Rapid Assessment of the Status of *Valencia letourneuxi*, the Greek Killifish

Final Report October 2006



Hellenic Centre for Marine Research



Zoological Society of London



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Hellenic Centre for Marine Research (H.C.M.R), Institute of Inland Waters

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1. Introduction

Valencia letourneuxi (Sauvage, 1880) is a distinctive endemic of the western Balkans (Stephanidis, 1974). It belongs to the small Mediterranean genus Valencia (family Valenciidae), which also includes V. hispanica (Valenciennes, 1846), endemic to the Valencian region of Spain (Planelles, 1996; Planelles & Reyna, 1996). In the past, there was some uncertainty about the taxonomic relationship of the two taxa and V. letourneuxi was often assigned to V. hispanica (Oliva 1965; Woeltjes, 1982). Today, the two taxa are accorded a separate specific status based on geographic and reproductive isolation, morphological variation and well established genetic differences (Vilwock et al., 1982; Perdices et al., 1996; Kottelat, 1997; Maltagliati et al. 2006). The genus Valencia is classified in the order Cyprinodontiformes (Parenti, 1981), which includes many widely distributed small fish, commonly known as killifishes. The term "killifish" derives from the Dutch word "kilde", which refers to a small puddle or creek, and describes the habitat of many species within this group. Earliest collections of V. letourneuxi, known as the Greek or the Corfu killifish, are from wetlands of the Corfu island (Sauvage, 1880). Later the species was reported from the Lefkas island and various aquatic systems in the mainland Greece in an area extending from North to South between the drainages of the rivers Kalamas (Thyamis) and Alfios (Stephanidis, 1939, 1974; Labhart, 1980; Das, 1985; Economidis, 1991, 1995; Bianco & Miller, 1989; Economou et al., 1999; Barbieri et al., 2000; Daoulas, 2003). Its historical range includes also wetlands around the lake Butrinto of southern Albania, where the species was recorded by Oliva (1961).

Throughout its range, V. letourneuxi has a disjunctive and spotty distribution, forming small localised populations isolated from each other. This fragmented range enhances the danger of extinction, prevents gene flow among populations and reduces the chance of the reestablishment of depleted populations by migration from other localities. Available records of occurrence (summarised by Das, 1985; Bianco & Miller, 1989; Barbieri et al., 2000, 2002a, 2002b) indicate the historical presence of V. letourneuxi in approx. 20 localities. In most localities, the species occurs at low densities and usually only a few individuals are collected when suitable habitat is sampled. Its patchy distribution and low population densities make the species vulnerable to environmental stochasticity and prone to extinction. In addition, V. letourneuxi exhibits a high habitat specificity that increases the probability of extirpation events to occur. The species inhabits springs, small streams and swampy areas with clear, cool, slow-flowing waters and requires rich aquatic vegetation for reproduction and shelter from predators (Das, 1985; Bianco & Miller, 1989; Barbieri et al., 2000). There are few habitat sites in the current landscape that satisfy these ecological requirements, which partly explains both the fragmented range of the species and its low local population densities. Having a relatively restricted distribution, being locally scarce and exhibiting strict habitat specialization, V. letourneuxi meets the criteria of rarity defined by Gaston and Lawton, (1990) and can be regarded as a threatened species highly susceptible to natural or human induced habitat change. However, it is still unclear which particular physicochemical and biological features are the most important determinants of habitat quality for V. letourneuxi, and to which extent natural causes and human activities are generating impacts on its specialised habitats.

In the last 30 years, the range and the number of populations of the species have been declining, most probably due to anthropogenic habitat modifications (Economidis, 1995; Barbieri *et al.*, 2000). The species does not appear to occur any longer in the islands Lefkas and Corfu (Economidis, 1995; Barbieri *et al.*,

2002a), despite its previously well-known occurrence there (Sauvage, 1880; Oliva 1965; Woelties, 1982, 1988). Broad-scale fish investigations in western Greece by HCMR ichthyologists in the 1990s, not specifically targeting this species, recorded its extirpation from at least one locality in mainland Greece (the Drepano marsh, Thyamis basin, first found there by Labhart, 1980) and raised the suspicion that other extirpation events may have occurred without yet being detected (Economou et al., 1999). Admittedly, there is no sufficient information to make a sound assessment of the conservation status of V. letourneuxi. However, because of its deteriorating status in recent years, the species has been included in the lists of the Bern Convention as an endangered and strictly protected species, and it has been characterised as endangered and priority species for conservation in Annex II of the Habitats Directive of the European Union (92/43/EEC). In 1996, this species was designated as Endangered by IUCN on the basis of few surviving populations which were presumed to be undergoing a continuing decline. Following a precautionary approach, the species was reclassified as Critically Endangered in the 2005 update of the IUCN Red List, based on restricted distribution, suspected population decline, projected decline in the next 10 years and impacts of introduced alien species. However, no conservation strategy for this species has yet been designed and no management measures have been implemented, except in NATURA 2000 designated areas which, however, cover only a small fraction of the species' range. Such measures (e.g. prevention of pollution, restrictions on land use) are not specifically addressed to V. letourneuxi and their effectiveness is unknown.

One of the obstacles in developing a realistic and effective conservation strategy for *V. letourneuxi* is that comprehensive data on the present-day distribution of the species and the status of its local populations are not available; another obstacle is that there is paucity of adequate or appropriate information on the biology and ecology of this species, especially as regards the traits and the environmental features that determine habitat profitability. These difficulties arise, at least partly, from the haphazard character of previous ichthyological investigations undertaken by HCMR, none of which specifically targeted this species. In fact, most of the data available for *V. letourneuxi* at the HCMR is the product of broader ichthyological investigations targeting many species or addressing issues of fisheries (Economou *et al.*, 1999, Economou *et al.*, 2001). In addition, the rarity of *V. letourneuxi* in most of its known habitats has prevented the collection of a sufficient number of individuals for population assessments and biological analysis.

This report presents the results of the first complete baseline investigation of *V. letourneuxi* aiming to provide appropriate information on its distribution, abundance, ecology, habitat conditions and local population status, upon which inferences about threats to the species, vulnerability to natural or human disturbances of its habitats and future viability prospects can be based. This information will also be used as a reference point for future surveys and can factor into the management plan for this species.

1.1 Project Background

The background of the investigation dates back to May 2004, when the ZSL hosted the first KilliTAG (Taxonomic Advisory Group) meeting over two days to assess the status of all European and Middle-eastern killifish species and prioritise those for conservation. The group of killifish experts from all over Europe identified *Valencia letourneuxi* as one of five priority species for the TAG to work with. An action plan was drawn up for this species that included a number of key objectives. One of the

objectives included working with local authorities within Greece to determine the status of the fish in the wild, in order to better recommend conservation actions.

In late 2004, the ZSL contacted the Hellenic Centre for Marine Research (HCMR), Institute of Inland Waters (IIW), to assess current knowledge on V. letourneuxi status and data availability on its distribution, abundance and ecology. IIW had done some sampling for this species but only as part of larger assessments and never as a focused investigation. Available distributional records spanned over several years but were patchy and had not been undertaken from a conservation perspective. The ZSL and the IIW jointly applied to EUAC (European Union of Aquarium Curators) for funding (7,700 euros) to undertake a rapid assessment of the known and suspected V. letourneuxi habitats in Greece. When funding was awarded in May 2005, the two organisations quickly assembled a team that could begin the field assessment. Due to other commitments, it was agreed to split the project into two sections. The first section would work out sampling methodology and cover two major river deltas in Greece's Western Central mainland (June 2005). These two areas were known to have V. letourneuxi historically but had not been thoroughly searched in recent years. These delta areas are vast, comprising many small rivers and cut irrigation channels and required a more in depth search to find the target species than in the other areas it was known or thought to exist. The second part of the survey took place later in the year (October 2005) and covered a larger number of water systems in north-western and south-western Greece.

1.2 Project Objectives

The project aimed to give baseline information on *V. letourneuxi* abundance, geographical distribution, ecological requirements and population status to assist conservation efforts.

Its stated objectives included: 1) to complete a comprehensive field survey of known, former and suspected habitats of *V. letourneuxi* in Greece, 2) to determine world-wide status of *V. letourneuxi*, 3) to conduct a preliminary assessment of *V. letourneuxi* habitats including potential threats, 4) to provide information to facilitate an update for IUCN Red List of Threatened Species and Fishbase, 5) to gather information and data to support the future drafting of a conservation management plan for *V. letourneuxi*, based on the findings of the survey.

We outline the development of these topics, in the context of the following questions:

• What is the current distributional range of *V. letourneuxi*? Does a comparison with the historical range of the species provide evidence of recession?

Which particular physical and biological features determine habitat profitability for this species?

• What habitat types are utilised by *V. letourneuxi*? Do they vary in their capacity to support the species?

Is the amount of habitat used by the species declining? To which extent do natural causes and human impacts contribute to the decline or limit population growth?

Is the species inherently vulnerable to disturbances due to its life history and biological characteristics? • Are there any unoccupied but suitable habitats for translocation, within or close to the species' range?

Is there sufficient connectivity between the localities in which the species occurs in terms of "water corridors" that can be effectively utilised for dispersal?

1.3 Project Team

Dr. Alcibiades N. Economou	Hydrobiologist-Fish Biologist, Scientific Co-ordinator (HCMR)			
Dr. Eleni Kalogianni	Biologist (HCMR)			
Stamatis Zogaris	Geographer-Biologist (HCMR)			
Sofia Giakoumi	Technician-Fish Biologist (HCMR)			
Roberta Barbieri	Fish Biologist (HCMR)			
Dr. MariaTh. Stoumboudi	Fish Biologist (HCMR)			
Y. Chatzinikolaou	Environmentalist (HCMR)			
Brian Zimmerman	Team Leader, Aquarium, (ZSL),			
	Coordinator of Killifish Taxonomic Advisory Group			
	Auvisory Group			

2. General Methodology

Field data were obtained during two field trips in June 2005 and October 2005 (22 field days). A total of 14 systems of Western Greece were sampled for *V. letourneuxi* presence, comprised of a total of 95 sampling sites in both known and suspected *V. letourneuxi* localities. These included springs, irrigation channels, river channels and stream outlets to the sea in both mainland and islandic areas.

2.1 Sampling Sites Selection

Using Greek ordinance survey maps, possible *V. letourneuxi* habitat sites were located, along with potentially favorable access points. Information from previously published material together with unpublished data from previous ichthyological surveys of HCMR biologists, have also proved useful in the sampling sites selection. Upon arriving at the area, local farmers and local officials were consulted to determine the exact location of possible *V. letourneuxi* habitats, some of them often accompanying the research team in the field and providing us with free access to their land, in the case of local farmers.





Consultation with local farmers.

I. Loizos, a local farmer, with two members of the team in the field.

Locating appropriate sites, in combination with the effort to sample as many and as diverse sites as possible, often meant that sampling lasted until dark (approximately 22:00).



Sampling at a late hour of the day.

2.2 Fish Sampling Methodology

The tendency of the fish to remain hidden in or near dense vegetation in water that was often fairly deep (over 1.5 meters), meant sampling methods had to be creative at many sites, therefore a variety of dip nets were tried with varying mesh sizes and configurations. A large net with extendible wooden handle that had the D-shaped frame at a right angle to the handle proved useful in deep sites or areas with dense floating vegetation. Smaller, traditional dip nets were used in areas with thick *Phragmites* reeds that the larger net could not access.



Sampling with a D-shaped frame net.

Sampling with a small mesh-size net.

In more accessible sites, seine netting was tried using two techniques. In one technique, an end of the net was circled around a cluster of aquatic plants and then dragged to shore. In the second technique, the net was run parallel to the shore and bank-side vegetation was disturbed to frighten the fish into the net.



Seine net sampling in a small stream.



Seine net sampling in a deep site.

Electrofishing was tried at some sites (Hans-Grassel GmbH battery powered backpack, Model IG200-2, DC (pulsed), 1,5 KW output power, 35-100 Hz, max. 850 V, Schönau, Germany) but water depth and access prevented its use at most sites. Snorkeling and underwater photography was also used to record the presence of the species at some sites but despite very clear water at most locations, access and thick sediments on the stream bottom quickly made the water turbid, so the use of this technique was also limited. As a result, a short visual observation period at the site usually preceded sampling.

It is important to note that due to the sampling difficulties encountered, the variability of the habitats sampled and the different methods used, sampling had only a semiquantitative character, in terms of abundance or density of the fish populations.



2.3 Fish Measurements and Field Protocols

Fish were photographed in a portable aquarium and after anesthetization, measured (standard length to nearest mm) and weighed (nearest 0,001 g). They were then returned to the water, with a small portion of the target and sympatric species preserved in 10% formalin (or 95% alcohol) for future investigation of dietary habits, gonad maturation cycle and genetic analyses.

A habitat description and pressure assessment protocol especially developed for the needs of the survey was used. More specifically, in the field protocol, records were taken of GPS, water temperature, flow, substrate, depth, width, vegetation, sample method used, water physicochemical parameters, species present and number of *V. letourneuxi* caught with sex ratio. All data obtained from field sampling were later incorporated to EXCEL spreadsheets.





Photographing the fish in the field in a portable aquarium.

Recording physicochemical parameters.

3. Project Results

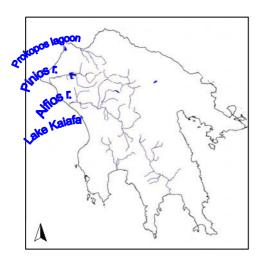
3.1. Analytical Project Results

Sampling sites' distribution, Pressures, Habitat characteristics, Fish species composition and Comparison of current versus historical presence of *V. letourneuxi* in each water system sampled, in the four major geographical regions of Western Greece, i.e. the Peloponnese, Western Central Greece, Ipeiros and the Ionian Islands.



