

# PROGETTO LIFE IP IMAGINE – ESTRATTO AZIONI PER AMBIENTI ACQUATICI

## ***ACTION A.9: Design of conservation interventions and action plans for aquatic ecosystems***

*Beneficiary responsible for implementation*

DCBB

This action contributes to the implementation of measure G.1.d (Pg. 63) of the PAF.

### *Description*

The action will begin in the first phase of the project and will end in the second phase.

The action involves the design of some conservation interventions, which will be carried out through the action C12, on the basis of cognitive insights related to both the biological habitats of species of cyclostomes, bonefish, crustaceans and aquatic insects of Annex II and IV (DH) (*Lampetra planeri*, *Lampetra zanandreae*, *Squalius lucumonis*, *Salmo cettii*, *Barbus tyberinus*, *Barbus plebejus*, *Cobitis billineata*, *Telestes muticellus*, *Sarmarutilus rubilio*, *Padogobius nigricans*, *Cottus gobio*, *Austropotamobius pallipes*, *Coenagrion mercuriale* coinciding with the habitats of Annex I. (DH), both to some particularly threatened habitats of Annex I.

The main objective of the interventions will be to reverse the current trend of loss of biodiversity and naturalness of the wetlands of Umbria, highlighted by a high rarefaction of species and habitats of Community interest, once abundant but currently confined in few small sites as a consequence of the cessation of traditional management practices, the impacts of some invasive alien species and ongoing and increasingly evident climate changes.

For a better description of the action, the planned activities are developed into sub-actions.

### ***A9.1 Cognitive insights and definition of rare habitat conservation interventions for the regional territory (Habitats 7140 new, 7210 \*, 7220 \*, 7230)***

The action, scheduled for the first and second year of the project, has the main objective of planning active interventions for the maintenance and restoration of rare habitats as they are present in very few stations in the regional territory. Through the preparatory action, the presence stations of the habitats in question will be investigated, both where they are present, and within potential sites, creating a precise picture of the occupied surfaces, any pressures in progress and therefore the state of conservation. These are the habitats: 7210 \*, 7220 \*, 7230, and 7140 (newly discovered for the Umbrian territory).

**The 7210\* habitat** is present exclusively along the shores of Lake Piediluco (TR) (the only station in Umbria), within the SAC IT5220018 and SPA IT5220026 sites. It is a habitat that, even in the absence of external perturbations, is subject to a natural dynamism that leads to a progressive replacement of the hygrophilous vegetation with a more mesophilic one and to an evolution towards aspects with woody plants (shrubby and / or arboreal). Therefore, the need to carry out maintenance and, where necessary, habitat restoration was highlighted in the PAF. In this action all habitat presence stations will be investigated and mapped (phytosociological surveys and GIS restitution) with indication of the area occupied (m<sup>2</sup>), along the entire perimeter of Lake Piediluco (about 14 km), as well as the possible presence of the habitat in the form of floating mats inside the lake mirror; the conservation status and any pressure factors will be reported for each point of presence detected. In the areas deemed suitable, restoration and / or expansion of the habitat will be defined, for the definition of which the experiences gained within LIFE11 / NAT / IT000187 TEN will be taken into consideration. The possible sources of pressure detected in the areas adjacent to those of intervention, providing long-term mitigation solutions to be implemented with concrete actions action C12.

Although **the 7220\* habitat** is extremely localized and reported in only three sites (SAC IT5210049, SAC IT5220017 and SPA IT5220025), it is in a good state of conservation. With this action, the potential sites where the habitat could be present will be investigated, through a precise mapping of the presence stations. The survey will be completed by surveys on the water regime of all the stations of the habitat since it is affected by its variations and therefore by any water uses.

**Habitat 7230** is present only in the swamp of Colfiorito (PG) site SAC / SPA IT5210072, (the only station in Umbria). These are habitats of great naturalistic value which, even in the absence of external perturbations, are subject to a natural dynamism with the progressive replacement of the hygrophilous vegetation with a more mesophilic one. The natural dynamics of vegetation towards shrubs or bushes are activated with great ease and the habitat is able to self-preserve only in the presence of high water level or good practices that allow its constant regeneration. In many cases the traditional uses of mowing and the cutting woody species have played a fundamental role among the regenerating and conservation actions. The habitat is also affected by the pressures of agricultural uses, has a high index of contrast with the surrounding areas, rarefaction phenomena and consequent worsening of the structure and function parameters. The habitat is in a poor state of conservation and in severe deterioration, highlighted by the presence of dynamic processes with shrub invasion and floristic depletion, caused by the synanthropic species that reduce the phytocenotic quality. The action will therefore aim to define the conservation and restoration interventions of habitat 7230 also through in-depth analyzes on: the state of conservation of the habitat according to the water regime of the area; identification and mapping of the presence of drainage channels that convey water inside the swamp; presence and consistency of vegetation evolution phenomena. The survey will concern the area within which the presence of the habitat is reported (approximately 2,5 ha) and the neighboring areas for a total of approximately 4 ha, so as to detect any pressure to be faced in defining the interventions that will be carried out with action C12. The definition of conservation interventions will be carried out starting from the experiences gained within LIFE11 / NAT / IT000187 T.E.N. and will concern interventions of hydraulic reorganization of the area, operations of removal of the arboreal and shrubby components, long-term mowing of the meadow crops.

**Habitat 7140** was recently found in Umbria, in the territory of the Sibillini Mountains National Park within the Natura 2000 site SAC/SPA IT5210071. It is a relict population of depleted acidophilic mires, outside its main northern range. The habitat has a significant biogeographical importance in the Apennine context since they represent the southern limit of these vegetations types of continental and alpine biogeographic areas. These phytocoenoses come out of the typology of high active sphagnum population; they cannot be considered ombrophylous in the strict sense because the ecological-environmental conditions in which they develop depends of the water table; moreover, for macroclimatic reasons, high levels of water stress are reached during the summer period. Finally, for biogeographical reasons, there is a strong impoverishment of the characteristic floristic composition: in fact, these plant communities survive as fragments of very small size, isolated and relict, within various systems of humid environments such as the karst plains and do not have a significant number of endemic or southern vicariant species. Given the importance that the habitat acquires, precisely because it is located outside the biogeographical context of belonging, since it contributes to the understanding of the floristic history and biodiversity of these areas, it deserves particular attention. Therefore, with this action, in-depth analyzes will be carried out to define an appropriate management strategy for the territorial area in which the habitat was found, with definition of the state of conservation and therefore, the identification of pressures and threats.

Finally in accordance with the interpretative manual of Italian habitats The residual populations of depleted acidophilic sphagnum mires, outside their main range, represented by a few species and not attributable to the type of high peat bogs, refer to Habitat 7140 'Transitional and unstable bogs'. so many authors prefer referring to the *Caricion nigrae* alliance. They are strongly conditional on unstable ecological factors (aridity, strong influence of the surface aquifer, anthropic influence); nevertheless, but they have a significant biogeographical importance in the Apennine context and deserve appropriate recognition.

The actions described will be carried out by the DCBB, which will take care of the technical-scientific part, in collaboration with AFOR, which will guarantee the construction of the structures and the specialized workforce for the implementation of the interventions.

### **A9.2 Development of Action Plans to improve the conservation state of temporary ponds (Annex I Habitats 3130 and 3170\*)**

This sub-action is focused on the development of Action Plans for two point-type Annex I Habitats with a very restricted distribution in Umbria, although with a high conservation value. The activities will include a complete and updated overview of the distribution, structural and floristic composition of H3130 (only the valuable sub-type with *Juncus bulbosus*) and H3170\* in the main sites of occurrence, with particular focus on the ZSC IT5210020 and IT5210013 which host the most valuable expressions of these two habitats. The other sites will be surveyed and an updated distribution will be drafted. Detailed distribution maps of the ponds will be drafted in the main sites of occurrence, at the scale 1:1.000. Based on the current conservation status, as resulting from the 4th Habitat Report ex-art. 17 updated by the *ad hoc* field surveys, and taking into account the pressures negatively affecting the conservation of these habitats (mostly represented by successional processes and nutrient accumulation), concrete actions to improve their conditions will be drafted and planned. The Action Plans will take into account the need of contrasting i) the shrub/tree invasion, ii) the accumulation of leaf-litter in the ponds (mainly deriving from long-lasting oak leaves) and iii) the invasion of the alien invasive moss *Campylopus introflexus*, recently documented. The guidelines in the Action Plans will start from the outcomes and management addresses developed by SUNLFE.

