LIFE Potamo Fauna

International Symposium on Conservation of Native European Freshwater Crayfish

Olot, Girona (Spain)
23 - 25 September 2015

ABSTRACTS BOOK

Organization:

CONSORCI DE l'estany

Generalitat de Catalunya

Parc Natural de la Zona Volcànica de la Garrotxa

Consorci del TER

Colaboration:

Ajuntament d'Olot

Diputació de Girona

CSIC

Presentation

The conservation of native crayfishes is one of the major challenges in inland waters of Europa. Most species are endangered by many factors, but specifically by introduced species and pests. This symposium will focus on recent research advances, and also on management strategies and specific experiences to achieve a long term conservation of our native freshwater decapods.

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KN.I

Status of the native crayfish (*Austropotamobius pallipes*) in Spain

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Austropotamobius pallipes populations declined sharply after the first diagnosis of crayfish plague in Spain in 1978. From a lowest around 600 the number of its populations has nearly doubled during the last decade, partly due to better knowledge but also to the success of restoration programs undertaken at some areas. Nevertheless, recovery has been uneven, and fragmentation and small population size are still a great problem.

Both actual distribution, regional contribution to total numbers, survey intensity and methods and active restoration programs are reviewed.

The recent dispersal pattern of American crayfishes and its relationship with fisheries policies and regulations and its availability to general public is also discussed.

Finally, a set of recommendations are given in relation with the future conservation of the species and its habitats.

KN.II

Crayfish conservation in the Alps: strategies and lessons learnt

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Crayfish have played a significant role in the social and cultural activities of Europe since the Middle Ages, today however, native populations have disappeared or are highly threatened. Also the autochthonous crayfish of the Alpine countries *Austropotamobius pallipes*, *A. torrentium*, and *Astacus astacus*, have been exposed to various threats and their populations still are strongly decreasing – most of it has been portrayed as a consequence of human activities. In ongoing species protection programs carried out in the Austrian and Italian Tyrol, we have been undertaking measures to enhance the situation of all three indigenous species. This study aimed at documenting activities for the support of the three autochthonous and endangered crayfish species. As these measures are being applied on three different species and in two states with different legislation, we elaborated specific management plans for crayfish conservation. All had in common a comprehensive survey of the species' distribution in the regions, their populations' phenotypical and genotypical characterization, their habitat conditions and future potential. Based on these data, species and country specific conservation measure where defined and carried out. Now, after several years of the implementation of these programs we evaluated them and record on their success and deficits.

KN.III

The complex issue of crayfish plague in Finland

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The populations of the native noble crayfish (*Astacus astacus*) have suffered major losses since the appearance of crayfish plague in Finland. Crayfish was an important fishery product and an export article about a century ago, and around 15 million crayfish were exported yearly. This activity was practically annihilated through the first wave of crayfish mortalities caused by the spread of the crayfish plague agent *Aphanomyces astaci*. As we know now, this first introduction in 1893 was the crayfish plague strain As (or A) that started its way through Europe in Italy in 1860's.

In a few decades the crayfish populations disappeared from the main water bodies, but scattered subpopulations still survived in smaller lakes, as well as recovering populations after restocking or spontaneous reappearance. However, recovering noble crayfish populations often suffered repeated population crashes due to crayfish plague induced mortality. This instability led to the introduction of the North American signal crayfish (*Pacifastacus leniusculus*) first in 1967. While the noble crayfish had expanded its original territory to more northern parts of Finland, mostly aided by human activity, the signal crayfish started to take over the southern Finland. Unfortunately, hidden in the cuticle of its natural host, crayfish plague agent of the signal crayfish type, strain Ps1 (or B) found its way to Finland.

The present day situation with crayfish and crayfish plague in Finland can be described as a search of a balance between two parasites; crayfish plague strains As and Ps1, and two hosts; the noble and the signal crayfish. The original host of the first introduced strain, As, is not known and thus we don't know if this strain originally had the variability that we see today between the isolates from different localities. The long presence of this strain in Finland may have led to adaptation to the new host and environment, although the host is still too vulnerable and the infection with this strain causes several population crashes yearly. The signal crayfish type strain Ps1 shows high virulence towards the noble crayfish, but even the natural host the signal crayfish may suffer from negative effects of infection in its new environment. Practically all populations of signal crayfish are carriers of crayfish plague strain Ps1, while strain As has never been detected from them. Whether this is due to the diminished virulence of strain As remains to be clarified. While the signal crayfish acts as a permanent reservoir of the highly virulent Ps1 strain, the noble crayfish populations recovering from a crayfish plague episode caused by the As strain can carry a latent or chronic infection, that often surfaces when the population is strengthening again. This complexity demands effective management measures and diagnostic abilities in order to keep both the noble and signal crayfish stocks thriving and productive.

KN.IV

Rescue transfers: How to make the best?

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In last decades, there is an obvious decline in native crayfish populations throughout Europe. Czech Republic is not an exception. Moreover, several negative impacts still strengthen their role. Human impact on crayfish can be partly reduced by active approach in cases when crayfish or their habitats are directly threatened. Crayfish must be secured in some cases such as civil engineering or water management works, as well as revitalizations of ponds or running waters. So called "crayfish rescue transfers" are one of the key possibilities how to reduce the eventual risks for crayfish. Usual transfer consists of catching crayfish on the influenced locality, transfer to another suitable one and once the danger is over, return of crayfish to their original habitat. The key component is to choose a suitable and safe temporal adoptive locality.