Water Systems of the Peloponnese

- 3.1.1 Alfios river (and Lake Kaiafa)
- 3.1.2 Pinios river
- 3.1.3 Prokopos lagoon





3.1.1 ALFIOS RIVER

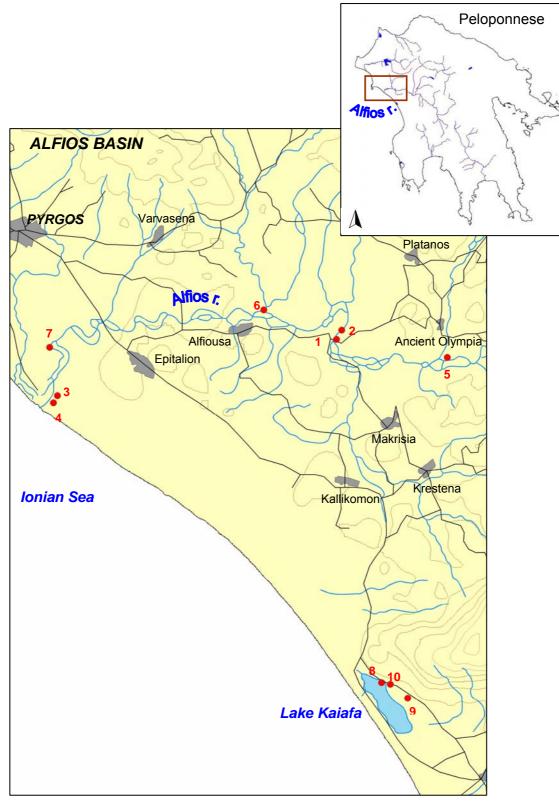
The Alfios river is the largest river of the Peloponnese both in terms of length (approx. length 120 km) and in discharge (average yearly discharge 66,6m³/sec at the Alfiousa bridge, 10 km from its mouth), with a basin area of 3.658 km² and a Delta area of 113 km². The river basin can be divided into three zones: the Upper Alfios, which drains the Arcadian plateaus, the Middle Alfios, draining the highland areas of Ilia, and the Lower Alfios, which drains the lowland area of Ilia. Due to the large size of its basin, and the high precipitation levels in the area, as well as its extensive karstic water supply, its base flow is high, with characteristic flooding phenomena. With its waters and its suspended sediments it has contributed to the creation of three lagoons, i.e. Agoulinitsa, Mouria (both now dried) and Kaiafa lake.

SAMPLING STATION DISTRIBUTION

The current survey focused on the lowland areas of the Alfios basin, with a total of 7 sampling sites, including the Kaiafa Lake area, with 3 sites. These included, apart from the Alfios proper, a tributary of Alfios (Lestenitsas river) and canals and riparian wetland areas (Table 1 provides a short description of these sites with the relevant pressure list and Map I shows the Alfios basin sampling sites).

TABLE 1. Sampling sites at the Alfios basin (red numbers indicate the species' absence, blue numbers the species' presence).

site	location	water body type	pressures
1	Alfiios dam, Alfios	Alfios river bank,	Dam hydropeaking
		meander-like	problems, gravel
		biotopes	extraction
2	Alfiousa, Alfios	Alfios river bank,	Dam hydropeaking
		secondary channel,	problems, gravel
		ponds, marshy edge	extraction
3	Zalos-South channel,	Canal	Agricultural
	Alfios		pollution,
			vegetation clearing,
			garbage disposal
4	Sotira –South channel,	Canal, near the river	Agricultural
	Alfios	mouth	pollution,
			vegetation clearing
5	Ancient Olympia, Alfios	Alfios river, oxbow	Agricultural
		ponds	pollution, gravel
			extraction, cement
			works
6	Lestenitsas river,	Lestenitsas river,	Gravel extraction
	confluence with Alfios gravelley wash with		
		islands	
7	North channel, Alfios	Canal	
8	Loutra, lake Kaiafa	Spring, canal	
9	Marsh, lake Kaiafa	Marshy area	Vegetation clearing
10	Springs, Lake Kaiafa	Sulfur springs	



MAP I. Sampling sites at the Alfios basin. Localities where *V. letourneuxi* was recorded (\bullet) – none in this system - and not recorded (\bullet) – sites 1-10.

PRESSURES

Frequent hydropeaking from the dam operation results in flooding/drying disturbances of the more inland sites sampled in this survey (such as sites 1 and 2, close to the Alfios dam, Fig.1, 2).



Fig.1. The Alfios dam.



Fig.2. Site 1 (top left) just beneath the Alfios dam.

In some inland sites gravel extraction also takes place, while several other sites, especially those closer to the river mouth, suffer from agricultural pollution, garbage disposal and vegetation clearing (sites 3 and 4, Fig. 3-6). Generally speaking, the major pressures for the lower parts of the Alfios system are irrigation works (water abstraction from the dam, as well as groundwater abstraction), gravel and sand extraction, some organic pollution from animal husbandry as well as pollution from urban waste disposal. At the lowland areas, high concentrations of nitrates, indicate agricultural pollution. Past drainage works have also altered significantly the delta area, notably with the drainage of Agoulinitsa and Mouria lagoons and the surrounding riverine wetlands.



Fig.3. Vegetation clearing and garbage disposal at site 3.



Fig.4. Trash disposal includes toxic chemical containers, such as this bottle of break fluid at site 3.



Fig.5. Disposed agrochemical containers at site 3.



Fig.6. Greenhouse near site 4.

HABITAT CHARACTERISTICS

In terms of habitat characteristics the Alfios sites can be subdivided in two habitat types. The first type includes relatively open riverine areas with meander-like habitats and oxbow ponds close to the river banks (sites 1, 2, 5, 6), with *Typha spp.* and *Arundo spp.* as riparian vegetation and very little aquatic vegetation (when present, *Potamogeton spp.* and some other aquatic plants) (Fig. 7-12).



Fig.7. Site 1 with Typha spp.



Fig.9. Open riverine area with dense riparian forest at site 5.



Fig.8. Site 1 with *Salix spp.* and *Phragmites* reeds.



Fig.10. Oxbow pond at site 5.

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Fig. 11. The confluence of Alfios river with Lestenitsas river (top right).



Fig. 12. Site 6, the Lestenitsas river, with *Salix spp., Arundo spp.* and *Phragmites* reeds.

The second habitat type includes canals feeding Alfios river (sites 3, 4, 7) with reeds *Arundo spp., Phragmites australis* and *Typha spp.* and a much richer aquatic vegetation of *Potamogeton spp., Ceratophyllum spp.*, water cress-like aquatic plants and notably *Lemna spp.* (Fig.13-16).



Fig. 13. Site 3 with sparse *Phragmites* reeds and a rich submerged and floating aquatic vegetation.



Fig. 15. Site 4, near the river mouth, with dense *Phragmites* vegetation.



Fig. 14. Pondweed (*Potamogeton spp.*), water cress-like plants and duckweed (*Lemna spp.*) at site 3.



Fig. 16. Site 7, solely with *Potamogeton spp.* aquatic plants.

The habitats of lake Kaiafa sites (8-10) were of the second type, namely canals and swamp areas, in some cases associated with springs, with a variety of riparian vegetation (Fig.17-20). Site 8 had slightly brackish water (salinity 2,6‰).



Fig. 17. Site 8, canal at lake Kaiafa near sulphur baths with dense *Phragmites* reeds but no aquatic macrophytes.



Fig.19. A close-up of site 9 with muskgrass (*Chara spp.*) beds (bottom right).



Fig.18. Site 9, marshy area with *Typha spp.*, *Scirpus spp.*, *Phragmites australis*, *Juncus spp.* and *Chara spp.* beds.



Fig. 20. Site 10, canal with springs, with *Phragmites* reeds and patches of *Typha spp.*

site	riparian	% r.v.	aquatic	% a.v.	surface
	vegetation		vegetation		cover
1	Typha spp. Salix spp. Lysimachia or Lythrum spp.	1	Potamogeton spp.	8	0
2	Typha spp.		None	0	0
3	Arundo spp.	40+	Water cress-like	2	55
	Phragmites aus.	40+	Lemna spp.	35	
			Potamogeton spp.	20	
4	Arundo spp.	30	Lemna spp.	50	65
	Typha spp.	5	Water cress-like	20	
5	Arundo spp.	5	Potamogeton spp.	1	5
	Salix spp.	85			
6	Arundo spp.	60	None	0	0
	Salix spp.	40			
	Phragmites aus.	10			
	Rubus spp.	5			
7	Eucalyptus spp.	5	Ceratophyllum spp.	2	20
	Arundo spp.	40	Potamogeton spp.	15	

TABLE 2. Data on the riparian and aquatic vegetation at the Alfios sampling sites.

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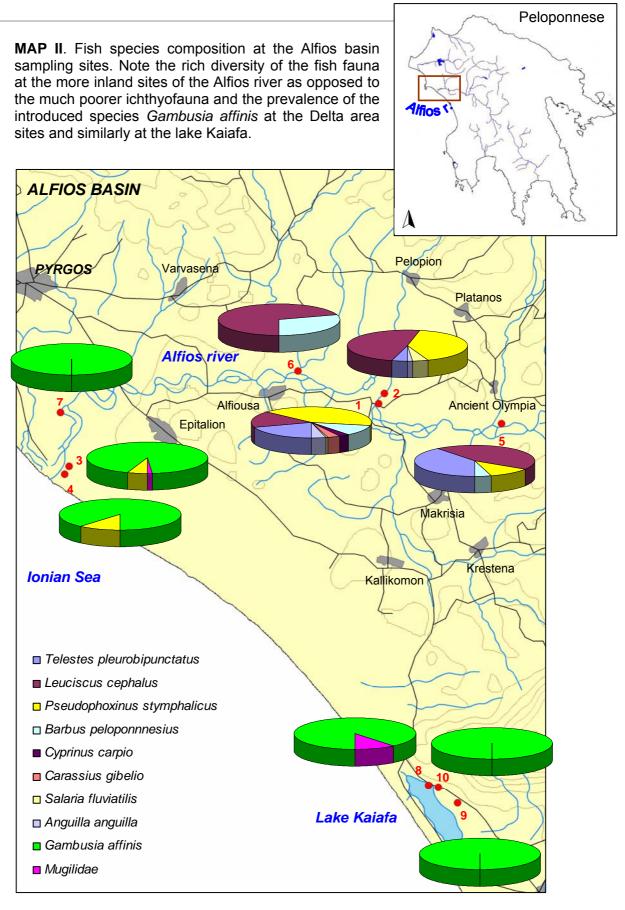
	Phragmites aus.	20			
	Typha spp.	25			
	Salix spp.	<1			
8	Eucalyptus spp.	20	Submerged plants	10	0
	Juncus spp.	20			
	Phragmites aus.	30			
	Grasses	20			
	Perennial forbs	10			
9	Typha spp.	20	Chara spp.	90	0
	Phragmites aus.	10			
	Scirpus spp.	5			
	Juncus spp.	1			
	Ficus spp.	1			
	Rubus spp.	1			
10	Typha spp.	10	None	0	0
	Phragmites aus.	80			

FISH SPECIES COMPOSITION

Table 3 and Map II (following page) summarize the fish species composition of the Alfios basin sampling stations. The fish species encountered were *Leuciscus cephalus peloponnensis, Telestes pleurobipunctatus, Barbus peloponnesius, Cyprinus carpio, Carassius gibelio* and *Pseudophoxinus stymphalicus* (Cyprinidae), *Salaria fluviatilis* (Blennidae), *Gambusia affinis* (Poecillidae), *Anguilla anguilla* (Anguillidae) and unidentified Mugilidae. Note the rich diversity of the fish fauna (e.g. 8 fish species at site 1) at the more inland sites of the Alfios river (sites 1, 2 and 5) as opposed to the much poorer ichthyofauna and the prevalence of the introduced species *Gambusia affinis* at the sites closer to the Alfios river mouth (sites 3, 4 and 7) and similarly at the Lake Kaiafa sampling sites (8, 9 and 10). In none however of the sampling sites was the occurrence of *V. letourneuxi* recorded.

TABLE 3. Fish species composition at the Alfios sampling sites (Red numbers and
dots indicate the species' absence, blue the species' presence).

site	Sampling	Fish species	%	No	V.
	equipment	-			letourneuxi
		Telestes pleurobipunctatus	19,7	208	
		Leuciscus cephalus	16,8		
		Barbus peloponnesius	41,8		
1	electrofishing	Carassius gibelio	11,1		
		Cyprinus carpio	2,9		•
		Pseudophoxinus stymphalicus	3,4		
		Salaria fluviatilis	0,5		
		Anguilla anguilla	3,8		
		Leuciscus cephalus	49,3	300	
		Telestes pleurobipunctatus	4,1		
2	electrofishing	Pseudophoxinus stymphalicus	41,1		•
		Salaria fluviatilis	4,1		
		Anguilla anguilla	1,4		
		Gambusia affinis	93,2	74	
3	electrofishing	Pseudophoxinus stymphalicus	5,4		•
	scoop net Mugilidae		1,4 88,9		
4	electrofishing	•		9	•
	scoop net Pseudophoxinus stymphalicus		11,1		
		Leuciscus cephalus	44,7	228	
_		Telestes pleurobipunctatus	39,5		
		Pseudophoxinus stymphalicus	11,4		•
		Barbus peloponnesius	4,4		
6 electrofishing <i>Leuciscus cephalus</i>			70,0	100	•
	Barbus peloponnesius		30,0		
7	electrofishing	Gambusia affinis	100,0	55	•
8	scoop net	Gambusia affinis	89,3	112	•
	dip net	Mugilidae	10,7		
9	scoop net	Gambusia affinis	100,0	51	•
10	scoop net	Gambusia affinis	100,0	35	•



CURRENT VERSUS HISTORICAL PRESENCE OF *V. LETOURNEUXI* IN THE ALFIOS BASIN

V. letourneuxi was first reported in Alfios by *Bianco & Miller* (1989), collected at a site near Epitalio village in 1987. It was found also once in 1992 - *Barbieri* et al. (2000) - at a deep creek discharging about 150 m upstream of the river mouth, which probably corresponds to site 3 or 4 of the current survey. It was also found at a similar date (*Economou* et al. 1999) near the Alfios dam, at site 1 of the current survey. Efforts to locate the species at a later date by the same group at site 1 were unsuccessful, probably due to habitat destruction through the operation of the dam and gravel extraction works at the area. Despite sustained localised effort, it was not possible, during the current survey, to locate the species in any of those sites (see Table below, red dots indicate the species' absence, blue dots the species' presence).

References	V. letourneuxi presence
Bianco & Miller (1989)	•
Barbieri et al. (2000)	•
Economou et al. (1999)	•
Current Survey	•

3.1.2 PINIOS RIVER

The Pinios river (approx. length 82 km), with a basin of 878 km² and a Delta area of 83 km², is fed by a karstic system of mount Erymanthos. It is a perennial water system, but with very low summer water discharge (average summer discharge $0.4m^3$ /s). The section of the river, downstream of the Pinios dam, shows signs of pollution and eutrophication due to urban waste disposal and agricultural and animal husbandry activities.

