Native Larvivorous Fish in an Endemic Malarious Area of Southern Iran, a Biological Alternative Factor for Chemical Larvicides in Malaria Control Program

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Abstract
Background: The widespread use of chemical insecticides, resistance in vectors and environmental problems, all have led to an increased interest in the use of biological agents in malaria control programs. The most important functional elements are the native fish. The aim of this study was to identify the native species of lavivorous fish in Rudan County, southern Iran, to introduce an effective species and to propose its’ implementation in the national malaria control program.

Methods: This ecologically descriptive study was conducted during 2011-2012 using random sampling from different fish habitats of Rudan County. The shoals of fish were caught using fishing net. Fish samples were then identified in the Ichthyology lab, Department of Fisheries and the Environment, Hormozgan University.

Results: Three species of larvivorous fish were identified as follows: Gambusia holbrooki, Aphaniusdispar dispar and Aphanius sp. The latter species has the most distribution in the study area and needs more morphological and molecular studies for identification at the species level.

Conclusion: Two species of native fish, i.e., A. dispar and A. sp. with larvivorous potential live in the area. Further studies on their predatory property are recommended in order to apply this local potential against malaria vectors in the area.

Keywords: Malaria, Biological control, Larvivorous fish, Aphanius, Iran

Introduction

Malaria is an endemic mosquito-borne disease in Iran, with active foci in southeastern part of the Country. Widespread use of chemical insecticides against malaria vectors which started about five decades ago, has resulted to insecticide resistance in some mosquitoes and therefore increased the environmental hazards by uncontrolled use of pesticides. This issue is one of the most important reasons for changing the national approaches in malaria campaign towards biological control methods for elimination of mosquito-borne diseases.

In addition, residual spraying has always been faced with problems such as human behavior, biology and ecology of vectors, insecticide resistance in target population of mosquitoes, op-
erational and financial defects. It also has side effects on non-target organisms as well. These problems caused the restriction of chemical methods and increased tendency to other control methods such as improving the environment, drainage, and integrated vector management. Due to the damaging effects of pesticides on beneficial insects, environmental pollution and contamination of food resources, as well as accumulation of insecticides in non-target organisms, governments are trying to improve pest control techniques.

Biological control is one of the ideal ways against vectors in natural and artificial habitats, because of the lack of environmental problems and cost-effectiveness (1). In the case of mosquitoes, biological control reduces their larval population using natural predators. In this method, certain larvivorous fish species have been used. Eastern Mediterranean Regional Office (EMRO) of WHO is promoting the integrated vector management methods; one of them is the use of fish. The advised fish species by this organization are from Poeciliidae, Cyprinidae, Cyprinodontidae and Cichlidae families (1,2). Therefore, national malaria control programs in different countries are supporting this strategy (3). Using biological agents in control of diseases have increased dramatically during the last two decades, while the most important agents against mosquito larvae are some Bacteria and larvivorous fish species (4). Gambusia affinis is a famous fish that is being used around the world for mosquito larval control. However, in some cases imported non-native fish may be a great threat to native species and could destroy the aquatic ecosystems. On the other hand, non-native fish, like Gambusia sp., may act as an opportunistic predator and feed on algae, zooplanktons, aquatic insects, fish and amphibian eggs. This behavior will lead to fundamental changes in non-native ecosystems (5). For these reasons, use of larvivorous fish in malaria control strategies has been revised. In a control program, selection of the species of a larvivorous fish varies, depending upon the type of larval environment. For example, Poecilia sp. is appropriate for a close ecosystem like well, while Gambusia sp. is more suitable for open ecosystems like pools and water canals (3). Tooth-carp fish, Aphanius spp., has an extent of distribution in brackish and fresh water bodies of Mediterranean region, Red sea, Persian Gulf and Arabian Peninsula. It lives in pools, lakes and small springs. The use of Gambusia fish in malaria control programs is a serious threat for native species like Aphanius in its distribution area. Gambusia fish was used for the first time in northern part of Iran to control the mosquito larvae (6). This fish is distributed along the coastal area of southern Iran. Aphanius dispar has one known subspecies in the area named A. dispar richardsoni, 1907, but it seems to have more subspecies. In Hormozgan, A. ginaonis lives in Geno warm mineral water of Bandar Abbas County (7, 8).

Study on native larvivorous fish will help us to have opportunity to use this biological agent without any change in the aquatic ecosystem and to find suitable alternative(s) for non-native fish such as Gambusia sp. as well as to discourage the use of chemical larvicides.