This sub-action will be implemented in the first phase of the project (years I and II).

### **A9.3 Development of an environmental remediation plan for the hydrological network**

In the second phase (third year), an environmental recovery plan for the river network will be drawn up on the basis of the results of the following studies that will be carried out during the first phase of the project:

#### **Collection of historical series of rainfall-thermal-hydrometric data and analysis of trends in water temperature, rainfall and flow rates**

The surveys will be carried out on all the rainfall, thermal and hydrological data collection stations available in Umbria (about 140) managed by the Regional Hydrographic Service. For each survey station, statistical analyses of the data series will be performed to analyse their trends and interpret their evolution over time. The action will be essential to recognize the most critical areas for water shortages and the absence of hydraulic continuity and to identify the most important physical obstacles (weirs, dams) that limit the movement of fish fauna. The presence of barriers is a particularly serious issue, since following the rise in temperature, most fish species will try to pursue their thermal optimum by moving upstream along the longitudinal gradient and that in this they will be limited by the presence of physical, hydrological or biological (alien species) obstacles.

#### **Analysis of summer flows in the hydrological network of Umbria Region and verification of the ecological flow**

Summer flow measurements will be carried out on 46 monitoring stations located on the waterways within the Umbrian RN2K. For each sampling station, the flow rate will be calculated in compliance with the ISO 1088 standard. The surveys will be carried out once a year for two years in each station in the lean period to highlight the critical issues related to the drought span and the data will be compared with the Minimum Ecological Flow (DMV) established by the Water Protection Plan (PTA).

#### **Analysis of environmental suitability in Umbrian waterways for aquatic species**

As a part of the action, an app for mobile devices will be developed and distributed to the main users of river systems (canoeists, anglers, hikers, etc ...) who will be able to report the obstacles that interrupt the river continuity in Umbria. When necessary, the information will be completed by surveys in the areas not covered by the census (max 15 travel). All the data collected in the activities a, b and c will be used to develop a GIS predicting scenarios related to climate change, including environmental suitability and variations in the distribution of aquatic species. These scenarios will be used in action C20 both to prioritize the obstacles to be removed (i.e those that prevent the movement of the aquatic fauna) and to identify the obstacles that, on the contrary, are important in limiting the expansion of invasive species.

Through the data collected in the activities a, b and c the most critical areas worth of a priority intervention will be identified. The intervention must provide an ecosystemic and integrated

approach between the Habitats Directive (DH) and the Water Framework Directive 2000/60 (DQA). The integration between the two directives, already considered by the articles 4.1c, 8.1, 11 of the same WFD, optimizes the use of the available data, increases knowledge, allows to choose the appropriate scale of intervention and to evaluate the effectiveness of the measures, optimizing the use of the economic resources.

#### ***A9.4 Proposal for a Regional Plan for the Adaptation of Aquatic Ecosystems to Climate Change***

The analyses of the Intergovernmental Panel on Climate Change (IPCC) have highlighted how human activities are changing the climate framework with unequivocal global warming. The effects of warming will be particularly negative on aquatic ecosystems in the Mediterranean area. For this reason, it is necessary to make an effort in planning adaptation measures to climate change, preventing and mitigating their future effects also at a regional scale, to be included in the wider context of environmental protection policies. In the fourth year of the project (second phase), the results of actions A9.1-A9.3 will be presented and discussed with the involvement of national authorities and stakeholders (Central Apennine District Authority, ARPA, environmental, scientific and sport fishing associations), in order to start a participatory process for the realization of a Regional Plan for the Adaptation of Aquatic Ecosystems to Climate Change (E.4)

The data acquired in action A9 will be used to develop the Recovery Plan for Umbrian river network and the Regional Plan for the Adaptation of Aquatic Ecosystems to Climate Change envisaged by the project. Moreover, thanks to this information it will be possible to improve the conservation status of fish species and aquatic habitats and to contribute to the implementation of integrated management of regional water resources in all levels of Regional planning. Data acquired within the action A9 are also preliminary to the restoration actions (considered in the Actions C12 and C19), to the defragmentation of aquatic ecosystems and promotion of ecological connections (Action C21). Thanks the action A.9, it will also be possible to define the Action Plans for Aquatic species (Action A12) and to specify the areas in which the control or eradication actions of the invasive species envisaged in the action C13 will have a greater chance of success.

During the action, the need of eventual permits and authorizations will be verified. In case of need, the authorizations and permits will be asked to the relevant Authority in order to carry out the correspondent C actions.

#### ***Reasons why this action is necessary***

The action is necessary because it allows i) to improve the conservation status of fish species and aquatic habitats, ii) to contribute to the implementation of integrated management of regional water resources in all levels of planning, iii) to implement restoration actions (Actions C12 and C19) and defragmentation of aquatic ecosystems and promoting ecological connections (Action C21). Thanks to this action it will also be possible to define the Action Plans for Aquatic species (Action A12).

A GIS predicting scenarios related to climate change will be realised, including environmental suitability and future variations in the distribution of aquatic species. This information will be used to predict the movements of fish fauna according to climatic variations and to establish which interruptions to river continuity must be primarily removed to facilitate such movements. Initially, the action didn't consider courses or educational activities related to the use of GIS, but they can be envisaged, if necessary. The results of the scenarios generated by the GIS will be available on the project website and on the Umbria Region website and they can contribute to the implementation of integrated management of regional water resources in all levels of Regional planning.

#### ***Constraints and assumptions***

No particular problems are expected in the implementation of the action, given the DCBB and DSA3's expertise in these sectors.

### *Expected results*

Habitat 7210\*: mapping of presence stations along the 14 km of Lake Piediluco shore, with census attached (number and size m<sup>2</sup>) in the body of water; pressure mapping; definition of interventions of restoration and expansion of the habitat surfaces for about 1000 m<sup>2</sup>

Habitat 7220\*: definition of an updated picture of the habitat presence stations (GIS georeferenced mapping); indication of the current water regime of the stations surveyed; evaluation and possible definition of interventions to modify the water regime where necessary.

Habitat 7230: definition of the water structure of the area where the habitat is present (survey on 4 ha); possible planning of interventions to adapt the network of canals that convey the waters to the swamp; Definition of interventions for the removal of the tree and shrub component and subsequent multi-year mowing plan, on approximately 2.5 ha.

Habitat 7140: definition of the conservation status and identification of a conservation strategy.

Through this action it will be possible to i) identify the most critical areas in the hydrological network of the N2K from the point of view of the quality and quantity of water, ii) realize suitability maps for all aquatic species, iii) predict future climate trends by implementing a Plan to mitigate the most negative effects. Complete floristic/structural/distributional characterization, over an area of about 4 ha corresponding to the most representative areas of occurrence of the two habitat types.

### *Cost estimation:*

#### **174.936 €**

119.642 € Personnel costs: they are calculated on the basis of the data collected by the national collective agreements of reference and refer to the personnel currently employed by the partner organizations, as well as to the human resources that will be used specifically for the project. The amounts have been calculated by multiplying the number of man-days foreseen for the relative daily cost.

The costs scheduled for the external GIS Expert (DCBB) were calculated by means of a market research, consulting specialized professionals in the sector.

13.094 € The travels to the sites of interventions were calculated on the basis of the cost of the means of transport and the daily allowances.

33.000 € For external assistance costs, an assessment was made taking into account the average market prices by consulting specialized subjects in the sector.

6.800 € to purchase equipment needed to implement the action.

For all other costs, an assessment was made taking into account the average market prices and by consulting specialized subjects in the sector.

### *Deliverables:*

Action Plans for the Annex I Habitats 3130 and 3170\* 30/09/2022 Distribution maps of H3130 and H3170\* at the scale 1:1.000 in the most representative areas 30/09/2022

Report on the climate trends of the Region 30/09/2022

Summer flow maps and verification of the DMV of 50 sampling stations in the waterways of the N2K 30/09/2022

Conservation improvement project Habitat 7210\* 30/09/2022

Conservation improvement project Habitat 7230 30/09/2022

### *Milestones:*

Request of permits and authorizations (if needed) 30/09/2022

## **ACTION A.12: Drafting action plans for species and planning of conservation interventions**

*Beneficiary responsible for implementation:*

DCBB

This action contributes to the implementation of measure G.1.a (Pg. 50-56) of the PAF.