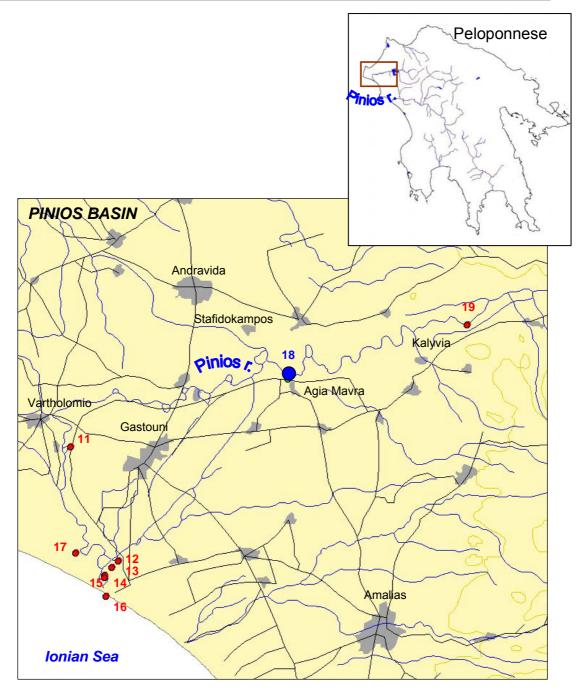
SAMPLING STATION DISTRIBUTION

The current survey focused on the lowland areas of the Pinios basin, with the majority of the sampling sites concentrated in the Delta area (sites 11,12,13,14 and 17 at different canals feeding the river and sites 15 and 16 at the main river close to its mouth), with two additional sites at the main river, further inland (sites 18, 19). (Table 1 provides a short description and pressure list of these sites and Map I a spatial distribution of the sampling sites).

TABLE 1. Sampling sites at the Pinios basin.

site	location	water body type	pressures
11	Agios Athanasios canal	Ditch/pool	
12	Agios Athanasios canal further downstream, before confluence with Margarita canal	Confluence of drainage canal with Margarita canal	Artificial drainage system, water abstraction for agricultutre
13	Margarita canal-wooden bridge	Canal	Vegetation clearing, potential pollution from agricultural run-off
14	Margarita canal- downstream from bridge	Canal	Riparian vegetation clearing, potential pollution from agricultural run-off
15	Pinios river –confluence with Margarita canal	River	Water abstraction and agricultural pollution causing low water flow and high turbidity respectively
16	Pinios river mouth	River	Structural changes, reinforced banks, etc.
17	Mantemi canal	Canal/pool	Citrus orchards and riparian vegetation clearing
18	Pinios river-Agia Mavra bailey bridge	River	Urban waste disposal
19	Pinios river –Kalyvia- Avgion	River	Evident river bed quarrying

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MAP I. Sampling sites at the Pinios basin. Localities where *V. letourneuxi* was recorded (\bullet) – site 18 - and not recorded (\bullet) – sites 11-17 & 19.

PRESSURES

A major threat to the Pinios system is water abstraction and water regulation from the dam (approx. 40 km from the river mouth) which at certain times of the year decrease significantly the water discharge. The area is cultivated extensively and thus the levels of agricultural pollution are high, as well as the point-source pollution from urban waste disposal (as in the case of Agia Mavra) and some animal husbandry waste. The effects of all of the latter are further accentuated by water abstraction and dam water regulation which almost zeroes summer water discharge. It has been calculated that the minimum discharge necessary is 5-7 m³/s (Economou *et al.*, 1999), much higher than the current average summer discharge. Finally there is evidence of river bed quarrying at the samplingsites further inland.

HABITAT CHARACTERISTICS

In terms of habitat characteristics the Pinios sampling sites included canals feeding the main river (sites 11, 12, 13, 14 and 17) as well as more open riverine areas (sites 15, 16 at the delta area and sites 18 and 19 further inland). Sites 11, 12, 13, 14 and 17 were relatively narrow canals with very low flow, rich riparian vegetation (mostly *Arundo spp.* and *Phragmites australis*) and some aquatic vegetation (mostly *Potamogeton spp.* and in one case *Lemna spp.*) (Figures 1-6).



Fig. 1. Site 11, Agios Athanasios canal with *Sparganium spp.* aquatic plants.



Fig. 2. Site 12 with rich riparian vegetation and *Potamogeton spp.* aquatic plants (bottom right).



Fig. 3. Close up of site 12, with *Phragmites* reeds and *Potamogeton spp.* aquatic plants (top right).



Fig. 4. Site 13, Margarita canal. *Arundo* and *Phragmites* reeds (right bank) and annual grasses after vegetation clearing (left bank).

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Fig. 5. Site 17, Mantemi canal.



Fig. 6. Site 17 with *Salix spp.* and *Arundo* reeds.

In contrast to the above, sites 15 and 16 were stretches of the main Pinios river, close to its mouth, both wide and deep. They were also characterized by rich riparian vegetation but had no aquatic vegetation (Fig. 7-9).



Fig. 7. Site 15, Pinios river – confluence with Margarita canal. Note the absence of any aquatic vegetation.



Fig. 8. Site 16, River mouth. Note the prevalence of *Phragmites* reeds on the distal river bank.



Fig. 9. Close up of site 16. Note the *Phragmites* reeds on the distal bank and the *Sparganium spp.* on the proximal bank.

Sites 18 and 19, at the main river further inland, were narrower and shallower sites (30 and 20 cm deep respectively) with thickly vegetated banks and some aquatic vegetation (Fig. 10-13).



Fig. 10. Site 18, Agia Mavra Bailey bridge. *Arundo* reeds, *Typha spp.* and *Iris spp.*



Fig. 12. Site 19, Pinios river, Kalyvia-Avgion, with *Arundo* and *Phragmites* reeds.



Fig. 11. Close-up of site 18 with *Potamogeton spp.* (bottom left).



Fig. 13. Site 19, with *Phragmites* reeds and stagnant waters with some decomposing floating algae. Evidence of gravel quarrying.

site	riparian vegetation	% r.v.	aquatic vegetation	% a.v.	surface cover
11	Ficus spp.	10	Sparganium spp.	60	60
	Populus spp.	60			
	Arundo spp.	30			
12	Salix spp.	10	Potamogeton spp.	50	50
	Arundo spp.	10	0 11		
	Phragmites aus.	40			
	Rubus spp.	30			
	Vitex agnus castus	20			
13	Arundo spp.	15	Potamogeton spp.	2	15
	Phragmites aus.	35	0 11		
	Annual grasses	50			
14	Arundo spp.	40	None	0	0
	Phragmites aus.	60			
15	Eucalyptus spp.	50	None	0	0
	Arundo spp.	50			
	Phragmites aus.	50			
	Populus alba	20			
16	Arundo spp.	5	None	0	0
	Phragmites aus.	70			
	Salix spp.	5			
	Sparganium spp.	5			
17	Salix spp.	20	Annual & perennial	10	40
	Fraxinus angustifolia	<1	emergent grasses		
	Arundo spp.	15	Lemna spp.	50	
	Iris spp.	20	Unknown submerged	30	
			plants		
18	Arundo spp.	40	Potamogeton spp.	10	15
	Typha spp.	50	Ceratophyllum spp.	5	
	Phragmites aus.	15			
	Iris spp.	2			
19	Arundo spp.	45	Floating algae	15	15
	Phragmites aus.	45			
	Juncus spp.	5			

TABLE 2. Data on the riparian and aquatic vegetation at the Pinios sampling sites.

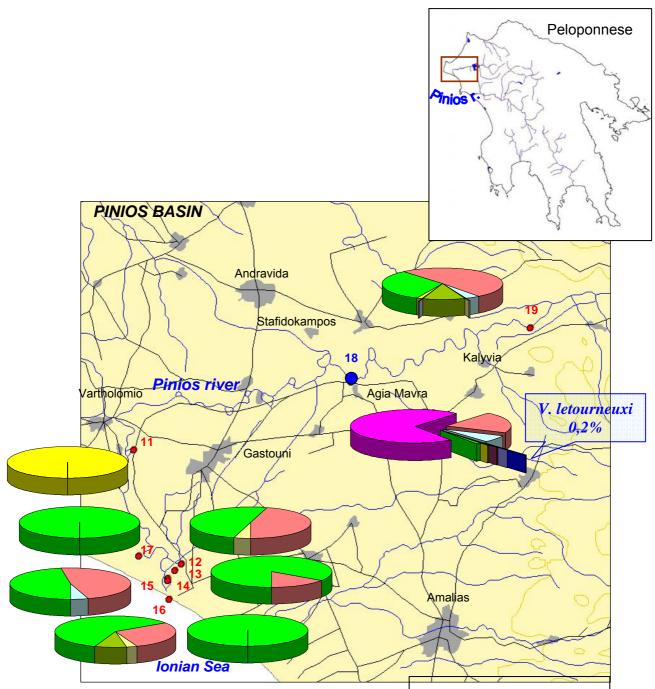
FISH SPECIES COMPOSITION

Table 3 summarizes the fish species composition at the Pinios basin sampling sites and Map II their spatial distribution at the Pinios basin. The fish species encountered peloponnensis. cephalus Telestes were Leuciscus pleurobipunctatus, Pseudophoxinus stymphalicus, Tropidophoxinellus hellenicus and Barbus albanicus (Cyprinidae), Knipowitschia sp. (Gobiidae), Salaria fluviatilis (Blennidae), Gambusia affinis (Poecillidae), Anguilla anguilla (Anguillidae), species of the Mugilidae family and Valencia letourneuxi at one site (site 18). Note the rich ichthyofauna diversity at the sites further inland (18 and 19), as opposed to the much poorer ichthyofauna at the delta area sites and the prevalence there of the introduced species G. affinis. Note also the presence of *Tropidophoxinellus hellenicus* in 6 Pinios sampling sites (sites 12, 13, 15, 16, 18 and 19) and Barbus albanicus in three sites (sites 15, 18 and 19). The Pinios river is the southernmost system of the geographical distribution of these two species and the only system where they occur in the Peloponnese. Finally, note the presence of Mugilidae species relatively further upstream (site 18).

V. letourneuxi was found in only one site (site 18) but at extremely low densities (2 individuals out of a total of 891).

site	Sampling	es composition at the Pinios samp <i>Fish species</i>	%	No	<i>V.</i>
	equipment	-			letourneuxi
11	scoop net	Pseudophoxinus stymphalicus	100,0	37	•
	electrofishing	Tropidophoxinellus hellenicus	45,5	22	
12	scoop net	Gambusia affinis	50,0		•
	dip net	Salaria fluviatilis	4,5		
13	scoop net	Tropidophoxinellus hellenicus	15,6	32	•
		Gambusia affinis	84,4		
14	electrofishing	Gambusia affinis	100,0	6	•
	scoop net				
		Tropidophoxinellus hellenicus	47,6	21	
15	electrofishing	Gambusia affinis	47,6		•
	scoop net	Barbus albanicus	4,8		
		Tropidophoxinellus hellenicus	28,6	35	
		Gambusia affinis	60,0		•
16	scoop net	Knipowitschia sp.	8,6		
		Salaria fluviatilis	2,8		
17	scoop net	Gambusia affinis	100,0	9	•
		Tropidophoxinellus hellenicus	20,6	891	
		Gambusia affinis	1,2		
		Barbus albanicus	7,0		
18	electrofishing	Telestes pleurobipunctatus	0,1		
		Leuciscus cephalus pelop.	0,1		•
		Pseudophoxinus styphalicus	0,2		
		Mugilidae	70,6		
		Valencia letourneuxi	0,2		
		Tropidophoxinellus hellenicus	50,8	382	
		Gambusia affinis	31,9		
		Barbus albanicus	3,9		
19	electrofishing	Pseudophoxinus styphalicus	1,3		•
	scoop net	Knipowitschia sp.	11,5		
		Salaria fluviatilis	0,3		
		Anguilla anguilla	0,3		

TABLE 3. Fish species composition at the Pinios sampling sites.



MAP II. Fish species composition at the Pinios basin sampling sites. Note the rich ichthyofauna diversity at the more inland sites as opposed to the much poorer ichthyofauna at the delta area sites and the prevalence there of the introduced species Gambusia Note also the affinis. presence of Tropidophoxinellus hellenicus and Barbus albanicus. This is the southernmost system of their geographical distribution and the only system where they occur in the Peloponnese. V. letourneuxi was found in only one Pinios site (site 18) but almost below detection in terms of densities (2 specimens in 891).

- Pseudophoxinus stymphalicus
- Gambusia affinis
- Tropidophoxinellus hellenicus
- Barbus albanicus
- 🗖 Salaria fluviatilis
- Knipowitschia spp.
- 🗆 Anguilla anguilla
- Telestes pleurobipunctatus
- Leuciscus cephalus
- Mugilidae
- Valencia letourneuxi



Fig. 14.

Pinios river, Agia Mavra (site 18). Specimens of some of the sympatric species of *V.letourneuxi*. From top to bottom (Mugilidae, *Tropidophoxinellus helenicus, Telestes pleurobipunctatus*).



Fig. 15.

Juvenile *V. letourneuxi*, approx. length 1 cm, caught close to the river bank at site 18, in association with *Gambusia affinis*.



Fig. 16. Dorsal view of juvenile *V.letourneuxi*, with the characteristic reflective spots over the eyes.

CURRENT VERSUS HISTORICAL PRESENCE OF *V. LETOURNEUXI* IN THE PINIOS BASIN

V. letourneuxi was first found by *Bianco & Miller* (1989) at a site under the bridge of Agia Mavra in 1987, which corresponds to site 18 of the current survey, where *V. letourneuxi* presence was also confirmed in October 2005 during the current survey but at extremely low densities. It had also been found by *Barbieri* et al. (2000) at a deep site of the river near Kalyvia village, d/s from Vartholomio bridge, where, however, *V. letourneuxi* presence could not be confirmed in the current survey (see Table below).

References	V. letourneuxi presence
Bianco & Miller (1989)	•
Economou <i>et al.</i> (1999)	•
Barbieri <i>et al.</i> (2000)	•
Current Survey	•

3.1.3 PROKOPOS LAGOON

Prokopos lagoon is part of a wider system of lagoons and wetlands at Strofilia in northwestern Peloponnese. These wetlands are watered by torrent streams lacking an outlet to the sea by an intermediate zone of sand dunes. Prokopos lagoon, 2 km south of Araxos village, covers an area of approx. 4-5 km² and its depth varies from 4,5 to 5,5 m. The major anthropogenic pressures in the area are surface water abstraction, agricultural pollution, limited touristic development, etc (Fig.1). During the current survey only one site was sampled at Prokopos lagoon, (site 20), a spring area connected with the lagoon, with brackish waters (salinity 7,3 ‰) and relatively high water temperature, 19,4°C. The surrounding area is covered with rush and *Phragmites* reeds. In the area of the springs there were some submerged algae (Fig.2).

site	location	water body type	pressures
20	Agios Georgios spring, Prokopos lagoon	Spring associated with lagoon	Water abstraction, tourist
	_	-	infrastructure.



Fig. 1. The Prokopos lagoon.



Fig. 2. Prokopos lagoon and the Strofilia pine forest zone along the Ionian coast.