Considering potential rescue transfer, firstly, it is necessary to become familiar with target locality and in situ crayfish status and quality of population of eventually affected crayfish. Wide range of aspects must be considered before rescue transfer itself, mainly for careful choosing of temporal locality: size of the locality, method of catching, length of supposed transfer, occurrence of NICS or diseases in surroundings, occurrence of predators which can affect transferred specimens or transmit diseases, potential manipulation with water or crayfish by third persons or influencing other endangered species (e.g. amphibians, fish, plants etc.). However, not only biologically relevant facts must be taken into account, but also official affairs play an important role. Manipulation with organisms protected by law cannot be done without confirmation of authorization for the persons, who will get into contact with the focal organism. Also agreements with owners of affected property must be well negotiated.

There is more than 10 years of experiences in crayfish rescue transfers at the Research Institute of Fish Culture and Hydrobiology in Vodňany (South Bohemia, Czech Republic). Our department was involved in rescue transfers of all three native or protected Czech crayfish species (*Astacus astacus*, *A. leptodactylus*, *Austropotamobius torrentium*). Our experiences were gained during different types of transfers from displacement of tens of stone crayfish from one part of the small brook to another one, up to weeks of collecting thousands of noble crayfish during the draining of water reservoir.

KN.V

Conservation plans in Spain. A management plan based on crayfish plague biology

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The Iberian Peninsula holds one single native species *Austropotamobius pallipes*, which is highly susceptible to crayfish plague. This disease was introduced into Spain in 1970s, via the North American crayfish, *Pacifastacus leniusculus* and *Procambarus clarkii*. Since them, the native species has experienced a dramatic decline and is now categorized as on risk of extinction by the majority of Local Governments. In the Pyrenean region of Navarra, it was implemented in 1996, a plan of management of native crayfish. This plan has been applied based on the knowledge on the biology of the crayfish plague pathogen, and has been complemented by a number of scientific and educational activities. These consisted of: (1) monitoring population of native and alien crayfish; (2) studying genetic diversity of native crayfish; (3) implementing health controls of both native and alien crayfish evaluating presence of crayfish plague and other disease; (3) evaluating disease resistance on native population and selecting more resistant individual for breeding; (4) establishing a breeding program of native crayfish; (4) identifying disease-risk free habitat for translocation of breaded specimens; (5) eradicating alien crayfish species by using biological control and other strategies; (6) implementing a diagnostic service for rapid diagnosing the causes of decline of native populations; (7) interacting with local fishing associations, and (8) having informative campaigns to local media and environmental local associations. As a result of the implementation of this plan, the native populations have increased from 55 in 1991 to 130 in 2014, and captures/year from 900 in 1991 to 25.000 in 2014. This plan has made possible to increase people awareness of native crayfish critical situation, and also that this species belongs to habitats that are threatened by destruction by humans, and alteration by alien species.

KN.VI

Active spreading of an invasive species challenges ecosystem-based management of crayfisheries

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We describe the impact of the alien crayfish introduction on the Finnish inland fisheries. The decision to introduce the alien signal crayfish was made to solve the problem of collapsing native noble crayfish stocks and crayfisheries due to the crayfish plague epidemics. The original idea was founded on overly optimistic expectations for adaptation of the alien signal crayfish, partially by ignoring the available warnings. The outcome of the massive stockings conducted during the last four decades now shows that the alien crayfish has not performed as expected and might even end up as being a failure resulting in catastrophical narrowing of the living space of the native noble crayfish. The alien crayfish are permanent reservoir hosts of the crayfish plague disease agent, *Aphanomyces astaci*, spread it efficiently and have been shown to experience elevated mortality when infected with *A. astaci*. In spite of this, the official policy in Finland is still driving towards the spreading of the alien crayfish to new areas and thus threatening original aquatic ecosystem functions and slowly leading in the elimination of the native crayfish. The policy includes, Fisheries Act (2015), National Alien Species Strategy (2013) and National Crayfisheries Strategy (2014), all contradicting EU alien species policy and ignoring commonly known behavior of people who tend to illegally transfer signal crayfish. We will also focus on the loss of the native crayfish and its cumulative effects in ecosystems and society. Part of the presentation is a 4 minute educational film on the relevance of a productive native crayfish population (in Finnish with English subtitles).

KN.VII

Alien crayfish management in Mediterranean areas. The Italian experience

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The North American red swamp crayfish *Procambarus clarkii* is the most widespread alien species in Italy as well as in the Mediterranean area. It is included in the 100 worst invasive species present in Europe for the heavy impacts caused on invaded ecosystem and native crayfish. In the light of the recent EU regulation on invasive species (1143/2014), the management of *P. clarkii* is thus necessary in order to mitigate its impacts and favour native species conservation. In Italy, within national and European projects (i.e. LIFE RARITY, LIFE SOS TUSCAN WETLANDS), several methods (e.g. intensive trapping, use of native fish predator, male sterilization technique, biocide as Pyblast) were developed and applied in different wetland areas. In the present contribution, the results of their applications together with their pro and cons are illustrated: an unique and efficacious method for all the habitat types seems not to exist, while the Integrated Pest Management approach, using a range of control and containment techniques to suit specific sites, is recommended to yield the best results.