*Description:*

The action aims to prepare Action Plans to improve the conservation status of some of the most threatened animal species among those present in Umbria listed in the Annex II and IV (DH): *Lampetra planeri*, *Lampetra zanandreaei*, *Squalius lucumonis*, *Salmo cettii*, *Barbus tyberinus*, *Barbus plebejus*, *Cobitis billineata*, *Telestes muticellus*, *Sarmarutilus rubilio*, *Padogobius nigricans*, *Cottus gobio*, *Austropotamobius pallipes*, *Bombina pachypus*, *Triturus carnifex*, *Emys orbicularis*, *Vipera ursinii*, all Chiroptera species. Furthermore, some of the interventions provided in the Plans will be designed for their execution under concrete conservation actions (C13, C16, C17, C18, C19 and C20) foreseen by the project.

In details, in addition to be needed for the Drafting action plans for aquatic animal species, the data acquired with action A12 are essential for achieving the objectives of conservation actions C18.1 (*Salmo cettii*) and C18.2 (*Padogobius nigricans*). Together with the information collected through action A.13, these data will also be used for identification of the sites in which carry out conservation interventions to be implemented with Actions C19 and C20 as a priority, for example for identifying the sites where river connectivity restoration interventions are carried out (Action C20), restoring otherwise isolated native fish populations and avoiding facilitating the propagation of invasive species.

For habitats, the collection data in the initial phases is fundamental since the conservation actions expected fall in aquatic environments which have as their intrinsic condition that they be extremely rapid in their evolution and subject to very large annual fluctuations. Being habitats (Habitats 7140 new, 7210\*, 7220\*, 7230, 3130, 3170\*) very rare and extremely localized, it is necessary to promptly define their structure, size, specific composition. An investigation supplement is then necessary for habitat 7140 for the first time reported in the Region.

### **A12.2 Drafting action plans for aquatic animal species and planning of conservation interventions**

The sub-action foresees to carry out the preparatory and preliminary activities for actions C13, C18, C19 and C20, and allows to gather the necessary information to coordinate future conservation interventions for all species of crustaceans, cyclostomes and bone fish listed in DH occurring in Umbria (*Lampetra planeri*, *Lampetra zanandreaei*, *Squalius lucumonis*, *Salmo cettii*, *Barbus tyberinus*, *Barbus plebejus*, *Cobitis billineata*, *Telestes muticellus*, *Sarmarutilus rubilio*, *Padogobius nigricans*, *Cottus gobio*, *Austropotamobius pallipes*).

For each species, a regional Action Plan will be defined containing the following elements:

- general framework, biology and ecology of the species and their conservation status;
- distribution and abundance of populations;
- regional, national, European and international regulatory framework;
- techniques and methods of population monitoring;
- identification of threats and limiting factors with particular reference to the presence and dissemination of IAS;
- identification of general and specific objectives;
- identification of action priorities;
- identification of the sites in which to carry out conservation interventions to be implemented with Actions C18, C19 and C20 as a priority;
- information and awareness strategies for the community and public administrations.

In order to investigate the composition of the fish assemblages, and the abundance and age structure of fish populations that make up the aquatic communities of the RN2K, quantitative samplings will be carried out in 52 aquatic environments of the Region, in addition to the data already available. The samplings will be conducted by DCBB and RU in autumn through electrofishing using the removal method. The abundance of populations of each species will be estimated using the two-pass removal method. For each population, the age structure will be determined (density ind/m<sup>2</sup> and standing crop gr/m<sup>2</sup> for each age group). Age determination and abundance estimation will be performed by DCBB on the basis of the data and the scales collected from each individual caught.

In addition to biological data, some information on the physical, chemical and hydrological characteristics of the sampled sites will be collected. The most important abiotic factors will include: water temperature, pH, dissolved oxygen, specific electrical conductivity, average current speed, geometry of the wet section, instant flow, substrate composition, cover, ecc...

In this step, the action involves also an initial screening of the genetic and demographic characteristics of *Salmo trutta* complex populations occurring in the Umbrian N2K network, according to the same methods tested during the LIFE12 NAT/IT/000940 (Life TROTA) and integrating with the information collected in the LIFE18 NAT/IT/000931 (Life Streams). Using nuclear and mitochondrial markers, pure or almost pure population will be selected and the wild breeders collected. These specimens will be used for breeding and artificial fertilization for the production of juvenile stages (eggs and fry) destined to supportive inbreeding for partially pure populations and to the reintroduction in the sites in which Atlantic trout has been eradicated. The level of introgression of the Mediterranean trout populations will be assessed. The samples for the genetic analyses using both nuclear and mitochondrial DNA (mtDNA) markers will be sent to a specialized laboratory. The genetic variability at mtDNA level will be analysed using polymerase chain reaction-single-strand conformation polymorphism (PCR-SSCP) and sequencing. In particular, sequencing will be performed on the complete Control Region (D-loop) of those individuals which will have exhibited SSCP polymorphism. This method will allow the detection of the mtDNA evolutionary lineages native and alien to the study area. The frequency of alien haplotypes will be used to estimate the level of introgression at mtDNA level. The presence of alien genes will be also estimated using the nuclear locus LDH-C1\*. The estimates of introgression values will be used to monitor the effect of conservation actions during the LIFE activities (actions D1). A total of 750 specimens (25 sampling sites x 30 specimens) will be analysed using the above molecular markers. A microsatellite DNA analysis will be also carried out on the populations for which mtDNA and LDH-C1\* analyses will have shown a high degree of genetic purity (introgression values with both mtDNA and LDH-C1\* locus  $\leq 20\%$ ). At least fifteen loci will be used to ensure high resolution power during the statistical treatment of the data.

Always in this step the action will provide an initial screening of the demographic characteristics of the populations of *P. nigricans* and *P. bonelli* in the Umbrian N2K network

to identify the areas where the two species cohabit and the sites where the invasive species has replaced the native one.

The data collected during the samplings, together with those of action A9.3 (for fish) will be used to update the conservation status of aquatic species in Umbria and for the drafting of Action Plans to improve the conservation status of all freshwater fish species and crustacean listed in the HD. Thanks to the results of this action, it will also be possible to identify the river stretches in which to focus the C18 actions for concrete conservation interventions in favor of *Salmo cettii* and *Padogobius nigricans*; it will also provide fundamental indications (for the aquatic part) in the management of IAS (Action C19), and for identifying the sites where river connectivity restoration interventions are carried out (Action C20), restoring otherwise isolated native fish populations and avoiding facilitating the propagation of invasive species.

Each specific Action Plan will identify:

- the areas with populations not having numerical abundances that allow their conservation over time;
- the main pressures for the species and individual populations;
- the areas that need habitat improvement measures and the type of intervention needed;
- the areas that need to contain interventions of alien species;
- the areas where river continuity can be restored avoiding the spread of IAS;
- the areas with populations having numerical abundances that allow their conservation over time.

For *Salmo cettii*, the Plan will also indicate (Action C18.1):

- the level of introgression of each population with the alien genome;
- the areas in which to collect the breeders to be used for artificial insemination in the regional ichthyogenic center of Borgo Cerreto;
- the number of breeders that can be collected from each site;
- the subject areas for the different concrete conservation actions (see Action C18.1).

For *Padogobius nigricans*, the Plan will also indicate (Action C18.2):

- the areas in which the species became extinct due to the competitive exclusion with the alien *Padogobius bonelli*;
- the percentage of native gobies compared to the alien ones, for the sites in which *Padogobius bonelli* was introduced;
- the subject areas for the different concrete conservation actions (see Action C18.2).

The drafting of the Action Plans will be carried out in the 1st year of the project, therefore it will be approved by the Umbria Region and made downloadable from both the project (Action E) and institutional (Umbria Region) websites.

This action will also produce the executive plans for the structural adjustment of the Regional ichthyogenic Centers of Borgo Cerreto and Sant'Arcangelo, to enable them to breed specimens of *Salmo cettii* and *Padogobius nigricans* for the action C18.

### **A12.3 Priority Intervention Plan for amphibians and reptiles species and conservation interventions projects**

The action entails carrying out preparatory and preliminary activities to action C17 (Conservation interventions for Amphibians and Reptiles) and the action E4 (Communication campaign for Amphibians and Reptiles).