Fig. 3, 4. Site 20, Agios Georgios spring, connected to the lagoon. Note the presence of submerged algae (bottom right) and the *Phragmites* reeds in the surrounding area.

site	riparian vegetation	% r.v.	aquatic vegetation	% a.v.	surface cover
20	Phragmites aus.	1	Algae	8	0

TABLE 2. Data on the riparian and aquatic vegetation at the Prokopos sampling site.

FISH SPECIES COMPOSITION

Table 3 summarizes the fish species composition at the Prokopos lagoon sampling site. The fish species encountered were the introduced species *Gambusia affinis* (Poecillidae), fish of the Mugilidae family, as well as *Pseudophoxinus stymphalicus* (Cyprinidae) in very small numbers.

TABLE 3. Fish species composition at the Prokopos lagoon sampling site.

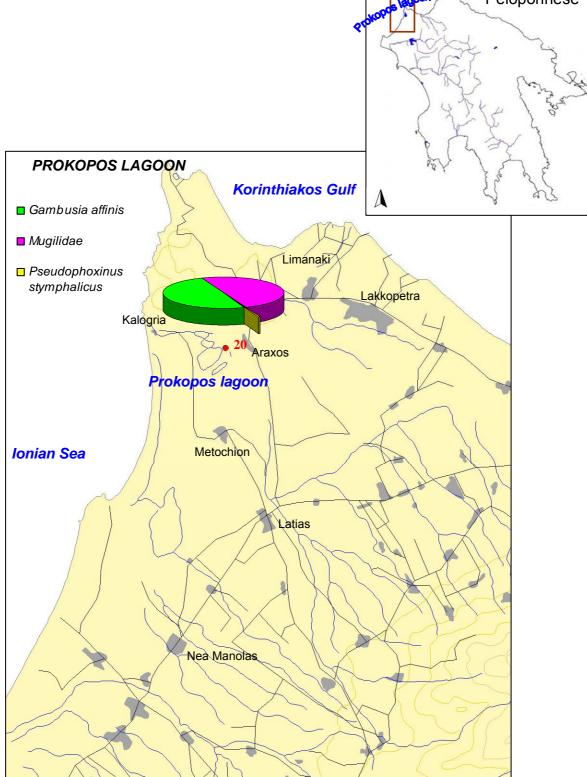
site	Sampling equipment	Fish species	%	No	V. Ietourneuxi
20	scoop net dip net	Gambusia affinis	49,8	201	
20	uphet	Pseudophoxinus stymphalicus Mugilidae	0,5 49,7		•

Map I (following page) shows the location of the Prokopos lagoon sampling site and the fish species composition in that site.

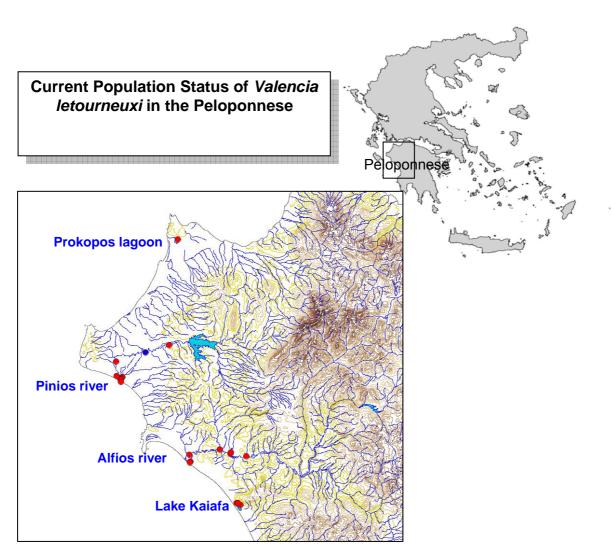
CURRENT VERSUS HISTORICAL PRESENCE OF *V. LETOURNEUXI* IN PROKOPOS LAGOON

There is no previous report of *V. letourneuxi* presence in this wetland system and the current survey confirmed its absence at the site investigated.

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MAP I. The fish species encountered at this site were the introduced species Gambusia affinis (Poecillidae), fish of the Mugilidae family, as well as Pseudophoxinus stymphalicus (Cyprinidae) in very small numbers. No V. letourneuxi was fished in that site.



In the frame of the current survey, three water systems of the Western Peloponnese were sampled for *V. letourneuxi* presence (from South to North, the Alfios river - with Lake Kaiafa, the Pinios river and the Prokopos lagoon). Out of a total of 20 sampling stations, *V. letourneuxi* presence was confirmed in only one sampling site (•) in the Pinios river and that at extremely low densities (2 individuals out of a total of 891). That, in combination with the absence of the species in the Alfios river, where it has been historically reported, possibly reflects its near extinction in the water systems of the Peloponnese.

Water Systems of Western Central Greece

- 3.1.4 Mornos river
- 3.1.5 Acheloos river (and Aitoliko streams)
- 3.1.6 Agios Dimitrios river (and Astakos springs)
- 3.1.7 Vlychos springs Western Central Greece

3.1.4 MORNOS RIVER

The Mornos river flows from mount Oiti and into the Korinthiakos Gulf, near the city of Nafpaktos (Lepanto) forming a delta area of 29 Km². The length of the river is 60 km and its average yearly discharge 484 hm³, of which almost half is diverted to Athens and the rest is used for irrigation of fields in the delta. At the delta area there are many springs, which are part of the same carstic system that discharges in the Mornos basin. There are also sedimentary aquifers that filter water from upstream parts of the river.

SAMPLING STATION DISTRIBUTION

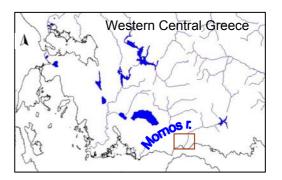
The current survey focused at four streams of the Mornos delta area, i.e. the Gouvos stream (sampling sites 21-23), the Koufosouda stream (sites 37 and 24-28) (which was connected in the past with the Gouvos stream), the Giara stream (site 29) and the Managouli stream (sites 31-34), fed by karstic springs. In addition, two sites very close to the Mornos river (sites 30 and 35) were sampled (Table 1 provides a short description and pressure list of these sites and Map I a spatial distribution of the sampling sites).

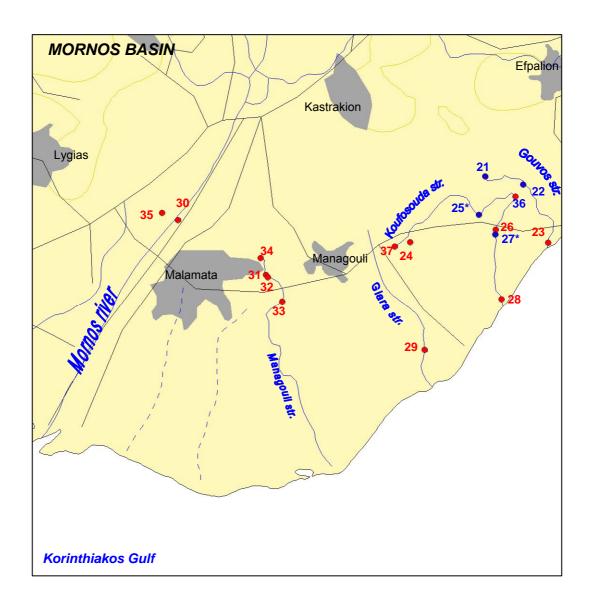
site	Subsystem	location	water body type	pressures
21		Chiliadou Springs	Spring fed stream	Pumping station, dredging, alfalfa fields
22	Gouvos str.	Ananti limnis	Stream	Pasture, olive groves, vegetation clearing
23		Gouvos outlet	Stream outlet to the sea	Tourist infrastructure
37		Koufosouda springs	springs	Agriculture
24		Koufosouda spring area	Spring fed stream	Agriculture
36	Koufosouda str.	Irrigation channel	Irrigation channel	Vegetation clearing
25		Bridge Elafia	Cement canal	Live-stock farm, bridge, channeling
26		Apothiki sikias	stream	Agriculture, buildings
27		Apothiki Elafia	stream	Agriculture
28		Koufosouda outlet	Stream outlet to the sea	Tourist infrastructure
29	Giara str.	Giara str.	Stream	Alfalfa, Maize, pasture
31	Managouli str.	Road bridge	Stream, with pools	Houses, some agriculture, vegetation clearing, dredging
32	1	Sluice	Stream	Alfalfa fields
33		Iron bridge, aquaduct	Stream	Agriculture, channeling, water abstraction

TABLE 1. Sampling sites at the Mornos basin.

34		Springs	Springs	Goat yard, scrap yard, dirt road.
35	Mornos embankment ditch	Mornos embankment ditch	Ditch parallel to main river course	Pasture, gravel road
30	Mornos river	Mornos river	Springs in Mornos river bed	Water abstraction, embankments, rubbish disposal.

MAP I. Sampling sites at the Mornos basin. Localities where *V. letourneuxi* was recorded (\bullet) – sites 21,22 at Gouvos str. and 25^{*}, 27^{*} at Koufosouda str. - and not recorded (\bullet) – site 23 at Gouvos str., sites 36, 26, 28 at Koufosouda str., site 29 at Giara str., sites 34, 31, 32, 33 at Managouli str. and sites 30, 35 at Mornos river. * First record





PRESSURES

A major threat for the system is water abstraction from the Mornos dam (supplying with water the city of Athens) and, as a result, the part of the river downstream from the dam dries up almost completely during the summer months. There is also water abstraction through drilling. Additional threats are river sand/gravel removal and agricultural pollution, especially at the delta area, due to the excessive use of fertilizers. Finally, the planned redistribution of land in the area is bound to affect land- use and consequently the water resources use in the delta. Specific threats of the areas V. letourneuxi stronghold (Chiliadou springs, see below) include possible increased water abstraction from the springs, already supplying the nearby community of Chiliadou (thus, limiting natural surface water flow).

HABITAT CHARACTERISTICS

During the current survey, four streams of the delta area were sampled.

Gouvos stream

The Gouvos stream is formed by the Chiliadou springs, possibly fed by an aquifer independent from Mornos, which renders the system relatively stable. At the site there is a pumping station supplying with water the small village of Chiliadou and the rest of the water flows between fields (alfalfa) and grassy areas, forming the Gouvos stream (Fig. 1 - 3). Sampling site 21 (a few meters from the springs) is characterized by rich riparian and aquatic vegetation and clear waters (Fig.4-6). Site 22 (Ananti limnis), further downstream was a ditch with dense *Phragmites* reeds and thick algae mats (Fig.7-9). The Gouvos outlet at the sea (site 23) was characterized by very cold water, very dense Phragmites reeds (4 m wide on each side) and very little aquatic vegetation (water cress-like plants, Fig.10-12).





Fig. 1. Chiliadou springs (site 21). On the left- Fig. 2. Alfalfa fields and grassy area at the site hand side is visible the white building of the of the springs. pumping station.



Fig.3. The location where water flows out from an underground water pipe at Chiliadou springs.



Fig. 5. Typha spp. at site 21.



Fig. 7. Site 22, Ananti Limnis. Dense *Phragmites* reeds with *Cirsium spp.* (right) and climbing *Calistygia spp.*



Fig.4. The Chiliadou springs sampling site with rich aquatic vegetation (and some open spots) and fringing reeds.



Fig. 6. Water cress-like plants and floating algae at site 21.



Fig. 8. Site 22, with Rubus spp. (left).



Fig. 9. Close up of site 22.



Fig. 10. Site 23, the outlet of Gouvos stream with dense *Phragmites* reeds and very little aquatic vegetation.



Fig. 11. Underwater photo of Gouvos outlet.



Fig. 12. Underwater photo of Gouvos outlet with *Pseudophoxinus stymphalicus*.

Koufosouda stream

The Koufosouda stream is fed by springs, located more easterly to Chiladou springs. Two sites were sampled at the area of the springs (sites 37 and 24) with thick riparian vegetation (Fig.13 and 14). Another four sites further downstream were sampled, which included a cement canal (site 25), an irrigation canal (site 36, Fig.15,16) and two more narrow sites with slow flowing water and thick vegetation (sites 26, 27, Fig. 17). The Koufosouda outlet site (site 28) is rather degraded with stagnant water and no aquatic vegetation (Fig. 18 & 19).



Fig. 13. The Koufosouda springs area (site 24)



Fig. 14. Koufosouda springs with *Sparganium spp*.



Fig. 15. Site 36, Irrigation canal.



Fig. 18. Koufosouda outlet (site 28) with stagnant water.



Fig. 16. Site 36 with flowering *Rubus spp.* (right)



Fig. 17. Site 26, Apothiki sikias, with fern (right) and *Cornus sanguinea* (further right).



Fig. 19. Koufosouda outlet.

Giara stream

Giara stream was also sampled at a site (Marathiades, site 29) with fast flowing water, large cobbled substrate and no aquatic vegetation (Fig. 20, 21).



Fig. 20. Site 29. Giara str., with Ficus tree and Phragmites reeds.



Fig. 21. Site 29, Giara str. with fast flowing water.

Managouli stream

4 sites at Managouli stream were sampled during the current survey, the Managouli spring area (site 34) and another three sites further downstream (31, 32 and 33). The Managouli springs are located in a big Platanus grove and characterized by slow flowing, clear waters with little aquatic vegetation (Fig. 21,22). Site 31 is a wider strech of the stream, heavily disturbed but with rich aquatic vegetation (80% surface cover, Fig. 23, 24). Further downstream site 32 (sluice), by a sluice, is a narrower stream section with fast flowing water, rich riparian vegetation but very little aquatic vegetation (Fig. 25-27). Finally, site 33 (iron bridge) is characterized by fast flowing water and rocky substrate The riparian vegetation consists mainly of *Phragmites* reeds and the aquatic vegetation solely of Ranunculus spp. (Fig.28-32).



Fig. 21. Site 34 (Managouli springs) with Fig. 22. Managouli springs. (Platanus, left), Juglans spp. and Rubus spp.





Fig. 23. Site 31. Managouli stream, road bridge, with rich riparian and aquatic vegetation.



Fig. 24. Site 31. Managouli stream, road bridge.





Fig. 25. Site 32, Managouli sluice.



Fig. 27. Site 32, with nettle, *Calistygia spp.* and various hydrophyllus plants.



Fig. 26. Site 32, with dense riparian but very little aquatic vegetation.



Fig. 28. Site 33 with *Phragmites* reeds and *Ranunculus* aquatic plants (bright green).



Fig. 29. Site 33 with Platanus trees, Phragmites reeds and *Ranunculus spp.* aquatic plants.





Fig. 30. Site 33 with fast flowing water.



Fig. 31. Site 33.

Mornos river

Two sites at and near the Mornos river bed were sampled during this survey (sites 30, 35). The first site (site 30) were some springs at a wide section of the Mornos river bed with no aquatic vegetation and the second site (site 35) at an adjacent location, a ditch parallel to the river with cool water, big *Eucalyptus* and *Ficus* trees at its banks and bright green water cress-like aquatic plants (Fig. 33,34).