KN.VIII

The decline of signal crayfish in Scandinavia

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Invasive species often have instable population dynamics and are known to collapse or oscillate heavily after passing through the initial lag/ growth phases. Long-term data-series documenting these fluctuations are however rare. We use long-term (starting in the early 1960s), semi-quantitative data on the invasive signal crayfish (*Pacifastacus leniusculus*), capturing its population development after introduction in 44 Swedish lakes. In total 18 (41 %) of these populations had experienced a collapse. A stepwise discriminant function analysis including 20 different ecological or physicochemical characteristics identified three variables explaining collapses in the following order: stocking year, population age and mean air temperature. Populations stocked in the 1980s were more likely to collapse than populations stocked in the 1970s. Lakes with collapses were located in areas with 0.4 C higher yearly mean air temperatures than the still viable populations. Collapses also depended on the time phase of the population and started to occur 12 years after stocking and were most frequent in the interval 16–20 years after stocking and after 11–15 years duration of the established phase with harvestable densities. An analysis of prevalence and pathogen load of crayfish plague was conducted. Plague was present in all populations but neither the level of prevalence nor the pathogen load in infested specimens differed between lakes with collapses and lakes without. Our results highlight the potential sensitivity and instability of introduced crayfish. The importance of density dependence and temperature suggest that both climate variability and/or fisheries can influence these processes. If the occurrence of these collapses were more generally known the illegal introductions of signal crayfish taking place today, threatening the native noble crayfish (*Astacus astacus*), would probably decline or even cease. This would be beneficial both for the conservation of the noble crayfish and for the fishery in itself.

Communications schedule

CODE	MAIN AUTHOR	TITLE
O.1	J. D. Reynolds	Increasing threats to last widespread populations of white-clawed crayfish austropotamobius pallipes in ireland
O.2	S. Llamas	Environmental education and awareness, fundamental tool in the conservation of the Iberian crab
O.3	N. Olarte	GIS as Guarantee in Success? Searching tool limits in freshwater crayfish management
O.4	B. Matallanas	Update of genetic information for the white-clawed crayfish in Spain
O.5	F. J. Oficialdegui	Invasion of fluvial ecosystems by red swamp crayfish, Procambarus clarkii
O.6	I. Vedia	Ethology of the invasive signal crayfish (Pacifastacus leniusculus) and its relationships with native fishes
O.7	F. C. Pricop	Assessing the crayfish ability to use anaerobic metabolism under microrefugia conditions
O.8	L. Martin-Torrijos	Susceptibility to the crayfish plague pathogen Aphanomcyes astaci of European native crayfish Austropotamobius pallipes: identification of a high resistant population

O.1

Increasing threats to last widespread populations of White-clawed crayfish *Asutropotamobius pallipes* in Ireland

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White-clawed crayfish occur in some 18 western countries of Europe, but in nearly all of these they are in sharp decline. This small European crayfish is protected under the EU Habitats Directive and no longer caught as food but it risks being overlooked or considered extinct.
All European crayfish are under pressure from environmental deterioration. Over most of its range this species is declining primarily because of crayfish plague carried by invasive North American species. Only in Ireland is this not so; there are no NICS, and white-clawed crayfish are widespread and considered close to stable.

While Northern Ireland is politically part of the UK, it shares environmental problems with the Republic of Ireland, and common drainage systems containing crayfish. Both territories must be vigilant against introductions of non-indigenous crayfish.

New threats have however arisen, chiefly through the ready availability of hundreds of exotic crayfish species on the Internet, in Europe chiefly supplied by German and Czech crayfish breeders and traders.
This presentation discusses threat levels to the last widespread stocks of *A. pallipes*, and outlines current conservation activities (ark sites, SACs, a crayfish farm with potential for restocking) and political and legal constraints to their successful protection.

O.2

Environmental education and awareness, fundamental tool in the conservation of the Iberian crayfish.

Sònia Llamas Comellas & Núria Valls Granero

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The Association of Defence and Study of Native Fauna & Flora (ADEFFA) runs a conservation program of the Iberian crayfish in the Llobregat river basin since 2005, including the study of populations, captive breeding, repopulation, removing invasive species, land custody contracts, habitat preservation and environmental education. All activities within the conservation program run from the Department of Agriculture, Livestock, Fisheries, Nutrition and Environment of the Generalitat de Catalunya. In addition, ADEFFA has signed with the Department a collaboration agreement for the conservation of native species.

ADEFFA believes that environmental education is an essential tool in the conservation of the Iberian crayfish, especially in the fight against the spread of invasive crayfish species. The main objective of ADEFFA's environmental education program is to present the risk factors that threat native crayfish populations and how people can contribute to their conservation. The activities are focused to general public, schools, and specific groups such as fishermen, hikers, etc.

ADEFFA owns an environmental education and wildlife conservation center in Santa Maria de Merlès (Barcelona) where many activities are held, aimed mainly for school and family audiences. More than 2500 school students and 2000 individuals of familiar public visit the center every year.
A highlight of the activities that has been under development for 5 years is the "Day of the invading crab catch at Merlès River" Merlès River is a protected area, which held large populations of Iberian crab, naiads and where native fish populations were well preserved. Due to the large tourist affluence, the space has been deteriorated and river middle and lower courses are currently presenting large populations of American red crayfish (*Procambarus clarkii*) and American signal crayfish (*Pacifastacus lenisculus*) in addition to numerous species of introduced fish.

The "Day of the invading crab catch" has a main purpose, whichis the environmental awareness of the participants related to the introduction of invasive crayfish species. Explaining how this species affect native crayfish, how invasive species deteriorate river ecosystems and the need for public to collaborate not expanding invasive species are the main goals of the date. Two or three dates are held annually. The activity takes place overnight and it is aimed at all ages. All participants to the event have a scientific authorization capture permit by the Department. The result is a great success of participation, more than 200 persons per event, a high disclosure in nearby villages and around 25000 crabs caught (mainly American signal crayfish) since 2010. A pioneering experience that is worth to be known.

O.3

GIS as Guarantee in Success? Searching tool limits in freshwater crayfish management.