During the first year of the project, a Plan of priority actions for the target species (*Bombina pachypus*, *Triturus carnifex*, *Emys orbicularis* e *Vipera ursinii*) will be drafted, based on current knowledge on the presence, distribution and size of the species (Ragni et al., 2006,



Regional Fauna Observatory Database, Regional Observatory for Biodiversity Database, Rural Landscape and Sustainable Design) and on the priorities highlighted in the PAF.

The Plan of the interventions foresees an extremely detailed feasibility study aimed at guaranteeing the success of the proposed activities. The Plan entails:

- identify the habitats which require improvement actions based on the state of conservation of the environments and existing populations.
- identify and selecting the populations of *Bombina pachypus* and *Emys orbicularis* capable of withstanding withdrawals of breeders, to be used in breeding facilities, and those requiring support to reach the numbers enabling their conservation over time.
- identify drinking ponds in Umbria colonised by *Triturus carnifex* and selecting those which require environmental improvement.
- identify the main pressure factors known for *Vipera ursinii* in the regional territory and identify the areas of intervention according to the creation of awareness products aimed at users of the areas characterized by the presence of *Vipera ursinii*.
- It will also contain a section in which the awareness raising actions to be carried out in action E4 (Communication campaign for amphibians and reptiles) will be defined.

In line with the contents of the Priority Intervention Plan for Amphibian and Reptile species, experimental interventions for the conservation of amphibians and reptiles to be carried out under C17 will be designed within the first project phase (2nd year). The necessary preparatory activities for their implementation will be carried out. Specifically, it is expected to proceed with:

- Designing environmental improvement actions for *Triturus carnifex* on wet biotopes (ponds) used by livestock and obtaining the necessary authorisations (Landscaping authorisation, Environmental Impact Assessment, etc.). In detail, the types of actions to be designed and subsequently implemented will regard creating natural aquatic biotopes adjacent to ponds currently used as tanks supplying livestock drinking troughs, and, where possible, the rewilding of sections of the pond shores. At least 10 biotopes, among those identified in the previously drafted plan of priority actions, will be involved in the final-executive planning of the environmental improvement actions for *Triturus carnifex*.
- The design of a breeding facility for *Bombina pachypus*. A portion of publicly owned land, within the Umbrian range of the species, will be identified already in the initial stages of the project, suitable for creating small ponds for *Bombina pachypus* to breed. This requires a small space and the town of Spoleto has shown its availability in cooperating in this aspect with a letter supporting the project. Furthermore, a space will be designed within the Science and Territory Museum – owned by the Town of Spoleto and managed by the Int.Geo.Mod. group and Studio Naturalistico Hyla – to set up tanks for the hatching of eggs and the growth of tadpoles in a controlled environment. Lastly, a breeding and reintroduction protocol will be drafted, to be applied during management of the facility. The necessary authorisations will be obtained in this phase.
- The design of improvement actions of breeding sites for *Bombina pachypus*. Among the sites identified in the plan of priority actions, at least 15 drinking troughs will be selected, where improvement actions will be designed to make them more suitable for *Bombina pachypus* to breed.

In addition to planning any replacement or repair of the tanks should water leaks be detected, and to guarantee the water supply, internal and external ramps will be designed on all fountains to make them suitable for Amphibians breeding and to reduce the potential trap effect which could be caused if the individuals are able to enter but the water level does not allow them to exit.

- The design of an *Emys orbicularis* breeding facility at the ichthyogenic centre of San Feliciano managed by the Umbria region and AFOR. Tanks will be designed in the outdoor areas of the facility to rear and reproduce breeders and individuals to be reintroduced. Furthermore an indoor area will be designed inside the facility with tanks for the early growth of the newborn (two years).

A breeding and reintroduction protocol will be drafted in this phase to be applied during management of the facility, which will continue to be carried out by the Umbria Region with the support of the herpetologists of Studio Naturalistico Hyla. The necessary authorisations will be obtained in this phase.

The actions on the biotopes will be designed by designers of the AFOR supported by the specialised herpetologists of Studio Naturalistico Hyla.

During the action, the need of eventual permits and authorizations will be verified. In case of need, the authorizations and permits will be asked to the relevant Authority in order to carry out the correspondent C actions.

*Reasons why this action is necessary:*

Due to the peculiar characteristics of aquatic environments, freshwater fish and crustaceans are the species that more than others need to be safeguarded as they are all strongly threatened. The action is necessary because it is preparatory to achieve the objectives of the actions C18, C19 e C20 of improving the conservation status of aquatic species in DH. For *Salmo cettii* the action is necessary because it allows increasing the knowledge on the alien Atlantic trout in Umbria, the level of introgression of native trout populations, and the demographic and genetic information necessary to select the intervention sites for action C18. The action is also preparatory in the management of the aquatic IAS (Action C19), because it will provide indications on their abundance and distribution, and will also facilitate the most suitable sites in which to focus the river connectivity restoration (Action C20), restoring native fish populations otherwise isolated and avoiding to facilitate the spread of invasive species. Through this action it will be possible to adapt: i) the Borgo Cerreto Regional ichthyogenic center to the breeding of Mediterranean trout within the action C18.1, and ii) the Regional ichthyogenic center of Sant'Arcangelo for the stocking of eggs and of the juvenile stages of *Padogobius nigricans* within the action C18.2.

The adverse state of conservation in which most of the Chiroptera species are currently found makes it necessary and paramount to implement the intended interventions in the region. The action, which entails identifying priorities and designing concrete interventions (action C16) makes it possible to implement real actions for the improvement of the shelter and foraging habitat.

For Amphibians and Reptiles, the action allows to implement measures to strengthen and conserve the populations of *Bombina pachypus*, *Triturus carnifex*, *Emys orbicularis* and *Vipera ursinii* (action C17) by improving and increasing potential breeding sites.

*Constraints and assumptions:*

No particular problems are expected in the implementation of the action, given the DCBB's and Hyla's expertise in this sector. No particular problems are foreseen for the issue of the authorizations since samplings by electric fishing must be authorized by the Umbria Region; no fish species are listed in Annex V of the Habitats Directive, for which a special authorization from the Ministry of the Environment, Land and sea Protection is required.

The regulation of caves of particular speleological interest might lead to objection and disagreement by their users, specifically, speleological associations. Considering this eventuality, the associations active in the region were involved already at the time of drafting the project, and the action entails jointly drafting a memorandum of understanding with the representatives of the various associations operating in the region.

No particular issues are expected in implementing the actions for the intended activities for Amphibians and Reptiles, as public places suitable to house the required equipment and facilities for the breeding centres have already been identified. Nor is the long term management of the intended breeding centres expected to be problematic. They will be set up inside facilities already existing, which have been operating for years in the region.

Management of the new activities can be guaranteed by the current employees through minimal measures.

*Expected results:*

Through this action it will be possible the i) identification and cartographic delimitation of the range in Umbria of all freshwater fish and crustaceans species in DH; ii) characterization of the biological habitat by measuring abiotic and biotic factors suitable for the presence of freshwater species in DH; iii) estimation of population abundance (n° of individuals, density, standing crop) and age structure of freshwater species in DH. By integrating the collected data with the results of A9.3 action, specific action plans will be drawn up with the interventions to be implemented to improve the conservation status of each freshwater species. The initial monitoring envisaged by the action will provide demographic information to select the sites in which to carry out the action 18 (1 e 2). Another result will be given by the definition of the distribution and abundance of the aquatic IAS (Action C19), that will also allow to identify the river stretches on which to carry out the river connectivity restoration interventions (Action C20).

As far as the Chiroptera are concerned, the action will allow common lines of intervention for the conservation and management of the Chiroptera population to be clearly defined, which shall be adhered to at regional level, and based on which future actions will be developed with greater synergy of aims and smaller expenditure in human and financial resources.

Furthermore, the action will make it possible to implement the initial improvement actions for the long-term conservation and protection of the habitats for Chiroptera sheltering, breeding, wintering and swarming, with a potential increase in the distribution of rare and localised species and increase in the number of populations in Umbria.

For Amphibians and Reptiles, the action will conserve and improve the reproduction habitats in Umbria as well as support the *Bombina pachypus* and *Emys orbicularis* populations at risk of extinction.

*Cost estimation:*

**172.255 €**

69.700 € Personnel costs: they are calculated on the basis of the data collected by the national collective agreements of reference and refer to the personnel currently employed by the partner organizations, as well as to the human resources that will be used specifically for the project. The amounts have been calculated by multiplying the number of man-days foreseen for the relative daily cost.