Fig. 33. Site 35, Mornos embankment ditch, with bright green water cress-like plants.



Fig. 34. Mornos embankment ditch.

site	riparian	% r.v.	aquatic	% a.v.	surface
	vegetation		vegetation		cover
21	Typha spp.	55	Lycopsis spp.	10	40
	Grasses	20	Typha spp.	10	
	Calistygia spp.	5	Grasses	10	
	Rubus spp.	10	Water cress-like	30	
	Juncus spp.	1	Floating algae	15	
	Purple geranium-like	2,5	Polygonum spp.	10	
22	Phragmites aus.	100	Floating algae	80	80
	Rubus spp.	20			
	Cirsium spp.	1			
23	Phragmites aus	80	Water cress-like	5	15
	Calistygia spp	5			
37	Phragmites aus.	1	Water cress-like	100	100
	Grasses	20			
24	Water cress	40	Sparganium spp.	20	80
	Grasses	30	Water cress	2	
	Ficus carica	10			
	Periploca graeca	10			
36	Phragmites aus	100	None	0	30
•••	Rubus spp.	15		·	
25	Nerium oleander	20	Water plantain	10	0
			(Alisma spp)		-
			Water cress	10	
			Submerged algae	60	
26	Phragmites aus.	5	Apion spp.	80	75
20	Ficus carica	100	Sparganium spp.	10	10
	Humulus lupus	2	opargamam opp.	10	
	Sparganium spp.	40			
	Grasses	30			
	Ferns	3			
	Cornus sanguinea	1			
	Plant of Apiacea	10			
	family	10			
27	Sparganium spp.	20	Sparganium spp	10	80
21		20	Sparganium spp. Water cress	20	00
	Phragmites aus. Water hemlock	20 10	Water cress Alisma spp.	20 10	
	Water Herniock	10		10	
28	Dhragmitae aug	100	Echium lycopsis Floating algae	5	5
<u>28</u> 29	Phragmites aus.		None	5 0	<u> </u>
23	Phragmites aus.	30 5	INOTIC	U	U
	Juncus spp. Figus carioa				
	Ficus carica	15			
	Urtica spp.	10			
	Plant of Apiaceae	20			
	family Domulus one	-			
	Populus spp.	5			
	Rubus spp.	5	14/- (4.5	00
31	Rubus spp.	15	Water cress-like	45	80
	Calystegia spp.	75	Lemna spp.	20	
	Arundo spp.	15	Floating algae	30	
	Cirsium spp.	2			
	Phragmites aus.	5			

TABLE 2. Data on the riparian and aquatic vegetation at the Mornos sampling sites.

	Urtica spp.	25			
		23 5			
	Ficus spp.	-			
32	Phragmites aus.	10	Lycopsis spp.	5	10
	Rubus spp.	30	Water-cardamon like	5	
	Calistygia spp.	60	Ranunculus spp.	5	
	Urtica spp.	10			
33	Salix spp.	2	Ranunculus spp.	20	20
	Platanus orientalis	5			
	Phragmites australis	70			
34	Platanus orientalis	30	Water-cardamon like	30	20
	Juglans spp.	20			
	Rubus spp.	70			
35	Ficus spp.	50	Water-cardamon like	20	0
	Eucalyptus spp.	80	(bright green)		
	Rubus spp.	20			
30	Platanus orientalis	1	None	0	0
	Nerium oleander	10			
	Vitex agnus	30			
	Arundo spp.	20			
	Eucalyptus spp.	5			
	Tamarix spp.	30			

FISH SPECIES COMPOSITION

Table 3 and Map II (following page) summarize the fish species composition of the Mornos basin sampling sites. The fish species encountered were *Pseudophoxinus stymphalicus* and *Leuciscus cephalus* (Cyprinidae), *Economidichthys pygmaeus* (Gobiidae) *Salaria fluviatilis* (Blennidae), *Anguilla anguilla* (Anguillidae) some Mugilidae fish and *Valencia letourneuxi*. *V. letourneuxi* was found in two systems (Gouvos stream and Koufosouda stream) which historically have been connected. Note especially the absence of the introduced species *Gambusia affinis* (Poecillidae), which is considered a competitor of *V. letourneuxi* for the same habitat. The relative abundance of *V. letourneuxi* in the Mornos streams (sites 21 and 25), although not high in absolute terms, is the highest in all systems investigated in the current survey.

TABLE 3. Fish species composition at the Mornos sampling sites.

site	Sampling	Fish species	%	No	<i>V.</i>
	equipment				letourneuxi
21	Scoop net	Pseudophoxinus stymphalicus	75,3	174	•
		Economidichthys pygmaeus	4,6		
		Valencia letourneuxi	20,1		
22	Scoop net	Pseudophoxinus stymphalicus	77,4	71	•
		Economidichthys pygmaeus	9,9		
		Mugilidae	1,4		
		Valencia letourneuxi	11,3		
23	Scoop net	Pseudophoxinus stymphalicus	100	20	•
37	Scoop net	No fish	0	0	•
24	Scoop net	Pseudophoxinus stymphalicus	100	5	•
36	Scoop net	Pseudophoxinus stymphalicus	100	30	•
25 *	Scoop net	Pseudophoxinus stymphalicus	60	5	
	Dip net	Economidichthys pygmaeus	20		•
		Valencia letourneuxi	20		

			400	4	
26	Scoop net	Pseudophoxinus stymphalicus	100	4	•
27*	Scoop net	Pseudophoxinus stymphalicus	57,1	7	•
		Economidichthys pygmaeus	28,6		
		Valencia letourneuxi	14,3		
28	Scoop net	Pseudophoxinus stymphalicus	30	60	•
		Mugilidae	70		
29	Electrofishin	Leuciscus cephalus	12,5	16	•
	g	Salaria fluviatilis	12,5		
	-	Anguila anguila	12,5		
		Mugilidae	62,5		
31	Scoop net	Pseudophoxinus stymphalicus	50	85	•
	Dip net	Economidichthys pygmaeus	50		
32	Scoop net	Pseudophoxinus stymphalicus	90,9	110	•
	dipnet	Economidichthys pygmaeus	9,1		
33	Scoop net	Pseudophoxinus stymphalicus	100	11	•
34	Scoop net	No fish	0	0	•
35	Sein net	Pseudophoxinus stymphalicus	100	3	•
30	Scoop net	Leuciscus cephalus	33,3	30	•
		Pseudophoxinus stymphalicus	66,7		

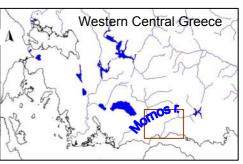
* First record

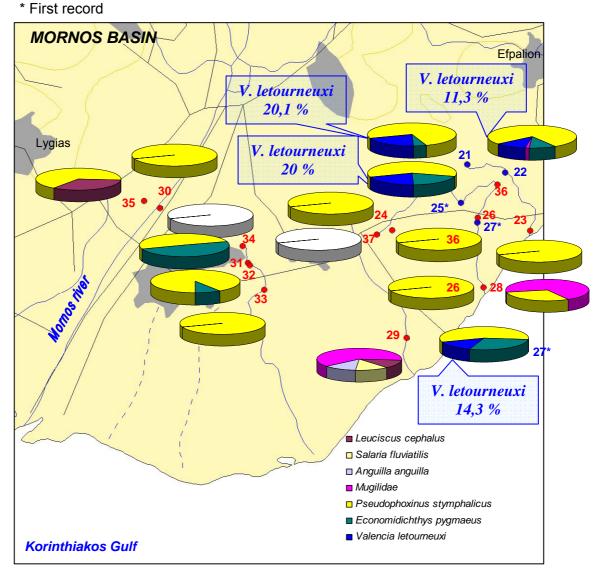


Fig.35.

A male *V. letourneuxi* from the Chiliadou springs population (bottom left), with characteristic transverse stripes and yellow coloration of the anal fin, among female *V. letourneuxi* fish.

MAP II. Fish species composition at the Mornos basin sampling sites. V. letourneuxi was found in two systems (Gouvos stream and Koufosouda stream, sites 21, 22 and 25*, 27* respectively) which historically have been connected. Note especially the absence of the introduced species G. affinis considered generally a competitor of V. letourneuxi for the same habitat. The relative abundance of V. letourneuxi in the Mornos streams is the highest in all systems investigated in the current survey.





CURRENT VERSUS HISTORICAL PRESENCE OF *V. LETOURNEUXI* IN THE MORNOS BASIN

V. letourneuxi was first reported in Mornos by *Barbieri* et al. (2000) fished at the deep ditch discharging the Chiliadou springs, probably corresponding to site 21 or 22 (Gouvos stream) of the current survey and also previously reported in the *Economou* et al. (1999) technical report at the same site. Its presence in this system has been confirmed during the current survey. The species was also found in a nearby system, the Koufosouda stream historically connected with the Gouvos stream but now separate, at two sites (see Table below).

References	V. letourneuxi presence
Barbieri <i>et al.</i> (2000)	•
Economou et al. (1999)	•
Current Survey	•

POPULATIONS FIRST RECORDED HERE

Koufosouda stream

3.1.5 ACHELOOS RIVER

The Acheloos river is the largest in discharge and one of the two longest Greek rivers (220 m, with the largest basin entirely in Greek territory) flowing from the mountain range of Pindos and discharging into the Ionian Sea. It forms a Delta of 66.5 km² which includes coastal and inland wetlands of brackish and fresh water. Given the high rainfall rates at this area of Greece (average yearly rainfall 1350-1400 mm), surface discharge is significant. In the wider Acheloos river basin there are four natural lakes as well as a number of reservoirs for hydroelectric power production.

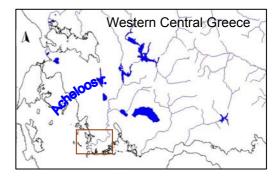
SAMPLING STATION DISTRIBUTION

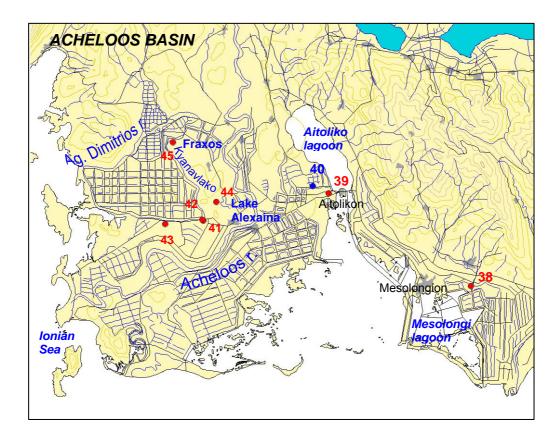
In the course of the current survey, a total of 8 sites were sampled at the lower Acheloos river Delta area for *V. letourneuxi* occurrence. These included two small systems in the Aitoliko-Mesolongi area, i.e. one canal, the Canal from Ai Simios (site 38) flowing into the Mesolongi lagoon and two small streams (sites 39 and 40) flowing into the Aitoliko lagoon. The remaining sampling sites were some water systems in connection with the Kyanavlako canal, a central canal, which serves as a major drainage canal of an extensive cultivated area at the site of the drained lake Meliti. These were some small springs flowing into Kyanavlako canal, such as site 41 (Rocky spring), 42 (Kaskariki) and 43 (by the ancient port of Oiniades). There were also two independent water systems, a lake (site 44 -Lake Alexaina) and the springs of the Fraxos water forest (site 45). Table 1 provides a short description of these sites with the relevant pressure list and Map I shows the sampling sites at the Acheloos river area.

site	location	water body type	pressures
38	Canal from Ai Simios	Canal	Riparian vegetation clearing, canal bed
			clearing
39	Stream of Aitoliko 1	Stream mouth	Adjacent road
40	Stream of Aitoliko 2	Spring fed stream	Water abstraction, agriculture
41	Rocky spring flowing into Kyanavlako canal	springs	Water abstraction, wetland drainage through Kyanavlako canal, grazing
42	Kaskariki, springs forming a small marsh	Marshy springs	Water abstraction, wetland drainage through Kyanavlako canal, grazing
43	Oiniades springs at the ancient site of Oiniades port	Springs, ephemeral pools	Wetland drainage through Kyanavlako canal
44	Lake Alexaina	Lake	Road building, grazing, potential organic pollution
45	Fraxos springs in the Fraxos water forest	Springs forming ponds in the forest	Wetland drainage

TABLE 1. Sampling sites at the Acheloos river area.

MAP I. Sampling sites at the Acheloos river basin. Localities where *Valencia letourneuxi* was recorded (\bullet) – site 40 – and not recorded (\bullet) – sites 38, 39 and 41-45.





PRESSURES

The waters of Acheloos river are mainly used for hydroelectric power production and for field irrigation. The main sources of pollution are urban waste and agricultural pollution. Due however to the high water discharge of the river, the effect of this pollution is low. In contrast, the Fraxos site, a unique water forest area, faces very particular problems, namely continuously decreased surface water levels due to drainage of underground waters through the Kynavlako canal, as well as inadequate water management measures (i.e. installation of a sprinkler system with polluted water from irrigation canals).

HABITAT CHARACTERISTICS

In terms of habitat characteristics the Acheloos sampling sites included canals (site 38), spring areas and small spring-fed streams (sites 39, 40, 41, 42, 43), a lake (site 44) and underground springs in a water forest (site 45).

Ai Simios canal

The canal from Ai Simios, near Mesolongi, site 38, is a canal with silty bottom and slow flowing water, with signs of recent riparian vegetation clearing as well as canal bed clearing. At its banks there were *Calistygia spp., Juncus spp.,* Typha *spp.* and *Phragmites* reeds. Its aquatic vegetation consisted of *Lemna spp.* and algae (Fig.1, 2).



Fig. 1. Canal from Ai Simios, site 38.



Fig. 2. Canal from Ai Simios, site 38.

Aitoliko streams. The two systems near Aitoliko sampled in this survey, are two small streams that flow into the Aitoliko lagoon. Aitoliko stream 1 is a relatively wide stream. Sampling site 39 was close to its mouth, and the riparian vegetation consisted of a big Eucalyptus tree, *Phragmites* reeds, and *Arundo* reeds. Its aquatic vegetation consisted of *Lemna spp.* and algae.



Fig. 3. Site 39, Aitoliko stream 1.



Fig. 4. Site 39 with emergent reeds and *Lemna spp.* (a close up of the site is shown to the right).



The second system that was investigated in the Aitoliko area was Aitoliko stream 2, with very dense *Phragmites* reeds and *Calistygia spp.* vegetation, as well as some water crawfoot (*Ranunculus aq.*)



Fig. 5. Site 40. Spring area of Aitoliko stream 2, thick with *Phragmites.*



Fig. 6. Site 40. Close up.