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The historical distribution of the autochthonous freshwater crayfish (*Austropotamobius pallipes*) in Biscay (Basque Country) is very wide ranging, as it inhabited almost all the fluvial reaches. After the crayfish plague and the effects of many further threats, the species became very scarce and nowadays it is in extinction risk. Recent studies show that the distribution of *A. pallipes* is increasingly restricted, and apart from the presence of non indigenous crayfish species and their effects, the most important factors are those derived of habitat deterioration. Monitoring the species under these conditions of patchy distribution and wide range of factors affecting it implies an important scientific and economic effort.
Nowadays, the advances in the management of cartographic or statistical information using Geographic Information Systems (GIS), allow simultaneous processing of large amounts of data for many variables; featuring both great accuracy and an important economic advantage over traditional methods. This tool has also been used to predict the location of potential, adequate and sustainable habitat for the native crayfish. However many biotic and abiotic factors and their influence on the species are difficult to estimate. This way, inappropriate use, or inaccurate caution in the interpretation of data, may lead to not adequate outcomes. In this work the suitability of the use of GIS as the main tool for the management of native crayfish populations is discussed. The interpretation of results must be accompanied by a high level of knowledge of the species and its requirements and in any case, lots of field work.

O.4

Update of genetic information for the White-clawed crayfish in Spain

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The white-clawed crayfish is endemic to western and southern Europe and its populations have decreased over recent decades. Genetic information available about Spanish populations of this species usually comes from a single molecular marker.

In the present work, we used two mitochondrial markers (Cytochrome Oxidase subunit I and rDNA 16S genes) to examine levels and patterns of genetic structure across the range of distribution of the species in Spain.

Data reveal the existence of two main genetic groups of white – clawed crayfish in Spain with the Ebro Basin as a possible contact zone. Processes occurred in historical and recent times, such as genetic drift and translocations, contribute greatly to this pattern.

Levels of genetic variability and genetic structure of Spanish populations together with demographic inferences suggest that the species is established in the Iberian Peninsula, at least since the Late Pleistocene.

Knowing the true origin of the Spanish populations is crucial when deciding upon the management policies that should be followed. Given the lack of any clear evidence against its indigenous status, we propose that current protection and conservation measures should be maintained. From a management point of view, we suggest that Spanish population should be considered as a single ESU with two MUs corresponding with the genetic clusters detected in the present study.

O.5

Invasion of fluvial ecosystems by red swamp crayfish, *Procambarus clarkii*

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Biological invasions are one of the biggest threats to biodiversity and conservation of native species, playing a key role in global change. To try to control them, it is important to understand two types of phenomena: mechanisms that allow a species to successfully establish in a new environment, and the ecological, evolutionary and socio-economic consequences of an invasion. We will address both types of phenomena using a multidisciplinary approach and multi-scale research, employing as study model an invasive species of global importance, which has invaded many inland waters: the red swamp crayfish, *Procambarus clarkii*. In its native area, this species mainly lives in marshes; nevertheless, it has been able to invade a new environment, streams, in invaded areas (in our case: West Andalusia in South Western Spain). Since its introduction over 40 years ago, this invasion is causing serious damage to native species like the European freshwater crayfish, *Austropotamobius pallipes*. We have five main objectives in this study: (1) to compare the genetic diversity of invasive populations with the native area, identifying patterns of introduction, propagation and gene flow; (2) to determine what mechanisms (at the level of gene expression in different tissues) allow *P. clarkii* to adapt to new conditions or environmental stress: identify genes and loci responsible for local adaptation; (3) to explore the ecological effects of *P. clarkii* in streams, in terms of structure and ecosystem functioning, and interactions with populations of native crayfish (*A. pallipes*) and native amphibians and fishes. To do this, we will compare the ecological effects of stream populations (which have potentially evolved in response to the new environment) with those of founding populations from marshes; (4) to compare the ecological effects of *P. clarkii* with those of *A. pallipes* and determine whether the invasive species is occupying the same ecological niche as the native species which has displaced; and (5) to examine the prevalence of *Aphanomyces astaci* in the red swamp crayfish, given that this invasive species is a chronic carrier of the crayfish plague, *aphanomycosis*, causing the death of European freshwater crayfish; and that of the fungus that causes *chytridiomycosis* in amphibians. In this project, we expect to find mechanisms that allow the red swamp crayfish to successfully establish in a new environment and the consequences that this entails for native species.

O.6

Ethology of the invasive signal crayfish (*Pacifastacus leniusculus*) and its relationships with native fishes

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The signal crayfish *Pacifastacus leniusculus* (Dana, 1825) is one of the most successful crayfish invaders in Europe and it was firstly introduced to Sweden in 1959. This crayfish is endemic to the northwestern U.S.A. and southwestern Canada and it has negative effects on macrophytes, benthonic invertebrates, amphibians, reptiles and benthic fishes. The behaviour and effects of this nocturnal species on fish communities (in particular benthic fish species) and habitat are widely reported in experimental aquaria. However, few studies analyses the ethology and interspecific behavioural interactions and competition *in situ*. The goals of this study were to analyse the behaviour of *P. leniusculus* and to study its interactions with benthonic native fishes such as the stone loach (*Barbatula quignardi*). There are some studies suggesting that invasive crayfish compete for shelter with native fishes. Considering that major of these studies has been carried out in experimental aquaria, we decided to check if the same happen in natural ecosystems. This study was located in the Mediterranean rivers of the province of Navarra (Ebro Basin, Spain). A total of five sampling points with good abundances of signal crayfish and native benthonic fishes were studied. At each site, five recordings of four hours each were obtained. A total of 100 hours of video recordings were analyzed.