The costs scheduled for the genetic analysis of fish (DCBB) were calculated by means of a market research and on the basis of the experience acquired during LIFE projects that included similar analyses (LIFE12 NAT/IT/000940 LIFE+TROTA e LIFE18 NAT/IT/000931 LIFE STREAMS).

7.655 € The travels to the sites of interventions were calculated on the basis of the cost of the means of transport and the daily allowances.

30.000 € For external assistance costs, an assessment was made taking into account the average market prices by consulting specialized subjects in the sector.

58.900 € to purchase equipment needed to implement the action. The action involves the purchase of a van (4x4) for an amount of 40.000 €. The van will be used for the transport of people and all the material necessary for fish sampling during all the actions and phases of the project.

For all other costs, an assessment was made taking into account the average market prices and by consulting specialized subjects in the sector.

*Deliverables:*

12 Action Plans for crayfish, bony fish and cyclostomes species present in Umbria 31/12/2022

### **ACTION A.13: Creation of lists of alien species present in Umbria and IAS management strategy**

*Beneficiary responsible for implementation:*

DCBB

This action contributes to the implementation of measure G.1.a (Pg. 50-56) of the PAF.

*Description:*

Invasive alien species (IAS) are considered a major threat to global biodiversity due to the serious ecological impacts that their diffusion entails. In 2014 the European Union (EU) introduced a regulation (EU) no. 1143/2014 relating to the prevention and management of the introduction and spread of IAS. The first priority list of IAS of Union interest was adopted on August 3, 2016. However, a complete overview of the situation at a regional level is still missing: the action, therefore, provides for in-depth studies related to the presence and abundance of alien species present in Umbria Region, defining the lists of those considered invasive (IAS) and implementing a strategy aimed at their prevention and control.

The data collection provide for the precise definition of the number of individuals of the species to "contain". The acquisition of abundance data of invasive allochthonous species within communities is fundamental to define the punctual interventions to be carried out through conservation actions. These interventions will constitute a model to be applied in similar contexts ensuring an improvement of riparian vegetation in general and of habitats 92A0 and 91E0\* in particular. The improvement interventions of the river ecosystem and in particular of the 92A0 and 91E0 \* Habitats, through the containment of the IAS (*Robinia pseudacacia*), was designed in order to pursue the long-term conservation objective, linked to the improvement of the such Habitats. This approach will produce lasting results that will be guaranteed by a change in the ordinary management of vegetation along water courses. The spread of IAS is in most cases due to maintenance of waterways that do not take into consideration the different types of vegetation and their structure. The project actions were structured so as to have a model definition in the preparatory phase. This is based on the mapping, through the preparation of a cartographic database (implementable), which will indicate in addition to the areas on which to intervene and the pressure factors present (practices harmful to habitats and biological habitats of the species), also the different techniques to be used for the improvement of habitats. This phase will therefore be useful to promptly define the concrete interventions, which in addition to improving the structure of the Habitats, will constitute a model that will be the responsibility of the site management body, of transferring the river and lake environments to ordinary practices of management. The guarantee of the effectiveness of the transfer of these management models in ordinary practice will be guaranteed by the presence in the project of two fundamental partners such as the Umbria Region, the managing body of the Natura 2000 sites and AFOR which in most cases is the operational structure that carries out the interventions.

For a better description of the action, the planned activities are developed into sub-actions.

#### **Creation of lists of alien species present in Umbria, recognition of IAS, horizon scanning and prioritization of species of regional importance.**

Through the analysis of the available bibliographic sources (both national/international, e.g Global Invasive Species Database GISD, ISPRA databases, and regional, e.g. Umbria Fish map, Umbria Amphibian/Reptiles Atlas, Umbria Mammal Atlas, Regional Fauna Observatory Database, Regional Observatory for Biodiversity Database, Rural Landscape and Sustainable Design; Alien Plant Cehcklists such as Galasso et al. 2018 on Plant Biosystems 152-3; data banks such as odonata.it and anarchiv.it), and field surveys foreseen in the Action A12, the alien species present in Umbria will be recognized, their distribution areas will be defined and both the sites of the RN2K involved will be identified and the Annex II-IV species affected by their spread. The lists of IAS included in the EU regulations 2016/1141, 2017/1263 and 2019/1262 will be taken into account, plus additional taxa with local role as IAS. On the basis of the information available in the literature and through the involvement of the major experts, a prioritization of the species capable of spreading in the regional territory and most dangerous for native biodiversity (IAS) will be performed. At the same time, an analysis of the main ways used by the IAS to reach Umbria Region will be carried out and a horizon scan will be realized to predict which species are most likely to appear in the near future, also taking into account the current distribution of these species

in the surrounding regions. Horizon scanning will provide a starting point for the design of prevention measures and early control actions for the management of potential IAS.

**Elaboration of the strategy aimed at the control and management of invasive alien species (IAS).**

For each of the IAS identified in the previous sub-action, a regional strategy will be defined aimed at:

- formulating the most effective operations to detect their early arrival (early warning systems), also developing tools to involve the support of the non-expert citizens,
- preventing their further spread, also by assessing Annex I habitats invasibility,
- limiting/removing their presence through eradication (when possible),
- controlling/mitigating the harmful effects on native species and habitats listed in DH.

The results of the horizon scanning will allow predicting which species are most likely to appear and the Annex I habitats most vulnerable to alien invasion (with a higher invasibility) in Umbria, permitting the adoption of strategies for the early containment, considering that eradication is the more effective the sooner it is implemented, since in the initial stages of the invasion IAS have still limited abundances and diffusion.

**Definition of interventions aimed at improving habitats 92A0 and 91E0\* through the containment of *Robinia pseudacacia* and the reconnection between habitat patches**

The sub-action, which will be implemented in the early stage of the project, provides for the definition of activities for the improvement of the conservation status of habitats 92A0 and 91E0\*, identified in the SUNLIFE project, among the habitats that need active intervention of management, since they are in an inadequate or bad state of conservation. In particular, the critical issues derive from a qualitative deterioration, following the presence of invasive alien species, such as *Robinia pseudacacia*, the excessive fragmentation of habitat patches and the presence of synanthropic and ruderal species, since these habitats are compressed into a narrow band between the river bank and cultivated fields. The action is part of the process of 1) prevention, 2) early detection 3) long-term control and containment of the IAS, which we want to design with action A13 and implement with actions C19 and C20.

With the implementation of the management plans of the Umbrian Natura 2000 sites, vegetation maps of the habitats (year 2007-2010) were made, which highlighted for the habitats 92A0 and 91E0\* many areas in which the plant communities are strongly invaded by *Robinia pseudacacia*. The first phase of the action will focus on the mapping and restitution in a GIS environment of the areas with a high presence of *Robinia pseudacacia*, starting from the information contained in the maps of the habitats in the N2000 sites in question. The investigations, which will also be carried out through the characterization of the floristic cortege of the communities (phytosociological characterization), will be concentrated in two sites which include the two main river (and ecological corridors too) of the Umbria region, the River Tiber in the SAC IT5210003 and the Valnerina on the SAC IT5210046 and SPA IT5220025 sites. The subsequent phases will be aimed at defining the containment interventions of *Robinia pseudacacia*, identified among the numerous experienced in projects carried out in different Italian regions (e.g. LIFE11/NAT/IT000187 TEN; LIFE 11 ENV/IT/000243 RII; LIFE15 GIE/IT/001039 ASAP). Given the territorial context of the interventions, which will be carried out in river areas, mechanical intervention techniques will be privileged, avoiding the use of chemical products. Simultaneously with the containment of black locust, planting of individuals of the shrub and tree species typical of the 92A0 and 91E0\* habitats will be planned, in order to improve their phytocenotic quality, restore continuity between the patches of habitats, while limiting the development of black locust same. Within the action, a handbook of good practices will be produced to limit the spread of invasive alien species in agriculture and forestry areas, which will also be disseminated by AFOR through dissemination actions.

The action that will be carried out in the first 2 years of the project will lead to the precise identification of the areas of intervention (georeferenced mapping), the characterization of the plant communities and the definition of the interventions that will be carried out with sub-action C19.3.

During the action, the need of eventual permits and authorizations will be verified. In case of need, the authorizations and permits will be asked to the relevant Authority in order to carry out the correspondent C actions.