Fig. 7. Site 40.



Fig. 9. Location further downstream from the spring area.



Fig. 8. Site 40, with *Ranunculus aq.* aquatic plants.



Fig. 10. Close up of the same location with water cress-like aquatic plants.

Rocky spring, Kaskariki marshy springs and Oiniades springs

All three systems discharge their water in the deep Kyanavlako drainage canal. The first site (41) is a spring area originating under a rock with slow flowing and clear water, rocky substrate and submerged algae. 100 m downstream from the site was a big cluster of the threatened plant species *Cladium mariscus* (Fig.11-12). Site 42

(Kaskariki springs) is a spring fed stream with fast flow, forming together with other nearby springs a marshy area with many reeds (Fig.13-14). The water discharge of both the Rocky spring and the Kaskariki springs has been extremely diminished due to water abstraction and wetland drainage through the Kynavlako canal. The third site, site 43 is a spring area at the ancient port of Oiniades with warm, very slow flowing and turbid water and no riparian or aquatic vegetation (Fig.15-16). The water of all three spring systems feeds the main drainage canal of the area, the Kynavlako canal, which during the current survey was being dredged and cleared of its vegetation (Fig.17-18).



Fig. 11. Site 41, Rocky spring with clear water, rocky substrate and submerged algae.



Fig. 12. A big cluster of *Cladium mariscus* at site 41.



Fig. 13. Site 42, Kaskariki springs with *Cladium mariscus* (left), *Typha spp.* (background) and forbs (right) .



Fig. 14. Close up of site 42. Shallow water with fast flow, rocky substrate and no aquatic vegetation.



Fig. 15. Site 43, Oiniades springs, with warm, turbid water.



Fig. 16. The water from Oiniades is diverted through a ditch to the Kyanvlako canal.



Fig. 17. The Kyanvlako canal. A very wide drainage canal into which is diverted the water of many springs in the area.



Fig. 18. The Kynavlako canal, being dredged and cleared of its vegetation.

Lake Alexaina

This is a big and deep karstic lake, lined with willows, chaste trees and Typha spp. with some characteristic Potamogeton spp. aquatic plants.



Fig. 19. Site 44, Lake Alexaina, a big lake with Fig. 20. Lake Alexaina. turbid water.







Fig. 21. Lake Alexaina with Vitex spp. (left and Fig. 22. Potamogeton spp. in Lake Alexaina. right bank)

Fraxos water forest

This is a unique system with underground springs that form deep ponds (called "eyes" by the locals) supporting a unique population of Fraxinus angustifolia trees. A diverse and unique ecosystem threatened in the past by constant encroachment to create arable land and currently by continuous lowering of its water levels and poor management (Fig.23-26).





Fig. 24. Site 45, a big spring pond in Fraxos water forest fed by underground springs.

Fig. 23. Site 45, the Fraxos water forest with tall Fraxinus trees.



Fig. 25. A big spring pond in Fraxos water forest with *Gambusia affinis* fish among dead tree trunks.



Fig. 26. A small pond in Fraxos water forest, almost dry.

TABLE 2. Data	on	the	riparian	and	aquatic	vegetation	at	the	Acheloos	sampling
sites.										

site	riparian	% r.v.	aquatic	% a.v.	surface
	vegetation		vegetation		cover
38	Calistygia spp.	50	Lemna spp.	30	70
	Typha spp.	10	Algae	30	
	Phragmites spp.	10			
	Arundo spp.	5			
	Forbs (non grass)	30			
39	Eucalyptus spp.	10	Lemna spp.	50	70
	Apiaceae	10	Algae	50	
	Phragmites spp.	60			
40	Calistygia spp.		Ranunculus aq.	60	70
	Phragmites spp.				
41	Grasses	10	Submerged algae	60	5
	Calitriche sp.	10	Calitriche sp.	10	
	Forbs	30			
	Typha spp.	20			
42	Typha spp.	40	None	0	0
	Forbs	20			
	Juncus spp.	10			
	Cladium mariscus	20			
	Water cress	10			
43	None	0	None	0	0
44	Scirpus spp.	10	Potamogeton spp.	5	
	Vitex spp.	80			
45	Fraxinus angustifolia	20	None	0	0
	Typha spp.	10			
	Bramble	70			
	Annual perennial	20			
	plants				

FISH SPECIES COMPOSITION

Table 3 summarizes the fish species composition at the Acheloos river sampling sites and Map II their spatial distribution at the area. The fish species encountered were *Pseudophoxinus stymphalicus* and *Tropidophoxinellus hellenicus* (Cyprinidae), *Knipowitschia sp.* and *Economidichthys pygmaeus* (Gobiidae), *Cobitis trichonica* (Cobitidae), *Gambusia affinis* (Poecillidae) and *Valencia letourneuxi* at one site (site 40). Note the presence of *Tropidophoxinellus hellenicus* (Cyprinidae) which is found only in western Central Greece and in Pinios river in the Peloponnese, that of *Cobitis trichonica*, a species endemic to the Acheloos basin only and that of the goby, *Economidichthys pygmaeus*, found in lakes and streams of Western Central Greece and Ipeiros.

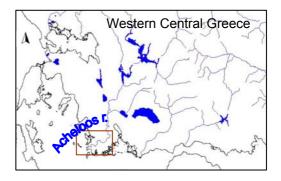
V. letourneuxi was found in one system (Aitoliko stream 2, site 40) but at low densities (1 individual out of a total of 39).

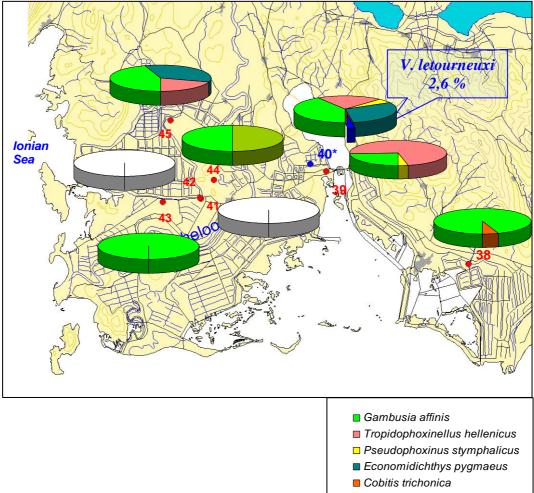
site	Sampling	Fish species	%	No	<i>V.</i>
	equipment				letourneuxi
38	scoop net	Gambusia affinis	94,7	19	•
		Cobitis trichonica	5,3		
39	scoop net	Gambusia affinis	24,2	62	
		Tropidophoxinellus hellenicus	72,6		•
		Pseudophoxinus styphalicus	3,2		
40 *	scoop net	Gambusia affinis	41,0	39	
		Tropidophoxinellus hellenicus	20,5		
		Pseudophoxinus styphalicus	5,1		•
		Economidichthys pygmaeus	30,8		
		Valencia letourneuxi	2,6		
41	scoop net	No fish	0	0	•
42	scoop net	No fish	0	0	•
43	scoop net	Gambusia affinis	100,0	30	•
44	Scoop net	Gambusia affinis	50,0	46	•
		Knipowitschia sp.	50,0		
45	Scoop net	Gambusia affinis	45,0	20	
		Economidichthys pygmaeus	35,0		•
		Tropidophoxinellus hellenicus	20,0		

TABLE 3. Fish species composition at the Acheloos estuarine area sampling sites.

* First record

MAP II. Species distribution at the Acheloos river sampling sites. *V. letourneuxi* was recorded at one site (•) – site 40* at low densities. * First record





- Knipowitschia sp.
- Valencia letourneuxi

CURRENT VERSUS HISTORICAL PRESENCE OF *V. LETOURNEUXI* IN THE ACHELOOS RIVER BASIN

V. letourneuxi was first reported in water systems of the Acheloos river basin by *Barbieri* et al. (2000). It was found at two sites (near ancient Oiniades and at Fraxos forest) which probably correspond to sites 43 and 45 of the current survey, in which however it was not possible to confirm the presence of the species. It had also been found by I. Leonardos in stream Ai Simios near the Technological Institute of Mesolongi (personal communication), a site which corresponds to site 38 of the current study, in which again it was not possible to confirm *V. letourneuxi* presence. However, a new population was found in a small stream near Aitoliko (site 40, see Table below), flowing to Aitoliko lagoon. I. Leonardos had kindly provided us with information about the existence there of a *V. letourneuxi* population.

References	V. letourneuxi presence
Barbieri <i>et al.</i> (2000)	•
Current Survey	•

POPULATIONS FIRST RECORDED HERE

Aitoliko stream 2

3.1.6 AGIOS DIMITRIOS RIVER

The Agios Dimitrios river is a rather small but important in discharge river system, fed mainly by the Lampra springs (Map I). Into Agios Dimitrios flow also the waters of Geroporos stream (largely an intermittent stream) from the north and those of Pentalofou springs from the west.

SAMPLING STATION DISTRIBUTION

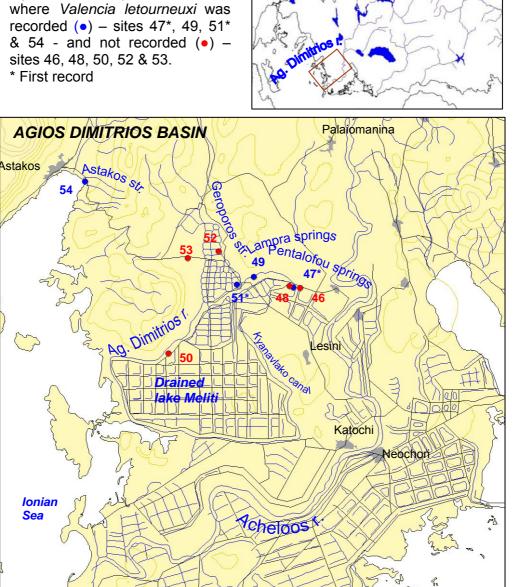
During the current survey a total of 9 sites were sampled at the Agios Dimitrios system. These included three sites of the Pentalofou springs system (site 46 - Mira Mill, 47-pumping station and 48 – downstream from the pumping station), two sites of the Agios Dimitrios river (site 49 – fish farm and 50 – further downstream), two sites at the Geroporos stream (one near its confluence with Agios Dimitrios, site 51 and one further upstream – site 52), an isolated pond (site 53 - pond by Markoutsa) and finally the Astakos springs, further north, a very small spring-fed stream near the town of Astakos (site 54). Table 1 provides a short description and pressure list of these sites and Map I a spatial distribution of the sampling sites.

site	location	water body type	pressures
46	Pentalofou springs - Mira Mill	Springs	Water abstraction, agriculture, wetland drainage
47	Pentalofou springs - pumping station	Springs	Water abstraction
48	Pentalofou springs - downstream	Spring fed canal	Water abstraction
49	Agios Dimitrios river - defunct fish farm	River	Pollution from small animal husbandry unit, irrigation, wetland drainage
50	Agios Dimitrios - downstream, close to the estuaries	River	Pollution from small animal husbandry unit, irrigation, wetland drainage
51	Geroporos stream -silos, confluence with Agios Dimitrios river	Stream	Canalization, irrigation, pollution from agrochemicals
52	Geroporos stream - upstream from silos	Stream	Canalization, irrigation
53	Pond by Markoutsa	Pond	Wetland drainage, road
54	Astakos springs	Springs	Pollution from animal husbandry, garbage disposal, road

TABLE 1. Sampling sites at the Agios Dimitrios basin.

Western Central Greece

MAP I. Sampling sites at the Agios Dimitrios system. Localities where Valencia letourneuxi was recorded (•) - sites 47*, 49, 51* & 54 - and not recorded (•) sites 46, 48, 50, 52 & 53. * First record



C

PRESSURES

The Agios Dimitrios system is a relatively pristine system with sole evident threats the water abstraction for irrigation purposes and some point source pollution from small husbandry units. In contrast, the Astakos springs (part af a larger wetland area), is a much degraded small system due to husbandry pollution, garbage disposal and past alignment and wetland drainage.

HABITAT CHARACTERISTICS

In terms of habitat characteristics the Agios Dimitrios sampling sites included spring areas and small spring-fed streams (sites 46, 47, 48 and 54), a pond (site 53), canals (51, 52) and open riverine areas (49, 50).

Pentalofou springs

This is a system of springs discharging their water in a canal connected with the Kyanavlako canal. Three sites were sampled at this system. The first site (site 46, Mira mill) was a small spring system, with clear water, dense riparian vegetation consisting of *Phragmites* reeds, *Typha spp.* and a lot of bramble and annual perennial plants (Fig.1-2). The second site (site 47, pumping station) is a wide spring area, rather pristine, with *Phragmites* reeds and a big *Ficus* tree at its banks and many aquatic plants, i.e. water cress-like plants and *Sparganium spp.* (Fig. 3-6). The third site (site 48) is a canal fed by the Pentalofou springs, rather wide (8 m), with cobble, pebble and gravel bottom and fast flowing water. Its banks are covered with *Typha spp.*, annual weeds and grasses and many aquatic plants (*Potamogeton spp.* and water cress, Fig.7-10).





Fig. 1. Site 46, Pentalofou springs – Mira mill, with dense vegetation, i.e. bramble (left) and forbs (right).

Fig. 2. Close up of site 46.



Fig. 3. Site 47, Pentalofou springs – pumping station.



Fig. 4. Site 47, with big *Ficus* tree, *Phragmites* reeds (left) and bramble.



Fig. 5. Site 47.



Fig. 7. Site 48, downstream from Pentalofou springs. A wide canal with grassy banks.



Fig. 9. Site 48, with *Potamogeton spp.* (brown aquatic plants) and some water cress-like plants (bright green).



Fig. 6. Site 47, with water cress-like plants and *Sparganium spp.*



Fig. 8. Site 48 with *Typha spp.* and annual weeds.



Fig. 10. A side pool of site 48, with gravel substrate and some water cress-like aquatic plants.

Agios Dimitrios river

The Agios Dimitrios river is mainly fed by the Lampra springs (Fig.11-13). Two sites along the river were sampled in the frame of the current survey, i.e. site 49 (fish farm) at a location with a now defunct fish farm, and a second site (site 50) further down stream. Site 49 is a wide section of the river (12 m), right by a big sluice, with some aquatic plants (water cress-like, *Potamogeton spp.*, *Ranunculus spp.* and *Luwidgia spp.*) This is a relatively undisturbed site with rich aquatic and insect life (Fig. 14-16). The second site, site 50 is also an open riverine area, further downstream, with *Phragmites* reeds and some *Potamogeton spp.* aquatic plants (Fig.17-18).