At each sampling point three underwater video cameras (GoPro4 modified for night vision) were mounted on an adjustable height, horizontal arm extending 1 meter of a set of iron. The height of the camera is set to just above the water the surface with the lens facing directly down toward the river bottom. Six infrared focuses are attached to the arm by adjustable extenders of 1.5 meters were used to illuminate the site during the evening and night in order to do not disrupt the normal behaviour. A battery was used to support lighting IR focuses. The videos of the three cameras gathered together with Adobe Premiere pro cs6. The program used to analyze the spatial positioning of crayfish and fishes was Tracker (Video Analysis and Modeling Tool). A digital multiparameter was used to measure *in situ* the physicochemical water variables (temperature, pH, conductivity and oxygen).

The main nocturnal behaviours of the signal crayfish (feeding, resting, aggressive interactions, home range, etc.) and some of the interactions with native fishes are analyzed and discussed. This research was funded by the Research Program of the University of Navarra (PIUNA 2014-15).

O.7

Assessing the crayfish ability to use anaerobic metabolism under microrefugia conditions

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Most of the freshwater invertebrates strongly depends on the quality of water in which they are living, the substrate also shaping their distribution. Ecologically, crayfish requires clean waters and stabile substrate in which they burrow gallery for shelters. Since crayfish tolerate the lack of oxygen by using anaerobic metabolism, we aimed to investigate if their sheltering behaviour is requiring the aforementioned use of anaerobic metabolism ability. For this objective, we analysed the oxygen requirements by simulating tubular shelters of different sizes. For this experiment we chosed two species of crayfish: *Astacus leptodactylus* and *Orconectes limosus* due to the reason that they are not protected. The crayfish were forced to live in submerged plastic tube in an aquarium simulating different water flow speed. Both, in the aquarium and tube we placed sensors for dissolved oxygen collecting the data regarding the saturation and temperature at the rate of 30 minutes for 24h in each experiment. Thus we computed the oxygen consumption of the crayfish inside its shelter. Using statistic analysis, we compared the data in different shelter conditions.

Our results showed that at least these two tested species of crayfish are able to produce low-hypoxic and even anaerobic conditions in tubular shelters longer than 40 cm. Consequently, our study highlight the importance of anaerobic metabolism as a legacy from its ancestral oceanic origin. This way rising a new question if the crayfish ectosymbionts or even pathogens are resistant at the lack of oxygen requiring further investigations.

O.8

Susceptibility of European native crayfish, *Austropotamobius pallipes* to the crayfish plague pathogen, *Aphanomyces astaci*: identification of a high resistant population

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Aphanomyces astaci is listed among the "100 of the World´s Worst Invasive Alien Species (http://www.issg.org/) and is responsible for the decline of the native crayfish species of Europe, and their endangered status. This pathogenic species is endemic of North America and only colonizes aquatic decapods. The North American crayfish species have a high resistance to this pathogen, while species from other regions are highly susceptible. However, recent field and laboratory observations indicate that they might exist some populations with higher degree of resistance. Thus, the objective of this study was to test the susceptibility of native European crayfish, *Austropotamobius pallipes*, by selecting 8 populations from the Pyreneans and challenging them towards the strain AP03 of *A. astaci* isolated the NorthAmerican red swamp crayfish, *Procambarus clarkii,* in the Garrotxa Natural Park (Girona). The results show that there are significant differences (P<0,001) among populations, although most of them show high mortality rates after exposure to *A. astaci* zoospores. However, one population from Girona exhibits a 100% survival during three months monitoring-period under the experimental conditions tested. Histological analyses revealed a high immune reaction in tissues examined, e.g., encapsulation and melanization of hyphae, similar to that found in NorthAmerican resistant species. These results represent the first observation of native European crayfish showing high resistance towards this pathogen. The identification of this population is of key importance for the management of these endangered species, and represents a crucial step forward towards the elucidation of the factors involved in the immune reaction against this devastating pathogen.

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P.1

Movements of the stone crayfish (*Austropotamobius torrentium*) in the Czech Republic: short-term movement patterns in small streams

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The stone crayfish *Austropotamobius torrentium* (Schrank, 1803) belongs to two indigenous crayfish species in the Czech Republic. It has the status of "critically endangered species" and is protected by national law (114/1992).

Within last few years, an intensive field research in the Czech Republic focusing on the stone crayfish has brought new knowledge about distribution, population densities, and ecological demands of this crayfish species. Nevertheless, only a few data dealing with movements and migration of this species were published.

This contribution describes short-term movement patterns in four small streams in the Pilsen region – one of the most important centres of distribution of this species in the Czech Republic. During 2013-2014, specimens of the stone crayfish were individually marked using VIE (visible implant elastomers) in 30m-long stream sections. Individuals were caught by hand, deposited in small plastic cups, marked and then carefully released under the same stone they were caught. The number of marked crayfish varied between 35 and 68 individuals. After one day, the recapture campaign was carried out on the same sites.

In this research, recapture ratios were very similar, varying between 13.2–17.6 % individuals. An estimation of population densities based on capture-recapture data varied on sites between 6.9-21.2 individuals (longer than 30 mm) per square meter. The recaptured individuals were divided into three categories: 1. non-migrating; 2. migrating up to 3 m along the stream; 3. migrating over 3 m along the stream. The first two categories created the majority of recaptured individuals (75-100 %) on all sites. In conclusion, the stone crayfish seems to be a sedentary crayfish species, as it is usually depicted.

P.2

Monitoring and implementation of the Recovery Plan of the White-clawed crayfish *(Austropotamobius pallipes)* in Aragon (Spain).

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OBJECTIVES: The main objective is to promote conservation actions necessary for halting and reversing the current process of regression of the species and ensure their long-term persistence in the Autonomous Community of Aragon (Spain).

STUDY AREA: SCOPE OF RECOVERY PLAN IN ARAGON. Decree 127/2006, of May 9 and extension of the recovery plan (Order of September 10, 2009) the Government of Aragon.