This study aimed to identify native larvivorous fish living in a malarious area of southern Iran, in order to find a suitable agent for biological control of mosquito larvae in the area.

Materials and Methods

Study area

Rudan with an area of 3257 square kilometers, is located in north of Hormozgan province, at a distance of 100 km from Bandar Abbas. The city center is located at 27° 27’ north latitude and 57° 11’ east longitude, and height above sea level is approximately 190 meters. Rudan is surrounded from the north with Hajjabad county and Kerman province, from the east with Kahanouj and Manoujan counties, from west to Bandar Abbas and from south by Minab County. Rudan has 3 cities, 4 districts, 10 rural districts and 187 villages with residents. This county is one of the agricultural centers of the Hormozgan Province (9). Climatically, Rudan is composed of mountainous and plain areas. There are two main rivers in the area,
Abnama and Jaghin, with a basin of about 6500 km².

**Study design**

An ecologically descriptive study was carried out in the aquatic habitats for fish living in different areas of Rudan County, during the year, 2012. For this purpose, a random sampling was conducted using fishing net from 13 aquatic habitats located in 4 districts of the study area (Markazi, Jaghian, Bika and Roodkhaneh) in 7 villages: Abnama, Deimatoon, Jaghian, Ziaratali, Hizbandagan, Janatabad and Berentin. These villages have different water bodies including river, swamp, pool and water canals. The observed data including name of district and village, as well as type of aquatic habitat were recorded in the relevant forms. Collected specimens were conserved in 75% ethanol and labeled. Species identification was carried out in Ichthyology laboratory, Department of Fisheries and Environment, Hormozgan University, Iran. Meristic characteristics of dorsal fin were used for species identification.

**Results**

During this study, 9 fish species from 4 families were collected and identified (Table 1).

| District | Habitat              | Family          | Species                  |
|----------|----------------------|-----------------|--------------------------|
| Markazi, Roodkhaneh | River, Swamp         | Balitoridae     | Paraschistura sargadensis |
| Markazi, Jaghian, Bika, Roodkhaneh | River, Water canal, Swamp | Cyprinidae      | Capoeta damascina         |
| Markazi | River                | Cyprinidae      | Luciobarbus kersin       |
| Markazi, Roodkhaneh | River, Swamp         | Cyprinidae      | Cyprinion watsoni         |
| Roodkhaneh | River, Swamp         | Cyprinidae      | Garra rufa               |
| Markazi, Roodkhaneh | River, Swamp         | Cyprinodontidae | Aphanius dispar dispar    |
| Markazi, Jaghian, Bika, Roodkhaneh | River, Water canal, Swamp | Cyprinodontidae | Aphanius sp.             |
| Jaghian  | Pool                 | Poeciliidae     | Gambusia holbrooki       |

Among them were three species of *Aphanius dispar dispar*, *Aphanius* sp. and *Gambusia holbrooki* which seem to have important roles in malaria vector control programs. Overall, Cyprinidae with 4 species had the highest frequency and diversity among the collected fish in Rudan county (Fig. 1), followed by Cyprinodontidae, Balitoridae and Poeciliidae. Figure 2 shows the spatial distribution of the species of fish caught in the study area.

![Fig. 1: Percentage abundance and diversity of fish families, Rudan County, 2012](https://example.com/image1.png)
Discussion

There are five malaria vectors in the southern part of Iran, i.e. Anopheles stephensi, An. culicifacies s.l., An. dthali, An. fluviatilis s.l. and An. superpictus s.l. (10). The last three species have exophilic and exophagic behavior, so indoor residual spraying (IRS), which is a common method in malaria vector control, is not effective against them. Therefore, larval management as well as the use of long lasting insecticide impregnated bednets (LLINs) are recommended to reduce their population and avoid their bites.

At present, predators and bacterial products such as Bacillus thuringiensis and B. sphaericus are the most important biological agents that can be used successfully against mosquito larvae. Provided the appropriate condition, fish are the best choice for performing the biological control against mosquitoes, because they have a high potential in this regard (11,12).