"Rapid Assessment of the Status of *Valencia letourneuxi*, the Greek Killifish" Final Report, October 2006



Fig. 11. A big spring feeding Agios Dimitrios river, with a pump installed by a local sheep farmer (Lampra springs).



Fig. 13. A second, smaller spring, with fern.



Fig. 15. Site 49. The big sluice.



Fig. 12. The big spring of the previous photo with a *Fraxinus* tree.



Fig. 14. Site 49, Agios Dimitrios river, sluice.



Fig. 16. Site 49 with submerged *Potamogeton spp.* (brown plants) and some water cress-like plants.

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Fig. 17. Site 50. Open riverine area with no floating vegetation.



Fig. 18. Site 50, with Phragmites reeds at its banks and some *Potamogeton spp.* aquatic plants (bottom left).

Geroporos stream

Two sites along the lower reaches of Geroporos stream were sampled during this survey. The first, site 51, is close to its confluence with the Agios Dimitrios river (near some large silos) and the second, site 52, further upstream. Site 51 is a rather wide canal section with rich aquatic vegetation (mostly *Potamogeton spp.,* Fig. 19-22) while site 52, further upstream, is a much narrower and undisturbed stream section, with dense *Phragmites* reeds at its banks, as well as emergent *Phragmites* reeds and many water cress-like plants (Fig. 23-24).



Fig. 19. Site 51. Canal by large silos.





Fig. 21. Site 51, with *Potamogeton spp.* mats (brown) and water cress-like plants (bottom right).

Fig. 20. Site 51, with *Phragmites* reeds at left bank (right bank cleared).



Fig. 22. Disposed bottle of agrochemicals at site 51.



Fig. 23. Site 52, with dense *Phragmites* reeds and some fern at its banks.



Fig. 24. Site 52, with water cress-like plants (bright green) and emergent *Phragmites* reeds.

Pond by Markoutsa and Astakos springs

Two additional sites were sampled to the north of the Agios Dimitrios river. The first site, site 53 (Pond by Markoutsa) is a lake-like pond with tall *Scirpus* reeds and warm and turbid water (Fig. 25-26). The second site, site 54 is a small spring-fed stream near the town of Astakos, with *Phragmites* reeds and very dense mats of brown thin *Zanichelia spp.* aquatic plants replaced by ulva algae closer to the sea (Fig.27-30).



Fig. 25. Site 53, a large lake-like pond by Markoutsa .



Fig. 27. Site 54, The Astakos stream with *Phragmites* reeds at one bank and dirt road at the other.



Fig. 26. Site 53.



Fig. 28. Site 54, the Astakos springs filled with garbage.



Fig. 29. The Astakos stream, close to its springs, covered with dense *Zanichelia spp.* aquatic plants.



Fig. 30. Closer to the sea, the Astakos stream is covered by dense mats of ulva algae.

TABLE 2. Data on the riparian and aquatic vegetation at the Agios Dimitrios sampling sites.

site	riparian	% r.v.	aquatic	% a.v.	surface
	vegetation		vegetation		cover
46	Phragmites australis	60	Water cress-like	5	5
	Brable	30			
	Forbs	20			
	Equicetum spp.	2			
	Maiden hair fern	5			
47	Phragmites australis	10	Water cress-like	40	50
	Ficus spp.	10	Sparganium spp.	20	
	Bramble	40			
	Forbs	20			
48	Typha spp.	10	Potamogeton spp.	30	10
	Grasses	10	Luwidgia spp.	5	
	Iris spp.	10	Water cress-like	10	
	Sparganium spp.	20			
	Forbs	30			
49	Phragmites australis	70	Potamogeton spp.	20	30
	Forbs	10	Water cress	25	
	Salix alba	5	Luwidgia spp.	5	
	Ficus spp.	5	Juncus spp.	5	
50	Phragmites australis	80	Potamogeton spp.	5	10
	Juncus spp.	60	Ceratophyllum spp.	5	
	Cladium mariscus	50	Luwidgia spp.	2	
51	Phragmites australis	50	Potamogeton spp.	30	40
			Phragmites australis	10	
			Water cress-like	70	
			Water lilies	20	
			(Nymphaea)		
52	Phragmites australis	90	Phragmites australis	20	40
	Water cress	5	Water cress-like	60	
	Calistygia spp.	5			
	Luwidgia spp.	5			

53	Scirpus spp.	10	Algae	10	10
	Phragmites australis	50			
	Typha spp.	20			
	Juncus spp.	20			
	Forbs	20			
	Tamarix spp.	5			
54	Phragmites australis	50	Zanichelia spp.	100	100
	Typha spp.	50	(ulva algae closer to		
			the sea)		

FISH SPECIES COMPOSITION

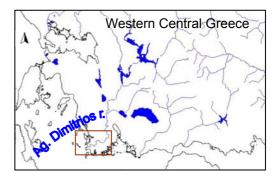
Table 3 summarizes the fish species composition at the Agios Dimitrios system sampling stations and Map II their spatial distribution at this area. The fish species encountered were *Telestes pleurobipunctatus, Pseudophoxinus stymphalicus, Tropidophoxinellus hellenicus* (Cyprinidae), *Economidichthys pygmaeus* (Gobiidae) *Gambusia affinis* (Poecillidae), *Anguilla anguilla* (Anguillidae), species of the Mugilidae family and *Valencia letourneuxi. V. letourneuxi* was recorded at four sites (47, 49, 51 and 54) with densities ranging from 2,8% to 5,7%). The 100% density of station 47 is misleading, as it was only one fish caught at this site.

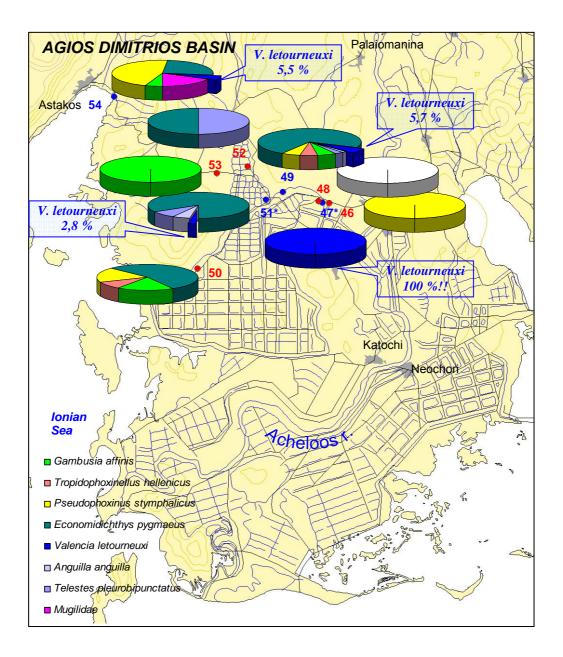
site	Sampling	Fish species	%	No	<i>V.</i>
	equipment				letourneuxi
46	Scoop net	Pseudophoxinus stymphalicus	100,0	2	•
47*	Scoop net	Valencia letourneuxi	100,0	1	•
48	Scoop net	No fish	0	0	•
49	Scoop net	Gambusia affinis	5,7	70	
	Sein net	Tropidophoxinellus hellenicus	5,7		
		Pseudophoxinus stymphalicus	5,7		
		Economidichthys pygmaeus	72,9		•
		Anguilla anguilla	1,4		
		Telestes pleurobipunctatus	2,9		
		Valencia letourneuxi	5,7		
50	Scoop net	Gambusia affinis	18,2	11	
		Tropidophoxinellus hellenicus	9,1		•
		Pseudophoxinus stymphalicus	18,2		
		Economidichthys pygmaeus	54,5		
51*	Scoop net	Economidichthys pygmaeus	83,3	36	
		Anguilla anguilla	5,6		•
		Telestes pleurobipunctatus	8,3		
		Valencia letourneuxi	2,8		
52	Scoop net	Economidichthys pygmaeus	50,0	6	•
		Telestes pleurobipunctatus	50,0		
53	Scoop net	Gambusia affinis	100,0	31	•
54	Scoop net	Gambusia affinis	5,5	91	
		Pseudophoxinus stymphalicus	46,1		
		Economidichthys pygmaeus	25,3		•
		Mugilidae	17,6		
		Valencia letourneuxi	5,5		

TABLE 3. Fish species composition at the Agios Dimitrios sampling sites.

* First record

MAP II. Species composition at the Agios Dimitrios sampling sites. *V. letourneuxi* densities ranging from 2,8% to 5,7%. The 100% density of station 47 is misleading, as only one fish was caught at this site.





CURRENT VERSUS HISTORICAL PRESENCE OF *V. LETOURNEUXI* IN THE AGIOS DIMITRIOS SYSTEM

V. letourneuxi was first reported by *Barbieri* et al. (2000) at the springs of Agios Dimitrios and at a site 200 m downstream (also reported in *Economou* et al. 1999, Technical report) which corresponds to site 49 of the current survey, where *V. letourneuxi* presence could be confirmed. At the Astakos springs, *V. letourneuxi* presence was first reported in *Daoulas* (2003) and confirmed in the current survey. Two new populations of *V. letourneuxi* were found during the current survey, the Pentalofou springs population (site 47) and the Geroporos stream one (site 51). Both systems are connected with the Agios Dimitrios river (see Table below).

References	V. letourneuxi presence
Barbieri <i>et al.</i> (2000)	•
Economou et al. (1999)	•
Daoulas (2003)	•
Current Survey	•

POPULATIONS FIRST RECORDED HERE

Pentalofou springs Geroporos stream

3.1.7 VLYCHOS SPRINGS

These are littoral springs with brackish water (3,6 %), adjacent to Myrtari or Vonitsa lagoon, flowing into the Amvrakikos Gulf, with average discharge 0.3 m³/s. At the area of the springs the aquatic vegetation is very rich.

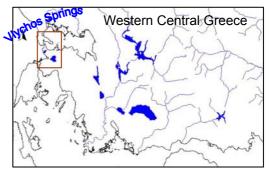
SAMPLING STATION DISTRIBUTION

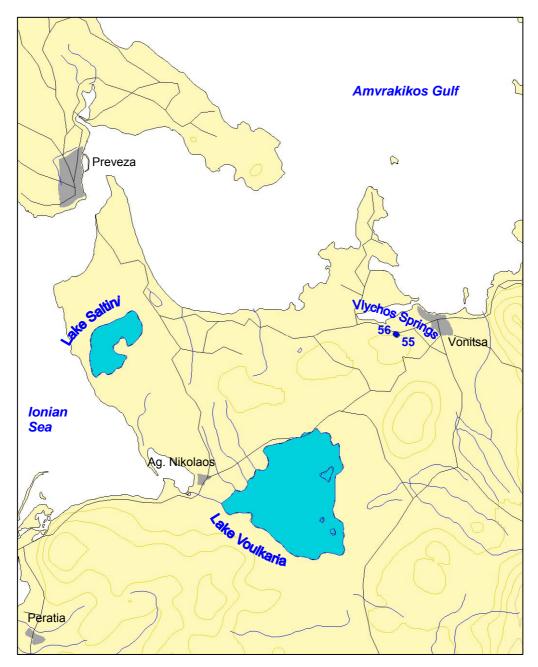
In the course of the current survey, two sites at the area of the Vlychos Springs were sampled for *V. letourneuxi* occurrence. The first site –site 56 was the actual area of the springs and the second site –site 55 - a ditch fed by the springs through a culvert. At the time the sampling took place (October) which was at the end of the dry season, this was cut off from the springs (Table 1 provides a short description of these sites with the relevant pressure list and Map I shows the sampling sites at the Vlychos Springs area).

TABLE 1. Sampling sites at the Vlychos Springs area.

site	location	water body type	pressures
55	Ditch associated with springs	Ditch	Adjacent main road, animal husbandry in the area
56	Vlychos springs	Springs	Adjacent main road, animal husbandry in the area

MAP I. Sampling sites at the Vlychos Springs area. *V. letourneuxi* was recorded at both sampling localities (•) – sites 55 and 56.





HABITAT CHARACTERISTICS

The ditch associated with the Vlychos springs – site 55 – is a 6 m wide, 30-70 cm deep canal, with slightly turbid and brackish water (3,5 ‰) and relatively high water temperature (21,4 °C). This ditch is connected with the springs through a culvert under a dirt road. The vegetation at the banks of the Vlychos ditch consisted of *Phragmites australis*, *Typha spp., Juncus spp. Arundo spp.,* and some water cress-like aquatic plants. The water surface was covered by a thick mat of algae, over submerged *Ceratophyllum spp.* aquatic plants (Fig.1-6).



Fig. 1. Ditch from Vlychos springs – site 55-, with dense *Phragmites* reeds on the right bank and *Juncus spp.* on the left bank. Note the extensive algae mats on the water surface.



Fig. 3. Close-up of site 55, with *Phragmites* reeds and some *Arundo* cane (top left).



Fig. 5. Site 55, with *Phragmites* reeds (left bank) and *Juncus spp.* (right bank).



Fig. 2. Ditch from Vlychos springs, with *Phragmites* reeds and water cress-like aquatic plants (top).



Fig. 4. Close up of site 55, with thick algae mat.



Fig. 6. Culvert under road connecting the ditch (site 55, left) with the Springs (site 56).

The second site, site 56, was the area of the Vlychos springs. These springs create a large marshy pool, connected with the Vlychos ditch through a culvert. The riparian vegetation of that site, was quite similar to that of the Vlychos ditch, with *Phragmites aus.*, *Typha spp.*, and *Juncus spp.* as well as some *Rubus spp.* and water cress-like plants. In contrast, however, to the Vlychos ditch, the aquatic vegetation was less thick, and consisted not only from *Ceratophyllum spp.* and algae but also from *Lemna spp.* (Fig. 7-10).



Fig. 7. The Vlychos springs - site 56 - which create a large marshy pool.



Fig. 9. Close up of the Vlychos springs, with some water cress-like plants (left).



Fig. 8. Site 56 with *Juncus spp.* (left), *Phragmites* reeds and *Typha spp.* (right).



Fig. 10. The culvert that connects the springs with the Vlychos ditch. Note the bright green *Lemna spp.* aquatic plants.