DESCRIPTION OF THE PROJECT: To conserve the genetic heritage of the specie, determining the populations of crayfish with an increased risk of disappearance and performing duplication compliance with its legislation in this regard (reintroduction plans).

DATA AND METHODS USED: Establishment of monitoring programs of populations and introductions. Take the necessary measures in epizootic processes. Update the general register of the crayfish populations. Update watershed states defining different areas, critical, sensitive, pending and null areas, and redefining strategies. Get a cartographic base with the latest results. Identify those parameters that determine the degree of development of the white-clawed crayfish, which guarantee the success of introductions. Improve understanding of the impact of the introduction of species of crayfish in sensitive aquatic species. Monitor the distribution and expansion of crayfish foreign species. Presentation of results, coordination and advisory groups involved in the conservation of the common river crab. Update information on the kind of research in Aragon and other communities.

RESULTS: Currently there are registered a total of 116 populations and 117 introductions. In Teruel there is registered a 63% of the total of populations, followed by Zaragoza (26%) and Huesca (11%).The main causes of disappearance of populations are: aphanomycosis (50%), unknown causes (36%) and water shortages (14%). Regarding Introductions are in the province of Teruel (75%), Zaragoza (15%) and Huesca (10%). Of all introductions made, a 67% have proved viable, and the causes that may explain the disappearance of introductions made have been established as follows: water scarcity (20%), fish placed in infectious process (15%), low number of individuals released (13%), posterior presence of red crab (13%), absence of barriers (10%), unknown causes (8%), floods (8%), uncontrolled discharges (5%), presence back signal crayfish (3%), ponds broken (3%) and aphanomycosis (2%).

P.3

Distribution and management of the stone crayfish (*Austroptamobius torrentium* Schrank) in the streams of the Nature Park Medvednica

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The stone crayfish (*Austropotamobius torrentium*) is one of the four native European crayfish species inhabiting Croatian freshwaters. It is considered as an endangered species, mainly due to the various anthropogenic impacts. This species is well adapted to lower water temperature and velocities typical for streams at higher altitudes. The aim of this study was to establish detailed distribution of the stone crayfish in the Nature Park (NP) Medvednica and to develop management plan of the species. Medvednica is a mountain located in the north-western part of Croatia. Nature Park encompasses approximately 75 waterbodies, scattered all over the mountain. Inventarisation of stone crayfish populations was conducted in 2010, with regular monitoring performed in 2012 and 2015. In 2010 altogether 71 locations on 52 streams were investigated by trapping and hand search, in 2012 and 2015 additional 10 new streams and tributaries were monitored as well. Sites included those in pristine state as well as heavily modified or polluted lower courses of certain streams. At each site, we performed a detailed characterization of each habitat and recorded basic physical and chemical parameters. The results of this extensive study show that 57 % of streams (43 streams) in NP Medvednica were inhabited by crayfish. There were no crayfish in polluted lower courses of streams located in inhabited parts of the NP. Among streams which were modified by minor hydrotechnical interventions (53 % of all investigated streams) 68 % were inhabited by stone crayfish. Results of this study were used for stone crayfish management plan in public institution NP Medvednica. Protection measures began in 2010 and included waste removal from the stream bed, educational activities related to the stone crayfish, like educational tables alongside educational trails, educational leaflets and workshops for stakeholders.

P.4

Could electric fish barriers help to manage native populations of European crayfish threatened by crayfish plague (*Aphanomyces astaci*)?

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European native crayfish species are threatened and their populations are declining due to habitat loss, water pollution, and the effects of competition and diseases from non-indigenous crayfish species. One of the main conservation problems is the crayfish plague caused by the oomycete *Aphanomyces astaci*. The pathogen lives in the cuticle of crayfish, and it is spread by asexual swimming zoospores, which serve as infective units. Every year, dozens of native crayfish populations disappear due to this disease. We decided to use an electric fish barrier during an event of crayfish plague to increase the discontinuity of the river to avoid the progression of individual infected crayfish upstream. The main objective of this poster is to transfer our expertise using this equipment for conservation purposes. As a result, we report a detailed description of the experience, as well as requirements, problems and opportunities of using an electric fish barrier to try to control crayfish plague *in-situ*.

P.5

First population of the spiny-cheek crayfish *Orconectes limosus* (Rafinesque, 1817) introduced to the Iberian Peninsula

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The spiny-cheek crayfish, *Orconectes limosus* (Rafinesque, 1817), is a crayfish native of North America, widely distributed on the Atlantic watersheds. The spiny-cheek crayfish was the first non-native crayfish to be intentionally introduced into Europe from the United States, in 1890.

This is the first record of the spiny-cheek crayfish, *Orconectes limosus* (Rafinesque, 1817), introduced into the Iberian Peninsula. Sixteen individuals of this crayfish species were detected in the Muga River, near to the Boadella Reservoir (Catalonia, NE Iberian Peninsula) on October 9, 2010. This reservoir is only 14 km away from France, where the spiny-cheek crayfish was introduced during the 20th century. The presence of this non-native crayfish is a pressure on the conservation of native crayfish populations of this area and a problem for the whole aquatic ecosystem. On this poster we expose new information of this population state, thanks to recent work within the Life Potamofauna.

