic insect consumptions by males and female fishes at different intervals of time entire experimental days.

S.no	Species name	Mean ± *SD* of eggs, larvae and aquatic consumption by fishes					
		Males			Females		
		Eggs	Larvae	Water insects	Eggs	Larvae	Water insects
1	*Punitus ambassis*	54.33±0.98	65±0.94	1.66±0.27	59±0.47	69.33±0.54	2±0.47
2	*Ompak bimaculatus*	49.66±0.72	59.33±1.90	11±0.47	52±0.81	61.66±1.65	10.33±0.27
3	*Chana murulius*	40±0.81	45.66±1.90	12±0.47	45±2.05	53.33±0.42	13.66±0.54
4	*Hetreopneustes fossalis*	30±0.94	35±2.35	13.66±0.27	35.66±1.90	40.33±0.72	15±0

The data on feeding consumptions averages with standard deviations (SD) of both the genders are given in (Table. 4). Three feeding consumption sequences of female genders are larvae consumption sequence P.A 69.33 ± 0.54 > O.B 61.66 ±1.65 > C.M 53.33 ±0.72 > H.F 40.33 ± 0.72, egg consumption sequence P.A 59 ± 0.47 > O.B 52 ± 0.81 > C.M 45 ± 2.05 > H.F 35.66 ± 1.90, and aquatic insect consumption sequence P.A 2 ± 0.47 < O.B 10.33 ± 0.27< C.M 13.66 ± 0.54 < H.F 15 ± 0 and three male gender feeding consumption sequences are larvae consumption sequence P.A 65 ± 0.94 > O.B 59.33 ± 1.90 > C.M 45.66 ± 1.90> H.F 35 ±2.35, egg consumption sequence P.A 54.33 ± 0.98 > O.B 49.66 ± 0.72 > C.M 40 ± 0.81 > H.F 30 ± 0.94 and aquatic insect consumption sequence P.A 1.66 ± 0.67 < O.B 11 ± 0.47 < C.M 12 ± 0.47< H.F 13.66 ± 0.27.

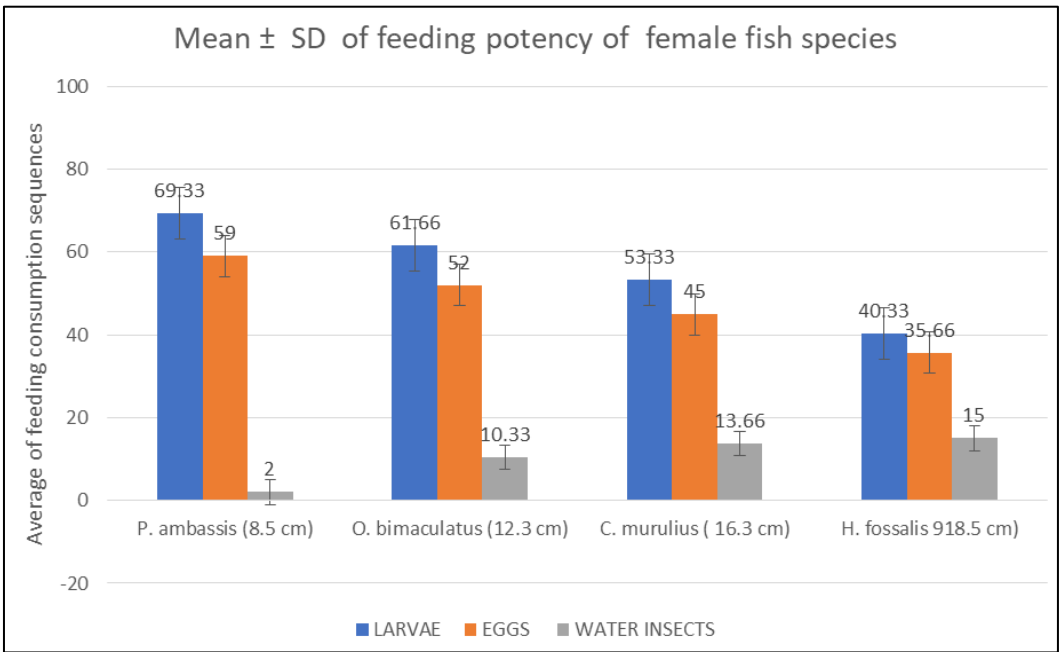

Fig 1: Comparative feeding potency of female genders of native fish species upon larvae, eggs, and aquatic insects.