TABLE 2. Data on the riparian and aquatic vegetation at the Vlychos Springs sampling sites.

site	riparian	% r.v.	aquatic	% a.v.	surface
	vegetation		vegetation		cover
55	Phragmites spp.	40	Ceratophyllum spp.	90	50
	Typha spp.	15	Algae	70	
	Arundo spp.	5	Water cress-like	3	
	Juncus spp.	10			
	Water cress	3			
56	Phragmites spp.	30	Ceratophyllum spp.	25	10
	Typha spp.	60	Algae	75	
	Juncus spp.	10	Lemna spp.	10	
	Rubus spp.	5			
	Water cress	5			

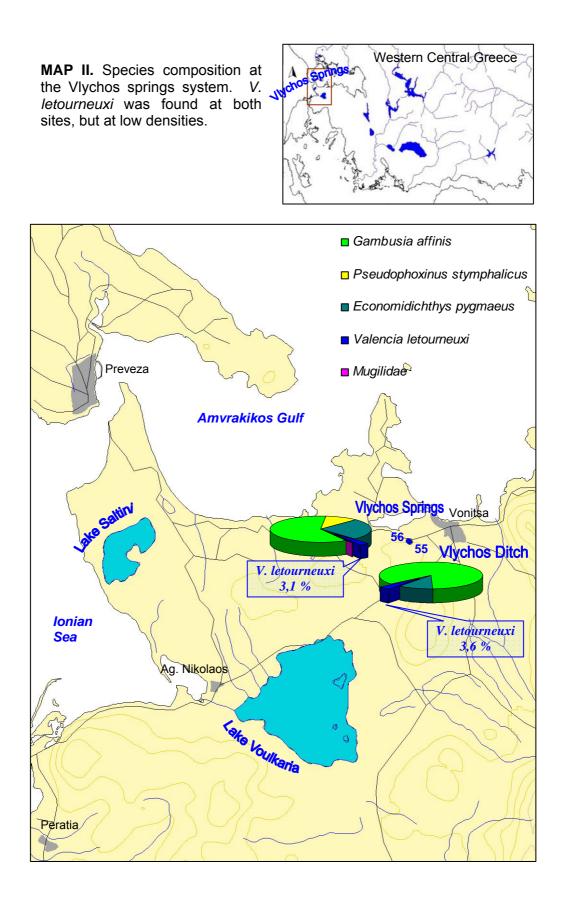
FISH SPECIES COMPOSITION

Table 3 summarizes the fish species composition at the Vlychos springs area sampling sites and Map II their spatial distribution at the area. The fish species encountered were *Pseudophoxinus stymphalicus* (Cyprinidae) – only at site 56, the actual spring area -, *Economidichthys pygmaeus* (Gobiidae), *Gambusia affinis* (Poecillidae) and *Valencia letourneuxi* at both sites. At site 56 was also fished 1 individual of the family of Mugilidae (out of a total of 325).

V. letourneuxi was found at both sites, but at low densities (11 individuals out of a total of 305 in the Vlychos ditch (3,6%) – site 55, and 10 individuals out of a total of 325 in the Vlychos springs (3,1%) -site 56).

site	Sampling equipment	Fish species	%	No	V. letourneuxi
55	scoop net dip net	Gambusia affinis Economidichthys pygmaeus Valencia letourneuxi	85,6 10,8 3,6	305	•
56	scoop net dip net	Gambusia affinis Pseudophoxinus stymphalicus Economidichthys pygmaeus Mugilidae Valencia letourneuxi	60,0 10,5 26,1 0,3 3,1	325	•

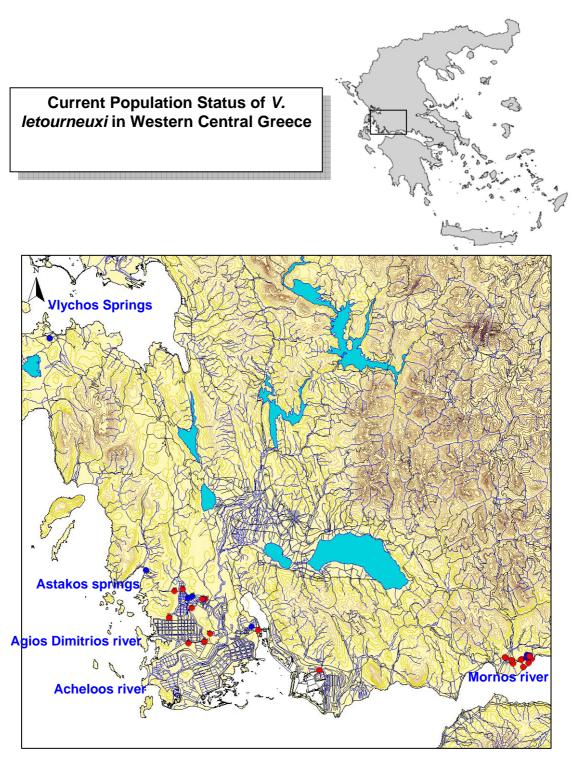
TABLE 3. Fish species composition at the Vlychos springs sampling sites.



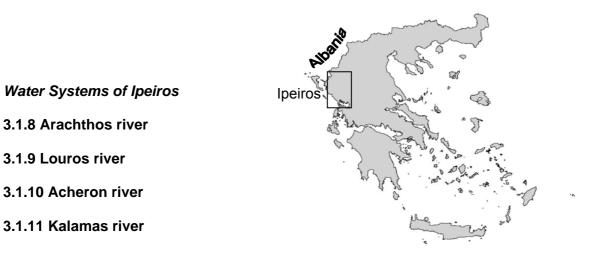
CURRENT VERSUS HISTORICAL PRESENCE OF *V. LETOURNEUXI* IN THE VLYCHOS SPRINGS

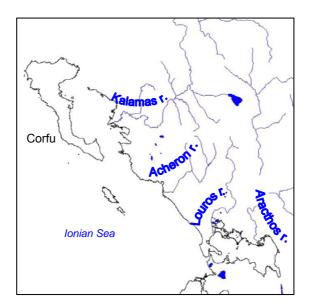
V. letourneuxi presence in the Vlychos system (confirmed also in the current survey) was first reported by *Barbieri* et al. (2000). It was first found in the ditch discharging from the Vlychos springs, (see Table below).

References	V. letourneuxi presence
Barbieri <i>et al.</i> (2000)	•
Current Survey	•



In the frame of the current survey, four water systems of Western Central Greece were sampled for *V. letourneuxi* presence (from East to West, the Mornos river, the Acheloos river, the Agios Dimitrios river - with the Astakos stream -, and the Vlychos springs). Out of a total of 36 sampling stations, *V. letourneuxi* presence was confirmed in a total of 11 sites (•). More specifically, it was found in the Chiliadou springs in Mornos basin at relatively high densities (ranging from 11,3 to 20,1% of total fish caught), in the Aitoliko stream 2 at low densities (2,6%), in the Pentalofou springs, the Agios Dimitrios river, the Geroporos stream and the Astakos stream of the Agios Dimitrios system at low densities, ranging from 2,8 to 5,7%, and at the Vlychos springs also at low densities (3,1 and 3,6%). It becomes thus obvious that, with the exception of the Chiliadou springs where the species is relatively abundant, *V. letourneuxi* can be characterised as locally rare in that region.





3.1.8 ARACHTHOS RIVER

Arachthos river is the largest river of Ipeiros both in terms of discharge as well as basin size. Its length is 106 km and the size of its basin approximately 2000 km² with 7 major tributaries. Its average annual discharge at the entrance of the Pournari Dam is 68 m³/s but with seasonal variation. Its springs forth from the Peristeri mountain range (Central Pindos) and, after flowing through the prefecture of Arta, it discharges into the Amvrakikos Gulf. It flows mostly through deep erosive valleys and as a result it is characterized by high and variable flow. Its basin consists mostly of impenetratable rockbeds and sites of high erosion, which causes fluctuations in its discharge. Also, the river is characterized by large quantities of sediments.

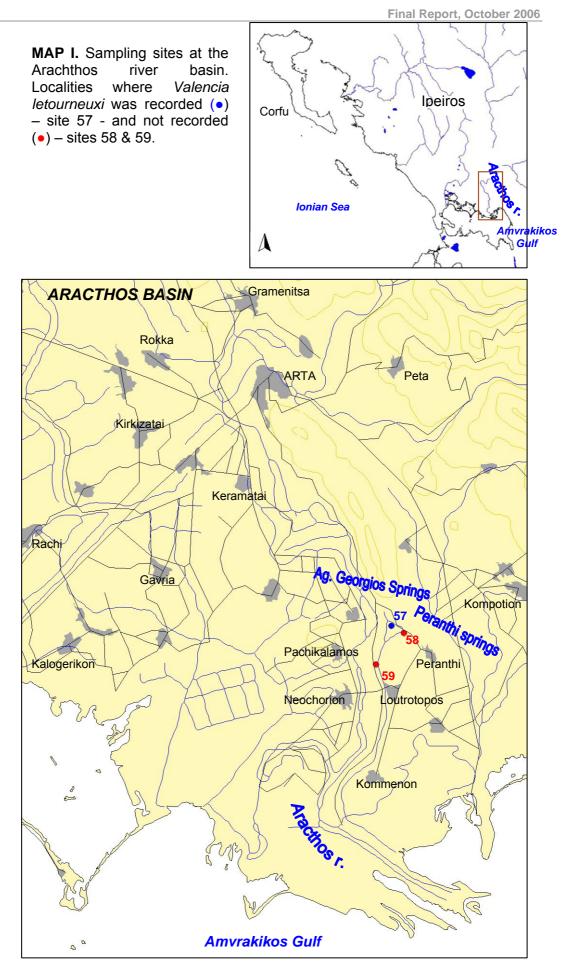
It's a pluvio-nival river. Due to the large size of its basin and the exceptionally high average annual precipitation (2400 mm/year at the Tzoumerka Mountains), surface discharge is high. It is however also fed by two big karstic systems. Two artificial reservoirs have been created at the site Pournari for hydroelectric, as well as irrigation, purposes. Its Delta (80 km²) is designated by the Ramsar treaty as a wetland of international importance and is protected by a series of Greek and European decrees.

SAMPLING STATION DISTRIBUTION

The current survey focused on the lower area of the Arachthos system and more specifically on two canals flowing into Arachthos river. A total of three sites were sampled, i.e. site 57 at a narrow canal fed by the waters of Agios Georgios Springs, very close to the springs, site 58 at the same canal near the discharge of Peranthi Springs and site 59 at a canal also fed by Agios Georgios Springs but much further downstream from the spring area.

site	location	water body type	pressures
57	Agios Georgios springs	Spring fed canal	Vegetation clearing, adjacent road, agrochemical pollution
58	Peranthi springs	Canal	Adjacent road
59	Agios Georgios ditch	Ditch	Citrus and olive groves

TABLE 1. Sampling sites at the Arachthos river basin.



PRESSURES

The main threats to the Arachthos river are the operation of dams, road works, point source pollution and water abstraction from upstream dams and adjacent springs. The operation of the dams causes fluctuations in the water discharge, affecting thus negatively the river's flora and fauna. The lower part of the river suffers from agricultural pollution as well as urban waste pollution.

HABITAT CHARACTERISTICS

All sites sampled during the current survey were spring-fed canals flowing into the Arachthos river.

Agios Georgios springs. The first site (site 57) was a canal, fed by the Agios Georgios Springs, adjacent to the road to Peranthi village, with very clear and relatively cool (17.7° C) water and low flow. One bank was cleared of its vegetation and that of the other bank consisted of some *Phragmites* reeds and bramble and mostly grasses and perennial plants (Fig.1, 2). Its aquatic vegetation consisted of water cress-like plants, *Lemna spp.* and an unidentified submerged plant (Fig. 3, 4). The canal is obviously often used for the dilution of agrochemicals (chemical fertilizers and pesticides, Fig. 5, 6).



Fig. 1. Site 57, a spring-fed canal with rich floating vegetation.



Fig. 3. Close up of site 57 with Lemna spp.



Fig. 2. Site 57, adjacent to road to Peranthi.



Fig. 4. Close –up of site 57 with *Lemna spp.* (top left) and unidentified submerged plant.

"Rapid Assessment of the Status of *Valencia letourneuxi*, the Greek Killifish" Final Report, October 2006



Fig. 5. Disposed bottles of agrochemical at site 57.



Fig. 6. The label reads "extremely toxic for aquatic organisms".

Peranthi springs. The second site (site 58, Peranthi springs) was a site further downstream of site 57, directly fed by karstic springs. The canal controlled by a sluice then bends sharply to the east (Fig.7-10). The water was sulphurus with a temperature of 20 °C. The surface was covered by thick algae mats and the bottom by submerged algae.



Fig. 7. Peranthi springs (site 58). Sulphurus springs with dense algae mats.



Fig. 9. Peranthi springs.



Fig. 8. Site 58 with sluice.



Fig. 10. Site 58 with algae bed.

Agios Georgios ditch. This narrow ditch (site 59) probably originating from Agios Georgios springs too, with thick riparian vegetation (mostly *Arundo spp., Phragmites aus.* and bramble) and some aquatic vegetation (*Lemna spp.*, water cress-like and submerged algae).



Fig. 11. Site 59. Narrow canal with thick vegetation.



Fig. 12. Canal, with *Lemna spp.* and water cress-like plants near the banks.

TABLE 2. Data on the riparian and aquatic vegetation at the Arachthos sampling sites.

site	riparian	% r.v.	aquatic	% a.v.	surface
	vegetation		vegetation		cover
57	Phragmites aus.	10	Water cress-like	30	70
	Bramble	10	Lemna spp.	50	
	Grasses and	80	Unidentified	20	
	perennial plants		submerged plant		
58	Arundo spp.	5	Algae (floating)	20	20
	Bramble	30	Algae (submerged)	80	
	Grasses	20			
	Water cress-like	20			
59	Platanus orientalis	15	Lemna spp.	20	50
	Arundo spp.	35	Water cress-like	25	
	Bramble	10	Submerged algae	50	
	Phragmites aus.	15			
	Iris	10			
	Grasses	5			

FISH SPECIES COMPOSITION

Table 3 summarizes the fish species composition at the Arachthos sampling stations and Map II their spatial distribution. The fish species encountered were *Pseudophoxinus st. thesproticus* and *Telestes pleurobipunctatus* (Cyprinidae), *Economidichthys pygmaeus* (Gobiidae), *Cobitis hellenicus** (Cobitidae), *Gambusia affinis* (Poecillidae) and *Valencia letourneuxi* at one site (site 57).

* It is considered conspecific with the *Cobitis arachthosensis*

V. letourneuxi was found in one location at densities (18,6%) relatively high for this species.

site	Sampling	Fish species	%	No	<i>V.</i>
	equipment				letourneuxi
57	electrofishing	Pseudophoxinus st. thesproticus	1,7	59	•
	scoop net	Economidichthys pygmaeus	64,4		
	dip net	Telestes pleurobipunctatus	5,1		
		Gambusia affinis	10,2		
		Valencia letourneuxi	18,6		
58	scoop net	No fish	0	0	•
	dip net				
59	scoop net	Pseudophoxinus st. thesproticus	5,9	17	•
		Economidichthys pygmaeus	88,2		
		Cobitis hellenicus	5,9		

TABLE 3.	Fish species	composition	at the	Arachthos	sampling si	tes.
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