P.6

LIFE Potamo Fauna, a project for the recovery and conservation of endangered river fauna in the basins of the Ter, Fluvià and Muga rivers (Catalonia)

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In January 2014 has started Potamo Fauna LIFE project: "Conservation of river fauna of European interest in the Natura 2000 Network of the basins of the Ter, Muga and Fluvia rivers" (LIFE12 NAT/ES/001091). With a total budget of 1.9 million euros, involve six partners (Consorci de l'Estany, Consorci del Ter, Generalitat de Catalunya, Associació d'Amics de la Tortuga de l'Albera, Forestal Catalana y Universidad del País Vasco) and 6 cofinancers, apart from the European Union, which provides 50% of the overall budget. The overall objective of this project is the recovery and long-term preservation 12 endangered native species of aquatic fauna, including 3 species endangered in Catalonia and Spain, through a wide range of measures: captive breeding, population reinforcements, habitat improvement, control of exotic species, and dissemination and research on the status of these species and the value of river and lake systems.

Among the planned conservation action, the main lines of action planned are:

- Conservation and recovery of riverine populations of three threatened species of aquatic fauna, mainly with specimens coming from captive breeding centers: *Unio elongatulus*, the native crayfish (*Austropotamobius pallipes*) and European Pond Turtle (*Emys orbicularis*).
- Conservation and recovery of populations of three endangered species of aquatic fauna, mainly through translocations of individuals coming from healthy populations into each basin: *Vertigo moulinsiana* and *V. angustior*, and Mediterranean Barbel (*Barbus meridionalis*).
- Improvement of populations of an aquatic turtle and of 5 amphibians in the Ter river, by creating micro wetlands: Mediterranean Turtle (*Mauremys leprosa*), Marbled Newt (*Triturus marmoratus*), Common Midwife Toad (*Alytes obstetricans*), Western Spadefoot (*Pelobates cultripes*), Natterjack Toad (*Bufo calamita*) y Mediterranean Tree Frog (*Hyla meridionalis*)
- Fight against various invasive alien species of crabs, fish, and freshwater mollusks, to mitigate its negative effects on aquatic fauna and their habitats, through a battery of different actions: population control in specific sectors, experiments against aphanomycosis, prevention of penetration, and other.

Plans and milestones of this project are presented.

P.7

Effect of invasive alien fish control over the population of red swamp crayfish (*Procambarus clarkii*) in Lake Banyoles.

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The red swamp crayfish (*Procambarus clarkii*) was mentioned on Lake Banyoles for the first time in 1982. It is especially abundant in irrigation channels, streams and ponds of the lake surroundings. The presence of native crayfish in its waters is not known. Their presence and abundance in Banyoles lake is determined by the abundance of alien predatory fish, especially largemouth bass (*Micropterus salmoides*), in which the red swamp crayfish diet plays a major role.

In 2010 a LIFE project entitled "A Demonstration Project Improvement of habitats and species of the Natura 2000 network of Banyoles" (LIFE08 NAT/E/000078) was started. Its main objective was to carry out a widespread intervention to fight, slow down and reverse the decline of species and habitats of community interest being caused by invasive species. Controlling alien fish species were necessary for the recovery of native species like Mediterranean barbell (*Barbus meridionalis*) and native freshwater mussels. These actions have been conducted mainly by electric fishing from a boat, trammel nets, traps and even water management in the lagoons around. The evolution of the red swamp crayfish population has been used as an indicator of the evolution of these control tasks. An increase of crayfish in the main lake is expected, while the largemouth bass populations were declining. The elimination of red swamp crayfish is not the aim of this project, although it's one of the most damaging invasive species on the planet, because it is considered impossible to eradicate of the lake. In 2014 a new project LIFE "Potamo Fauna" (LIFE12 NAT/ES/001091) reinforces the experimental control actions over alien predatory fish so it has continued the monitoring that began in 2010. We have used a standardized methodology for monitoring species at long term. Two campaigns a year, in summer and autumn, have been made, based on the sampling of 21 points on the lake and surroundings. At each point five traps have been placed during 24 hours baited with chicken liver. The population densities of crayfish on Lake Banyoles at the beginning of the project were very low (0.03 to 0.5 CPUE crabs per day and trap) and these increased to higher values (1.07 to 1.62 CPUE) in 2012 and 2013 with the implementation of measures of alien fish control. However, they remained below the density values of the locations of the lake surroundings (2.67 to 5.36 CPUE). A significant increase was also observed in crayfish in Vilar lake from the autumn 2011campaign. A significant increase in crayfish in Vilar lake was also observed after the autumn campaign of 2011. Catches of crayfish maintain a ratio of males to females close to 1 in the summer campaigns, while in the autumn campaign fewer males were captured. There is also an increase in catches of individuals of more than 100mm in the lake which is consistent with the general trend. We caught crabs until a depth of 17 meters, although most were captured between 0 and 2 meters. The evolution of red swamp crayfish density can be used as an indirect indicator of the density of alien predatory fish in the lake.

P.8

Mediterranean rivers with low hydromorphological impacts constitute a refuge for native fish and amphibians, in front expansion of exotic aquatic species; the case of several basins in northeast Catalonia.

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Several fish surveys in some Mediterranean rivers in northeast Catalonia have been done from 2006 to 2011. These rivers include a wide range of hydromorphological situations, from pristine status to highly degraded situations with hard modifications of flow regime, river bed, riparian forest and even the presence of artificial barriers. Over 300 stations have been sampled, all along 7 hydrologic basins. Fish surveys were based on passive capture techniques. In each sampling station between 3 and 8 small fyke-nets were left on the river along a day, at least, to estimate relative density (CPUE). This capture technique has been useful to detect other species of aquatic fauna, mainly herpetofauna, and great crustaceans. All the amphibians and great crustaceans potentially present have been captured.

In the surveyed rivers 7 native freshwater fish are present, and till 11 species of amphibians can appear on these river habitats. On the other hand, a large number of exotic fish and other aquatic fauna, including red swamp crayfish (*Procambarus clarkii*), have been established in the area, and have appeared on surveys.