Based on the averages of feeding consumption sequences of males and females, we plotted two graphs (fig.1 and fig.2) separately for males and females. Females possess greater larvicidal potency as compared to males. The body size of the fish species also affects their larvivorous potency. The small fish species have a greater value of larvicidal potency than that of large fishes. Small fishes usually select small prey as food like mosquito larvae and large fishes usually select largely prays as food like aquatic insects. Out of the four native species, *Punitus ambassis* possess greater power of larvivorous property followed by O*mpak bimaculatus*, because of their small body sizes. While the other two species namely *Chana murulius* and *Heteropneustes fossalis* show less interest in larvae because their body size is larger and hence usually prefers large food like aquatic insects for their survival. Thus, the carnivore's sequence of the native fishes is PA<OM<CM <HF. This sequence shows that the carnivorous nature of these four native fish species increases as the fish size increases.

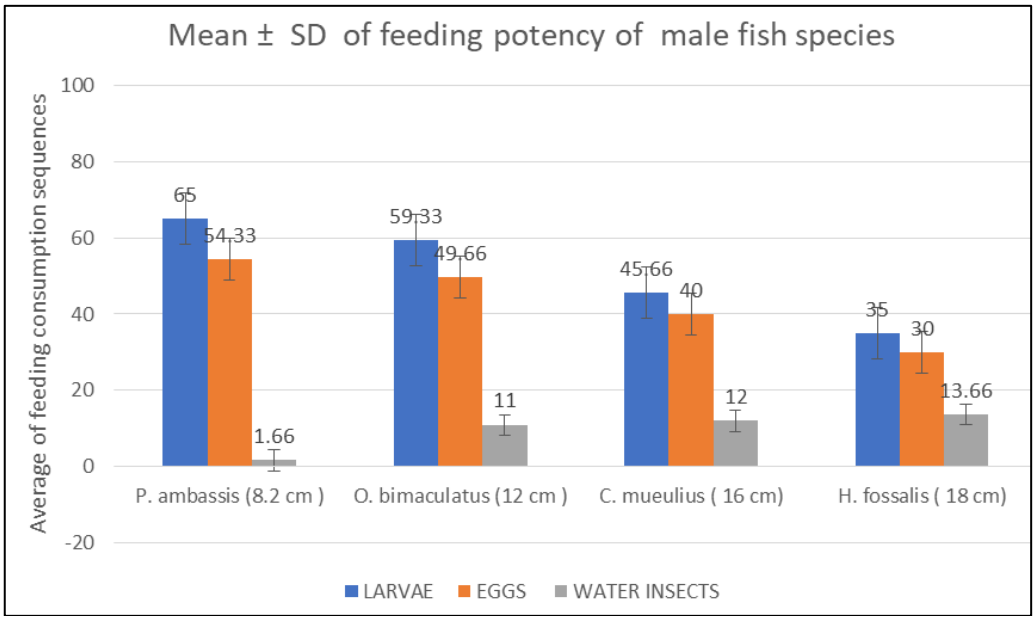

Fig 2: Comparative feeding potency of male genders of native fish species upon larvae, eggs, and aquatic insects.

5. Conclusion

The comparative study of larvivorous native fish species revealed that Larvae consummation was positively related to the small fish species having female gender and prefers mostly small prey as food. Thus, small-sized female fish species possess greater predation efficacy on mosquito larvae and the sequence of predation potency of female fish species was noted in the present study is *Punitus ambassis*> *Ompak bimaculatus*> *Chana murulius*> *Heteropneustes fossalis*. As per the results of our study, it is predicted that females have a greater value of larvicidal potency than that of males. Thus, female gender plays an important and positive role in

mosquito biocontrol at the source level. The indigenous larvivorous fish species showed excellent results with high predation efficiency and good survival ability in small volumes of water containers. Besides this, they don't cause any harm to other native species of fishes and also breed naturally. As the size of the mosquito larvae and eggs are minute, hence are easily predated by the small native larvivorous fish species especially with feminine character. The carnivorous nature of the female native larvivorous fish species increases as the body size of the fish species increases. Large-sized native larvivorous fish species feed voraciously on large prey like aquatic insects etc. Out of all the four native larvivorous species of Gwalior (M.P) selected for larvivorous activity, *Punitus ambassis* with female gender seems the most effective and powerful in larvicidal potency followed by *Ompak bimaculatus*. Thus, the small the female larvivorous fish, the more will be its larvivorous property than that of male larvivorous fish. Female genders of native larvivorous fish species are more powerful than that of male genders in mosquito-biocontrol. Biological control by fishes is considered effective, eco-friendly, low-cost, and safe to human and other non-target population and suggested that only native fish species should be used as biological control of mosquitoes to avoid the invasive nature of exotic species such as Gambusia and Poecilia [41, 44, 48, 49].