*Reasons why this action is necessary:*

The project envisages a holistic approach to the management of invasive species which starts from the recognition of the exotic species occurring in the Umbria Region (A.12), the selection of those most capable of spreading in the regional territory and most dangerous for native biodiversity (priority IAS). Prevention will be guaranteed by Horizon Scanning (HS) with which the Region Umbria will equip itself with a tool capable of predict which species are most likely to appear in the near future, also taking into account the current distribution of these species in the surrounding areas. Another important preventive measure will be guaranteed by raising awareness of the IAS problem and involving citizens through actions to detect (citizens science) the early arrival of IAS (early warning systems).

For each of the IAS identified as present in the Region or HS coming soon, a regional plan will be defined aimed at:

- formulating the most effective operations to detect their early arrival (HS);
- preventing their further spread (priority IAS);
- limiting/removing their presence through control or eradication actions (when possible) (priority IAS);
- controlling/mitigating the harmful effects on native species and habitats listed in DH (priority IAS).

Some control and / or eradication actions of some particularly important IAS will be undertaken already during the project and will become an integral part of regional policies, as in the case of *Robinia pseudoacacia* (C.19), *Trachemys* spp. (C.19), *Salmo trutta* (C.18), *Padogobius bonelli* (C.18), and so on.

The presence and diffusion of alien species will also be taken into account in the interventions of Ecological reconnections (C.20) to avoid that they take advantage of the new ecological corridors created.

The action is necessary because it allows increasing the knowledge on the IAS in Umbria Region, limit their impact and improve the conservation status of the native species and Habitats in DH. The action is necessary to define the interventions to be carried out within actions C19 and C20, in response to the needs identified in the RN2000 regional management strategy and in the PAF, aimed at achieving the objectives of recovery and conservation of habitats threatened by rarefaction and fragmentation processes, triggered or accelerated by disturbing factors such as: presence of invasive alien species, synanthropic and ruderal species, structural simplification of forest communities.

It also responds to the need for concrete interventions aimed at limiting the spread of IAS and consequently improving the conservation status of Habitats 92A0 and 91E0 \*

*Constraints and assumptions:*

No particular problems are expected in the implementation of the action, given the contribution in the project of the major experts for the main systematic groups who have already joined in the prioritization of alien species and in the definition of the IAS lists in Italy.

*Expected results:*

Thanks to this action it will be possible to i) identify the most critical areas in the Umbrian N2K sites due to the presence of IAS, ii) implement strategies for their control, iii) improve the conservation status of native species in DH threatened by the IAS, iv) predict which alien species are most likely to appear in the near future and develop strategies to prevent their arrival. A handbook of good practices will be created to limit the spread of invasive alien species in agriculture and forestry (manual of good practices).

*Cost estimation:*

**140.977 €**

118.007 € Personnel costs: they are calculated on the basis of the data collected by the national collective agreements of reference and refer to the personnel currently employed by the partner organizations, as well as to the human resources that will be used specifically for the project. The amounts have been calculated by multiplying the number of man-days foreseen for the relative daily cost.

4.470 € The travels to the sites of interventions were calculated on the basis of the cost of the means of transport and the daily allowances.

12.000 € For external assistance costs, an assessment was made taking into account the average market prices by consulting specialized subjects in the sector.

6.500 € to purchase equipment needed to implement the action.

*Deliverables:*

List of IAS and Prioritized IAS for Umbria 31/12/2021

IAS Strategy 31/12/2022

Distribution Map of *Robinia pseudacacia* in the SAC IT5210003 sites; SAC IT5210046 and SPA IT5220025 30/09/2022

Vegetational characterization of the areas mapped and identified for the realization of the interventions (Acrion C19) (phytosociological surveys) 30/09/2022

Good practices manual for the management of agricultural and forestry IAS 30/09/2022

Projects of interventions for habitats 92A0 and 91E0\* 30/09/2022

*Milestones:*

Request of permits and authorizations (if needed) 31/12/2022

## **ACTION C.18: Concrete interventions for conservation of *Salmo cettii* and *Padogobius nigricans***

*Beneficiary responsible for implementation:*

DCBB

This action contributes to the implementation of measure G.1.d (Pg. 63) of the PAF.

### *Description:*

The action will be carried out in all phases of the project.

The Mediterranean trout *Salmo cettii* is in sharp decline mainly due to the introduction of Atlantic trout (*Salmo trutta*), an alien invasive species (IAS) with which the native trout can hybridize, leading to a loss of genetic integrity. *Salmo cettii* is considered critically endangered according to the Red List of Italian Vertebrates. The action involves a series of interventions for the *ex situ* conservation of wild breeding in the Regional Centre of Borgo Cerreto regional that can be used to start the production of juvenile specimens for the restocking and reintroduction of the species in natural environments.

The Arno goby *Padogobius nigricans*, considered as Vulnerable according to the Red List of Italian Vertebrate, is in sharp decline throughout its range of distribution, mainly due to the competition with *Padogobius bonelli* an alien invasive species that can outcompete *P. nigricans* for spawning sites preventing its reproduction. In several watercourses, *P. bonelli* has led to the reduction of *P. nigricans* populations and, in some cases, the local extinction. The action involves the development of breeding techniques for the production of juvenile stages of *P. nigricans* in the regional Ichthyogenic Centre of Sant'Arcangelo, starting from egg clutch collected in the wild. Up to now the breeding of *P. nigricans* for conservation purposes has never been attempted; through such experimentation, the production of juvenile specimens destined for restocking and reintroduction of the species in the rivers is expected.

### **C18.1 Interventions for *Salmo cettii* conservation.**

For *Salmo cettii*, the sites object of the interventions will be chosen on the basis of the results of the preparatory action A12.2, among all those in which the species is present: IT5210055-Gola del Corno - Stretta di Biselli; IT5210059-Marcite di Norcia; IT5210048-Valle di Campiano (Preci); IT5210049-Torrente Argentina (Sellano); IT5210065-Roccaporena - Monte della Sassa; IT5220015-Fosso Salto del Cieco (Ferentillo); IT5210071-Monti Sibillini; IT5220020-Gole di Narni; IT5220017-Cascata delle Marmore; IT5210041-Fiume Menotre (Rasiglia); IT5210053-Fiume e Fonti del Clitunno; IT5210044-Boschi di Terne – Pupaggi; IT5210045-Fiume Vigi ;IT5210046-Valnerina.

The interventions are articulated in the following main steps:

the first step of the action, we will involve the **structural adjustment of the Borgo Cerreto fish farm**, whose executive project was carried out within the action A12, for its following use for the breeding of wild specimens and juvenile stages of *S. cettii*. The fish farm currently produces adult brown trout as support for sport fishing and it is able to execute all the techniques for breeding coldwater fish. However, some structural adjustment works must be carried out to allow the correct housing of wild specimens and the survival of the juvenile stages of Mediterranean trout and, above all, to avoid the mixing of the different stocks that will be present in the fish farm until the structure will not be completely converted to the production of Mediterranean trout only. In addition, the environmental education classroom already present in Borgo Cerreto must be equipped to adequately accommodate students within the communication campaign planned by Action E4. Prevention is certainly one of the best strategies to combat the introduction of the IAS and the environmental education can be used to inform students about this phenomenon that is deeply felt in Valnerina, a territory where trout fishing covers an important social and economic role.

**Production of the pure wild trout** to be used for restocking and restoration of *S. cettii* populations. The river stretches with pure (or almost pure) populations in which wild spawners will be captured by electrofishing and selected by the use of molecular markers will be identified according to the results of the action A12.



The field activities aimed to catch the wild spawners will be carried just before the mating season. The DCBB staff will carry out a series of two samplings for 6 years (second – 7<sup>th</sup> year): the number of wild spawners to be collected in each watercourse will be estimated on the basis of the demographic analyses carried out during the action A12. After electrofishing collection, the wild spawners will be tagged (by Passive Integrated Transponder and elastomers). From each fish, a small fragment of fin will be taken and biometric and morphological parameters will be recorded. The spawners will be transported in Borgo Cerreto hatchery awaiting the results of genetic analysis. The adipose fin clips will be transferred to the selected genetic laboratory for DNA extraction and subsequently genetically analyzed with the same methodologies used in the action C19.2. Genetic analyses will be carried out on a total of 1200 wild native spawners (300 x years in the second – 4<sup>th</sup> year, 100 x year 5<sup>th</sup> - 7<sup>th</sup> year). Only pure wild spawners, with a level of introgression null will be used for the artificial reproduction.