Non altered Mediterranean rivers, with a low or none hydromorphological impacts, are the principal refuge for all these native species, where on the other hand the presence of exotic species is generally very low. These rivers have orders between 3 and 4, mainly. On the lowland plains, in natural conditions, this kind of rivers are typically intermittent during summer, when most of the river bed is dry and the only refuge for fish are isolated pools. In contrast, most of the principal fluvial axis of the area (orders above 4), with high degree of hydromorphological transformations, are intensively invaded with exotic species, native species are absent or scarce, both fish and amphibians.

In this context, only some well preserved Mediterranean rivers arise as refuges for native species in front the progressive establishment and expansion of exotics in impacted fluvial rivers. Unfortunately, these refuges constitute isolated river sections in the context of basins widely modified.

P.9

Biological control of red swamp crayfish (*Procambarus clarkii*) by Mediterranean pond turtle (*Mauremys leprosa*)

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Since 2008 the Government of Catalonia has been working on the control of red swamp crayfish (*Procambarus clarkii*) populations in sensitive areas in contact with European native freshwater crayfish populations (*Austropotamubius pallipes*), in different basins in southern Catalonia.
The removing of these populations turned effective for a while, but the rapid recovery of the invasive species made any control measure ineffective if these actions were not taken periodically throughout the life cycle of the species.

On the other hand, parallel sampling work on Mediterranean pond turtle made in the same watersheds since 2013, showed a clear relationship between the abundance of the two species. In the stretches where the turtle had higher population abundances, the presence of invasive crab was by contrast scarce or even nonexistent, and vice-versa; where the turtle was absent or its presence was anecdotal, crab abundances soared significantly.

Samplings obtained in 24 locations in the 6 major basins of southern Catalonia (Anoia, Foix, Gaia, Francolí, Siurana-Montsant and Algars) demonstrate the significance and correlation of the abundance of the two species, so we can conclude that the conservation and recovery of pond turtle populations would be a very effective control measure against the spread of the invasive species.

P.10

Management of the native White-clawed crayfish (*Austropotamobius pallipes*) in the regions within the province of Girona

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The native white-clawed crayfish (*Austropotamobius pallipes*) is a species that was commonly found in most watercourses in the Upper Fluvià River Basin in the La Garrotxa Region until the end of the 1970s, when the crayfish plague *Aphanomyces astaci* arrived. The native white-clawed crayfish populations are currently found in first or second-order seasonal rivers and fast-flowing streams where, due to some kind of natural barrier (droughts in the lower reaches of rivers, cliffs, etc.), the fungus was unable to proliferate. Moreover, at the end of the 1990s, the red swamp crayfish (*Procambarus clarkii*) population expanded rapidly and the situation became extremely complicated.

In the Fluvià River Basin, white-clawed crayfish populations have been monitored since 1992. In the first year of the study, a total of 24 populations were located and, five years later, this number had decreased by 50%. After this, sampling intensified and new populations were found. However, studies also revealed that half of these populations disappeared after a few years.

In 2004, a recovery plan for the species was launched in the Volcanic Region of La Garrotxa Natural Park. The aim of this plan was to recover the species, so as to re-establish the balance of the ecosystem it occupied in suitable sectors of the water network. The activities planned were: the localisation and health assessment of all the populations, control of the crayfish plague, reinforcement of the populations, and reintroductions based on a breeding site or by translocation. These activities were included in a work plan with the following objectives:

Objective 1: To avoid the basin being invaded by non-native crayfish species.

Objective 2: To favour the white-clawed crayfish's colonisation of the areas described as crayfish zones.

Objective 3: To establish a periodical control system, allowing problems specific to each population to be detected, as well as any crayfish plague outbreaks.

Objective 4: To promote the development of lines of research applied to management, allowing for the redefinition of white-clawed crayfish conservation strategies to be adopted in the future.

Objective 5: To set up educational programmes, which promote respect for the native species and raise awareness of the crayfish plague.

P.11

Management of the White-clawed crayfish breeding center in the Volcanic region of la Garrotxa Natural Park (Girona)

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The recovery plan for the native white-clawed crayfish (*Austropotamobius pallipes*) in the Fluvià River Basin included a series of actions, with specific objectives aimed at ensuring the conservation of this species. In order to achieve the following objectives: 2: *To favour the white-clawed crayfish's colonisation of the areas described as crayfish zones,* objective 4: *To promote the development of lines of research applied to management, allowing for the redefinition of white-clawed crayfish conservation strategies to be adopted in the future*, and objective 5: *To set up educational programmes, which promote respect for the native species and raise awareness of the crayfish plague*, it was necessary to create a captive breeding centre for this species.

Therefore, in 2004 the white-clawed crayfish farm was created in the Upper Fluvià River Basin and in the heart of the Volcanic Region of La Garrotxa Natural Park.

At first, there were several issues with the crayfish farm facilities that made them unsuitable for breeding this species, since they had been designed to be used as a fish farm for brown trout. Thus, certain adaptations had to be made in order to breed crayfish in captivity.

The initial idea was to divide up the various ponds in order to keep the breeding individuals, those to be reared and the juveniles in their first year separate, thereby facilitating handling and decreasing losses resulting from cannibalism among different sized crayfish.

Individuals from different water basins were also placed in separate ponds, in order to study the genetics of each population.

Since then, the Natural Park crayfish farm has allowed for the captive breeding of crayfish from different water basins across Catalonia and, since 2008 repopulations have ben carried out in several streams in river basins in northeast Catalonia.

At present, thanks to support from the LIFE Potamo Fauna Project, improvements have been made to the crayfish farm, in particular with regard to the controlled conditions required to help monitor the species' breeding cycle better and improved health research

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