In this study, fish collected from two main larvivorous families were found to be G. holbrooki (Poeciliidae) as well as Aphanopus dispar and Aphanius sp. (Cyprinodontidae). The first species is non-native to the area. Almost 315 species in 7 genera are recorded as larvivorous fish around the world (3). Gambusia affinis has an extensive use in biological control of mosquito larvae. By the way, imported non-native fish sometimes poses great threat to native species in the aquatic ecosystems. Now, the use of Gambusia fish has no restriction in most malarious areas of south and southeast of the country. However, using non-native fish in some habitats is prohibited due to their negative effect on fauna and diversity of native fish, amphibians and other non-target organisms, as confirmed by studies on Poeciliidae fish, especially Gambusia sp (3, 4, 13). Observations in the present study showed the low abundance of aquatic insects like dragonflies in the habitats of this fish. In addition, it is confirmed by low abundance and species diversity for Aphanopus fish in those habitats. Therefore, it is necessary to avoid release of non-native fish into water bodies with native species.

We found Aphanopus dispar and Aphanius sp. in the study area. Studies in recent years have shown the high diversity of species belonging to Aphanopus genus, so that 10 species of this fish are reported from different parts of Iran including: A. dispar
**Aphanius dispar** as in basins of Persian Gulf and Oman sea, *A. ginaonis* Holly, 1929 from Geno warm water spring in south, *A. isfahanensis* Hrbek, Keivany & Coad, 2006 from Isfahan, *A. farsicus* Teimori, Esmaii & Richenbacher 2011 from Maharlu lake, *A. sophiae* Heckel, 1847 from Kor river Basin in Fars province, *A. vladykovi* Coad, 1988 from central areas of Zagros, *A. pluristriatus* Jenkins, 1910 from drainage water of Mond river of Bushehr province, *A. arakensis* from the sluices of Salt Lake in Arak County, central Iran; *A. mento* in Arvandrood basin, and *A. mesopotamicus* Coad, 2009 from the Tigris–Euphrates basin (8,14-18). However, little data exists on diversity of different populations of *Aphanius* in Iran, so it is important to do more studies in this field. So, more studies on the morphological and molecular aspects of taxonomy are recommended for accurate diagnosis of this fish at the species level in Hormozgan Province. *Aphanius* fish can be found in the lower river basins and sluices covered with aquatic plants. They have tolerance to a wide range of salinity and can tolerate organic/inorganic contamination and low oxygen level. Some species of this genus are reported from environments with high salinity that makes them uninhabitable for other species. These killifish are abundant in wetlands, saltwater and brackish coastal waters as well as stagnant and semi-stagnant shallow waters (18-20).

*Aphanius dispar* Holly, 1929 with local names such as Zebra fish, Flag tail fish, Long fin toothed crap belongs to the crap fish family, and prefers the salty waters at the coastal areas of Iran. The geographical distribution for this species is reported to be India, Eastern Mediterranean region, i.e. Pakistan, Iran, Iraq, United Arab Emirates, Saudi Arabia, Kuwait, Bahrain, Palestine, Oman and Yemen; as well as Djibouti, Eritrea, Egypt, Sudan, Ethiopia and Somalia in Africa. This species has controlled the mosquito larvae in the streams of Karachi, Pakistan (20-21). In addition, there is a report of successful use of this species in mosquito larvae control in a coastal area of Ethiopia (2). Other successful applications of this species are reported from Saudi Arabia against *Culex quinquefasciatus*, in urban areas of Djibouti against *Anopheles arabiensis* and *An. gambiae*, and in Ethio-pia against *An. culicifacies adenensis* and *An. arabiensis* (1,2,5). Using *A. dispar* in water tanks reduced 93% larval counts by day 7 and 98% by day 21. This study showed high larvivorous potential of this fish (21).

Because the preference of using native organisms in biological control programs, creating a data-bank for native species and improving knowledge on their ecology, biology as well as ability to hunt target pest is critical, especially in malarious areas. Hence, the use of native fish is recommended in the study area as well as other malarious foci of Hormozgan Province.

**Conclusion**

The mosquito density depends on limited aquatic habitats in semi-arid/arid areas of southern Iran. Therefore, proper management of larval habitats and the use of native biological agents, including *Aphanius* larvivorous fish can be an appropriate solution to the risk of malaria transmission problem. Having paid due attention to the effectiveness of the use of native fish in malaria control program in other countries such as Oman, this potential should be highlighted in Iran’s national program as well. Biological control will drop the use of insecticides and reduce the risk of resistance in mosquito vectors. The present study showed this potential in Hormozgan Province and this requires accurate research and planning for mass rearing and distribution of native fish.

**Ethical considerations**

Ethical issues (Including plagiarism, informed consent, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc.) have been completely observed by the authors.

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