Once the genetic analyses will have been completed, artificial reproduction will be carried out at Borgo Cerreto during the winter season (from December to March) by RU technicians with DCBB supervision. After each reproductive season, non-pure spawners will be translocated into fishing ponds isolated from the river networks. Thanks to this action, 100-150 pure wild breeders are expected to be kept at the end of the 3 years of sampling, with an average of about 100 breeding specimens usable per year for artificial fertilization and a mortality of 50% of the adults once artificial fertilization has occurred. On the basis of these data it is presumed to obtain a number of eggs equal to 46 000, thus subdivided:

2<sup>nd</sup> year: 50 females weighing 100 g, average fecundity 2 000 eggs/kg) -> 10 000 eggs;

3<sup>rd</sup> year: 25 females weighing 200 g of the previous year+100 females weighing 100 g -> 15 000 eggs;

4<sup>th</sup> year: 12 females weighing 250 g two year earlier,+ 25 females weighing 200 g of the previous year+100 females weighing 100 g -> 21 000 eggs;

During the 5<sup>th</sup> year, specimens born in the Borgo Cerreto hatchery from the first reproduction attempt (first generation) can reproduce and the production of eggs in the following years will be mainly used for the **restocking** activities, allocating about 15 000 fry per year for the last 3 years of the project (5-6-7 years). From the 5<sup>th</sup> year onwards, wild breeders will be used to supplement the stock and limit the loss of genetic variability. The overall egg production deriving from the fertilization of the Mediterranean trout breeders can be dedicated to the conversion of the Borgo Cerreto hatchery to the exclusive production of stock for the restocking of the entire regional territory. The restocking action will continue after project end. Only stocking of fish from the same river basins will be allowed.

An important part of the action will be dedicated to the **eradication of some Atlantic trout populations** that will be replaced with newly settled Mediterranean trout. This conservation activity will be performed adopting two-pass electrofishing method. A priori, a total of 3 sites are foreseen. The rationale of the above number of sites is related with the potential of fry production of Borgo Cerreto during the last 3 years of the project (5<sup>th</sup> -7<sup>th</sup>). A total of nine eradication events per site in three years will be programmed between summer and autumn (3 events in the 2<sup>nd</sup>-3<sup>rd</sup>-4<sup>th</sup> years of the project). Only streams of modest dimensions will be selected (e.g., no longer than 3 km). In this case the genetic and demographic characterization of trout population will be of crucial importance: only low abundance and completely alien genome-introgressed populations will be removed. Habitat characterization made during Action A9.3, as well as the detection of barriers that prevent the spread from downstream of alien trout will play a key role in the selection of suitable sites in which carry out removal activities. The action will be carried out by DCBB, RU and AFOR, with the support of the volunteers of the sport fishermen's associations. During each field phases the demographic parameters (trout population density and biomass) will be collected by DCBB for subsequent evaluations on the effectiveness of the removal activities (action D.1). Finally, alien specimens will be transferred (no profit) to small fishing ponds isolated from the river networks. The transfer will be carried out using trucks outfitted with tanks.

After the last season of alien trout removal (4<sup>th</sup> year), the corresponding river stretches will be involved in **restoration** activities with the native fry produced in the Borgo Cerreto Farm:

restoration actions are foreseen in the period March – June of the last three years of the project (5<sup>th</sup>-6<sup>th</sup>-7<sup>th</sup> years), the restoration action will continue after project end.

### **C18.2 Interventions for *Padogobius nigricans* conservation.**

For *Padogobius nigricans*, the sites object of the interventions will be chosen on the basis of the results of the preparatory action A12.2, among all those in which the species is present: IT5220006-Gola del Forello; IT5220003-Bosco dell'Elmo (Monte Peglia); IT5220002-Selva di Meana (Allerona); IT5210040-Boschi dell'alta Valle del Nestore; IT5220011-Lago di Alviano; IT5210073-Alto Bacino del Torrente Lama; IT5210039-Fiume Timia (Bevagna - Cannara); IT5210061-Torrente Naia; IT5210001-Boschi di Monti di Sodalungo - Rosso (Città di Castello); IT5210003-Fiume Tevere tra San Giustino e Pierantonio; IT5210006-Boschi di Morra – Marzana; IT5210011-Torrente Vetorno; IT5210012-Boschi di Montelovesco - Monte delle Portole; IT5210013-Boschi del Bacino di Gubbio; IT5220024-Valle del Tevere: Laghi di Corbara – Alviano; IT5210025-Ansa degli Ornari (Perugia); IT5210022-Fiume Tescio (parte alta) ZSC-ZSC IT5220001-Bagno Minerale (Parrano); IT5210054-Fiume Tevere tra Monte Molino e Pontecuti (Tevere Morto); IT5210075-Boschi e pascoli di Fratticiola Selvatica (Valfabbrica).

The interventions are articulated in the following main steps:

The action will start at the third year of the project and it will involve the **structural adjustment of the Regional Ichthyogenic Centre of Sant'Arcangelo sul Trasimeno** for the production of *Padogobius nigricans* juvenile stages. This is necessary because the structure is currently dedicated to the breeding of fish species (cyprinids and Italian pike), mainly to support the populations of Lake Trasimeno. Therefore, although it is able to support all the breeding techniques of warm water fish species, some adjustment works must be undertaken to allow the correct housing of the eggs and the survival of the juvenile stages of *P. nigricans*, very delicate phases of a species sensitive to the alteration of the water quality. For this reason, it is expected that the Ichthyogenic Center of Sant'Arcangelo will be equipped with a more efficient filtering system, allowing reaching a higher level in the quality of the feed water of the breeding tanks.

Always in the 3<sup>rd</sup> year of the project in the areas chosen on the basis of the results of the preparatory action A12.2 (approximately 10 sites) an egg sample will be collected and subjected to genetic analysis during the following two months by means of a PCR-RFLP protocol already developed by DCBB, using a fragment of the 16S rRNA gene and specific restriction enzymes capable of discriminating between the two species. Genetic analyses are necessary since the nests of *P. nigricans* and *P. bonelli* are indistinguishable and a correct diagnosis is fundamental for the continuation of the action. The eggs of *P. nigricans* will then be kept on the Sant'Arcangelo al Trasimeno facilities and the most suitable techniques will be identified to allow hatching and optimize the survival in different experimental conditions (at least 3 different conditions).

In the last step of the action, the **removal of the nest of *P. bonelli* and the stocking with *P. nigricans* fry** will be expected. This phase will be carried out starting from the 4<sup>th</sup> year of the project and for the subsequent 4 years (4<sup>th</sup>-7<sup>th</sup>) in the reproductive period and it provides for the collection of the nests of both species from the sites where they cohabit. At least 300 nests per year will be collected and brought to the Sant'Arcangelo facilities using tanks equipped with an oxygenator. DCBB will participate in the sampling with 2 people for 10 travels per year in 4 years. The eggs will be kept in the tanks of the Ichthyogenic Center and determined to identify the parent species by the genetic analyses already described. The eggs belonging to *P. bonelli* will be destroyed, while those of *P. nigricans* will be kept in captivity until the reintroduction in their original locations. Depending on the results obtained from the experimental breeding techniques developed in the previous point, the most suitable vital stage for reintroduction (eggs, juvenile fry with calf sack, juveniles), i.e. able to maximize the survival in the natural environment, will be decided. In this way, a minimum of 10,000 juveniles of the native species is expected to be introduced every year (150 nests x 100 eggs = 15,000 - 30% of mortality) and to suppress 15,000 eggs (150 nests x 100 eggs) of the invasive species. The transport of the juveniles to the restocking sites will be carried out by DCBB with vehicles equipped with tanks and oxygenators: 2 people will participate in the sampling for 5 travel per year in 4 years. During the action, techniques for the recognition of the parental species of the nests will be tested directly in the field, in order to increase the number of the nests

of the alien species to be eliminated, thus making the action adoptable at a larger scale, much cheaper and incisive.

*Reasons why this action is necessary:*

The action guarantees the full conversion of the Borgo Cerreto regional fish farm to the production of the Mediterranean trout juveniles to be used for the restocking, with the potential to produce several million specimens per year. In this way, a rapid improvement in the purity level of the populations affected by the interventions will be guaranteed. Through the eradication activities, new pure populations of Mediterranean trout will be settled: overall, therefore, the action will allow improving the conservation status of *Salmo cettii*.