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7. References

1. Sarwar M. Insect-borne diseases transmitted by some important vectors of class insecta hurtling public health. International Journal of Bioinformatics and Biomedical Engineering. 2015;1(3):311-317.
2. Ghosh SK, Dash AP. Larvivorous fish against malaria vectors: a new outlook. Transactions of the Royal Society of Tropical Medicine and Hygiene 2007;101(11):1063-1064.
3. Foster WA, Walker ED. Mosquitoes (Culicidae). In Medical and veterinary entomology Academic press 2019, 261-325.
4. Anyaele O, Obembe A. Comparative studies of the feeding capacity and preference of *Aphyosemion gularis* (Boulenger 1901) on some aquatic macroinvertebrates. Journal of vector borne diseases 2011;48(4):231.
5. Lehane MJ, Lehane M. The biology of blood-sucking in insects. Cambridge University Press 2005.
6. Russell RC. Constructed wetlands and mosquitoes: health hazards and management options an Australian perspective. Ecological Engineering, 1999;12(1, 2):107-124.
7. Hancock PA, Sinkins SP, Godfray HCJ. Strategies for introducing Wolbachia to reduce transmission of mosquito-borne diseases. PLoS Negl Trop Dis 2011;5(4):e1024.
8. Ramírez-Lepe M, Ramírez-Suero M. Biological control of mosquito larvae by *Bacillus thuringiensis* subsp. israelensis. Insecticides-Pest Engineering 2012.
9. Lacey LA, Frutos R, Kaya HK, Vail P. Insect pathogens as biological control agents: do they have a future? Biological control 2001;*21*(3):230-248.
10. Culbert NJ, Balestrino F, Dor A, Herranz GS, Yamada H, Wallner T *et al.* A rapid quality control test to foster the development of genetic control in mosquitoes. Scientific Reports 2018;8(1):1-9.
11. Liu N, Xu Q, Zhu F, Zhang LEE. Pyrethroid resistance in mosquitoes. Insect Science 2006;13(3):159-166.
12. Zaman MS. Malarial control through fish. Pakistan Journal of science 1980;32:163-168.
13. Holt RA, Subramanian GM, Halpern A, Sutton GG, Charlab R, Nusskern DR *et al*. The genome sequence of the malaria mosquito *Anopheles gambiae*. Science 2002;298(5591):129-149.
14. Gu W, Novak RJ. Habitat-based modelling of impacts of mosquito larval interventions on entomological inoculation rates, incidence, and prevalence of malaria. The American journal of tropical medicine and hygiene 2005;73(3):546-552.
15. WHO Study Group. WHO vector control for malaria and other mosquito borne diseases? WHO Tech. Rep. Ser 1995, 857.
16. Cabral JA, Mieiro CL, Marques JC. Environmental and biological factors influence the relationship between a predator fish, Gambusia holbrooki, and its main prey in rice fields of the Lower Mondego River Valley (Portugal). Hydrobiologia 1998;*382*(1-3)41-51.
17. Louca V, Lucas MC, Green C, Majambere S, Fillinger U, Lindsay SW. Role of fish as predators of mosquito larvae on the floodplain of the Gambia River. Journal of medical entomology 2014;46(3):546-556.
18. Phukon HK, Biswas SP. An investigation on larvicidal efficacy of some indigenous fish species of Assam, India. Adv Biores 2013;4(3):22-5.
19. Ghosh A, Mandal S, Bhattacharjee I, Chandra G. Biological control of vector mosquitoes by some common exotic fish predators. Turkish journal of biology 2005;29(3):167-171
20. Chala B, Erko B, Animut A, Degarege A, Petros B. Assessment of *Carias gariepinus* as a biological control agent against mosquito larvae. BMC ecology 2016;16(1):27.
21. Kumar R, Hwang JS. Larvicidal efficiency of aquatic predators: a perspective for mosquito biocontrol. Zoological Studies-Taipei 2006;45(4):447.
22. Ramírez-Lepe M, Ramírez-Suero M. Biological control of mosquito larvae by *Bacillus thuringiensis* subsp. israelensis. Insecticides-Pest Engineering 2012.
23. Walker K. A review of control methods for African malaria vectors. Environmental Health Project 2002;2:618-627.
24. Vilela CLS, Bassin JP, Peixoto RS. Water contamination by endocrine disruptors: Impacts, microbiological aspects and trends for environmental protection. Environmental pollution 2018;235:546-559.
25. Effects of the removal or reduction in density of the malaria mosquito, *Anopheles gambiae* species on interacting predators and competitors in local ecosystems. Medical and veterinary entomology 33(1), 15.
26. Collins CM, Bonds JAS, Quinlan MM, Mumford JD. Effects of the removal or reduction in density of the malaria mosquito, *Anopheles gambiae* species on interacting predators and competitors in local ecosystems. Medical and veterinary entomology 2019;33(1):1-15.

27. Angelon KA, Petranka JW. Chemicals of predatory mosquitofish (*Gambusia affinis*) influence the selection of oviposition sites by Culex mosquitoes. Journal of chemical ecology 2002;28(4):797-8
28. Mohamed AA. Study of larvivorous fish for malaria vector control in Somalia, 2002. EMHJ-Eastern Mediterranean Health Journal 2003;9(4):618-626, 2003.
29. Chandra G, Ghosh A, Bhattacharjee I, Ghosh SK. Use of larvivorous fish in biological and environmental control of disease vectors. Cameron MM, Lorenz LM, eds 2013, 25-41.
30. Aditya G, Pal S, Saha N, Saha GK. Efficacy of indigenous larvivorous fishes against *Culex quinquefasciatus* in the presence of alternative prey: Implications for biological control. Journal of Vector-Borne Diseases 2012;49(4):217.
31. Kendie FA. Potential biological control agents against mosquito vector in the case of larvae stage: A review. World News of Natural Sciences, 2020;28:34-50.
32. Bano F, Serajuddin M. Comparative Study of Larvicidal Efficacy of Four Indigenous Fish with an Exotic Top Water Minnow, *Gambusia affinis*. Journal of Ecophysiology and Occupational Health 2017;*16*(1, 2):7-12.
33. Bhattacharjee I, Aditya G, Chandra G. Laboratory and field assessment of the potential of larvivorous, air-breathing fishes as predators of culicine mosquitoes. Biological control 2009;49(2):126-133.
34. Hoddle MS. Restoring balance: using exotic species to control invasive exotic species. Conservation Biology 2004;18(1):38-49.
35. Homski D, Goren M, Gasith A. Comparative evaluation of the larvivorous fish *Gambusia affinis* and *Aphanius dispar* as mosquito control agents. Hydrobiologia 1994; 284(2):137-146.
36. Kamareddine L. The biological control of the malaria vector. Toxins 2012;*4*(9):748-767.
37. Gu W, Novak RJ. Habitat-based modeling of impacts of mosquito larval interventions on entomological inoculation rates, incidence, and prevalence of malaria. The American journal of tropical medicine and hygiene 2005;73(3):546-552.
38. Fang W, Azimzadeh P, Leger RJS. Strain improvement of fungal insecticides for controlling insect pests and vector-borne diseases. Current opinion in microbiology 2012;15(3):232-238.
39. Kalogianni E, Koutsikos N, Vardakas L, Giakoumi S, Chatzinikolaou Y, Oikonomou A. Impacts of the alien mosquitofish on the abundance and condition of two Mediterranean native fish. Mediterranean Marine Science 2019;20(4):727-735.
40. Hurlbert SH, Mulla MS. Impacts of mosquitofish (*Gambusia affinis*) predation on plankton communities. Hydrobiologia 1981;*83*(1):125-151.
41. Lloyd LN. Ecology and distribution of the small native fish of the lower River Murray, South Australia and their interactions with the exotic mosquitofish, *Gambusia affinis* holbrooki (Girard) (Doctoral dissertation).
42. Baber MJ, Babbitt KJ. The relative impacts of native and introduced predatory fish on a temporary wetland tadpole assemblage. Oecologia 2003;136(2):289-295.
43. Martinez-Ibarra JA, Guillén YG, Arredondo-Jimenez JI, Rodriguez-Lopez MH. Indigenous fish species for the control of *Aedes aegypti* in water storage tanks in Southern Mexico. Bio Control 2002;*47*(4):481-486.
44. Miura T, Takahashi RM, Wilder WH. Impact of the mosquitofish (*Gambusia affinis*) on a rice field ecosystem when used as a mosquito control agent. Mosq. News 1984;44(4):510-517.
45. Morgan LA, Buttemer WA. Predation by the non-native fish Gambusia holbrooki on small *Litoria aurea* and *L. dentata* tadpoles. Australian Zoologist 1996;30(2):143-149.
46. Nelson SM, Keenan LC. Use of an indigenous fish species, *Fundulus zebrinus*, in a mosquito abatement program: a field comparison with the mosquitofish, *Gambusia affinis*. Journal of the American Mosquito Control Association 1992;*8*(3):301-304.
47. Noreen M, Arijo AG, Ahmad L, Sethar A, Leghari MF, Sethar GH *et al*. Biological control of mosquito larvae using edible fish. Int. J of Inov and App Res 2017;5(1-6):526.
48. Van Dam AR, Walton WE. Comparison of mosquito control provided by the arroyo chub (*Gila orcutti*) and the mosquitofish (*Gambusia affinis*). Journal of the American Mosquito Control Association 2007;23(4):430-442.
49. Verma AK, Pal RRRK, Tripathi L. Biological control of vector of communicable disease in Pawai, Panna MP using larvivorous fish, Gambusia. Biological Control 2016, 3(1).