For *Padogobius nigricans* the action is necessary because it allows stopping the decline of *P. nigricans*, an endemic species of the Tyrrhenian regions of central Italy, preventing the spread of *P. bonelli*. By developing methods to recognize the parental species of the nests, it will be possible to undertake concrete action to improve the conservation status of the native species.

*Constraints and assumptions:*

No particular problems are expected in the implementation of the action, given the contribution in the project of the experts who participated in the LIFE12 NAT/IT/000940 "TROTA" and LIFE18 NAT/IT/000931 "STREAMS". For eradication, the experiences already carried out with LIFE12 NAT / IT / 000940 "TROUT" have shown that if the sites are selected with the right criteria, a removal of up to 90% of the individuals initially present can be guaranteed; this value will allow the replacement of the alien species with the native one with a not too high restocking effort.

As regards the recognition of goby eggs, no particular problems are expected to arise because molecular diagnosis techniques have already been developed and used by DCBB. Some problems could exist for the breeding and the hatching of the eggs and the survival of the fry since the breeding of *Padogobius nigricans* has never been experienced before. In any case, if the breeding will be impossible or very difficult, it will be possible to reduce the time of housing of the specimens on the Centre to the bare minimum or even plan to reposition the eggs in the places of origin. Another possibility is to collect eggs from sites where only abundant populations of the native species are present, and use them for restocking activities (after genetic characterization).

*Expected results:*

Thanks to this action it will be possible to i) identify the most critical areas in the Umbrian N2K sites due to the presence of native trout population with high level of genetic introgression ii) implement strategies for the restoration of native populations through removal of alien specimens and restocking, iii) significantly increase the number of specimens of the native species compared to the densities and to the percentages of introgression observed in the initial monitoring.

Through this action, it is expected to significantly increase the number of individuals of the native species and to reduce those of the invasive one in the intervention sites with respect to the densities and percentages of relative abundance obtained in the initial monitoring action.

*Cost estimation:*

**721.162 €**

482.282 € Personnel costs: they are calculated on the basis of the data collected by the national collective agreements of reference and refer to the personnel currently employed by the partner organizations, as well as to the human resources that will be used specifically for the project. The amounts have been calculated by multiplying the number of man-days foreseen for the relative daily cost.

18.880 € The travels to the sites of interventions were calculated on the basis of the cost of the means of transport and the daily allowances.

90.000 € For external assistance costs, an assessment was made taking into account the average market prices by consulting specialized subjects in the sector.

65.400 € to purchase equipment needed to implement the action

For all other costs, an assessment was made taking into account the average market prices and by consulting specialized subjects in the sector.

*Deliverables:*

First report on artificial reproduction results for *Salmo cettii* 30/04/2023

Second report on artificial reproduction results for *Salmo cettii* 30/04/2025

Report on reproduction and restocking activities of *Salmo cettii* 31/07/2027

Report on reproduction and restocking activities of *Padogobius nigricans* 30/06/2024

Second record on artificial reproduction and restocking results for *Padogobius nigricans* 30/06/2026

*Milestones:*

Structural adjustment of Borgo Cerreto Regional fish farm 01/09/2022

Structural adjustment of Sant'Arcangelo Regional fish farm 01/12/2023

Production of first stock of native fry of *Salmo cettii* 31/03/2023

Production of first stock of native fry of *Padogobius nigricans* 31/05/2024

## II FASE

### **ACTION C.12: Conservation interventions and action plans implementation for aquatic ecosystems**

*Beneficiary responsible for implementation:*

DCBB

This action contributes to the implementation of measure G.1.d (Pg. 63) of the PAF.

*Description:*

#### **C12.1 Implementation of conservation interventions of rare habitats for the regional territory (Habitats 7210\*, 7230)**

The action involves a series of activities planned in sub-action A9.1 aimed at maintaining and restoring rare habitats as they are present in very few stations in the regional territory. The actions will be carried out between the 3rd and 7th year of the project.

For **habitat 7210\***, present exclusively along the shores of Lake Piediluco (TR) (the only station in Umbria), within the SAC IT5220018 and SPA IT5220026 sites, maintenance interventions will be carried out and, where necessary, restoration of the habitat for about 1000 m<sup>2</sup> of surface. Some portions of floating mats in the most natural and less frequented part may also be made, taking advantage of the experience gained with action A8, in which prototypes of structures for the formation of floating mats will be made and tested. The restoration and expansion of habitat 7210\*, the definition of which will also take into account the experiences gained within LIFE11 / NAT / IT000187 T.E.N., will take into account any sources of pressure by providing long-term mitigation solutions. The maintenance interventions will be carried out through mowing of the helophytic herbaceous vegetation and removal of all the shrubby and arboreal component. Where provided, the expansion interventions will be carried out by means of the collection and preparation of *Cladium mariscus* rhizomes, taken from the species' presence stations at the N2000 site and then planted in the identified areas.

For **habitat 7230**, present only in the marsh of Colfiorito (PG) site SAC IT5210072, (the only station in Umbria), maintenance and, where necessary, restoration of the habitat, defined with sub-action A9, will be carried out. The planned interventions will concern:

- Cutting of the herbaceous, shrubby and arboreal vegetation, with subsequent removal of the plant material, on a surface of about 2.5 ha. (repeated over at least 3 years)

Restoration / adaptation of the network of canals that convey the water from the cultivated fields, towards the swamp, relative to a surface of about 4 ha around the intervention area.

The actions described will be carried out by the DCBB, which will take care of the technical-scientific part, in collaboration with AFOR, which will guarantee the construction of the structures and the specialized workforce for the implementation of the interventions.

#### **C12.2 Application of Action Plans to improve the conservation state of temporary ponds (Annex I Habitats 3130 and 3170\*)**

This sub-action is aimed at performing concrete conservation actions reported in the habitats action plans for the two point-type Annex I Habitats 3130 and 3170\* in Umbria in the main sites of occurrence (ZSC IT5210020 and IT5210013 which host the most valuable expressions of these two habitats), based on the Action Plans produced by sub-action A9.2. The activities will include, for instance, the removal (pruning and, when needed, eradication) of shrub/tree species and perennial nitrophilous herbs invading the ponds; the removal of the leaf-litter accumulated in the ponds; the removal of the alien invasive moss *Campylopus introflexus*. In all cases, the removal will be done manually, due to the fragile structure and the small dimension of the patches of occurrence of these habitats.

This sub-action will be implemented in the second phase of the project and will be repeated for at least 3 consecutive years (years III to V, possibly extending till year VI in case of need of repetition for failed activities). This will also give the opportunity to monitor the speed of accumulation of leaf-litter and the re-growth of invading species (shrub/tree species and perennial nitrophilous herbs, *Campylopus introflexus*), in order to adjust and complement the Action Plans.

The sub-action C12.2 will be conducted by DSA3 staff. In addition, external assistance (junior experts in botany) will be provided to collaborate in all the activities.

*Reasons why this action is necessary:*

Sub-action C12.1 is necessary to implement the interventions for the conservation and improvement of the conservation status of very rare habitats for the 7210\* and 7230 regional territory.

The sub-action C12.2 is needed in order to concretely contrast the phenomena (i.e. successional processes, alien invasions, nutrient accumulation) currently acting as pressures on the Habitats 3130 and 3170\* and seriously threatening their conservation status. The Application of specific Action Plans will help halting further degradation and recovering significant occurrences of these peculiar habitat types in Umbria.

*Constraints and assumptions*

No particular problems are expected in the implementation of the action, given the skills of DCBB and DSA3 in the sector.

*Cost estimation:*

**368.938 €**

## **ACTION E4: Dissemination campaigns on habitats and species conservation in Umbria**

*Beneficiary responsible for implementation:*

DCBB

This action contributes to the implementation of measure G.1.a (Pg. 50-56) of the PAF.

*Description:*

The aim is to disseminate the results of conservation activities and projects and the best practices identified to extend the benefits of the activities carried out. The activities will be carried out starting from the second phase of the project.

### **Dissemination of good practices for grazing and mowing**

a) Drafting of Guidelines for sustainable livestock and "biodiversity-focused" production systems, with an active role in the preservation of Annex I Habitats and Annex II-IV plant species.

This activity will be based on the results achieved in A10, C14 and C15. Specifically, it will be built on the following pillars:

- Management of the extensive farms, taking into account: Nutritional behaviour of grazing animals; Palatability of grasses and forbs characterising semi-natural grasslands and shrublands in the studied areas; Grazing strategies in balance with the territory; Plant macronutrient characteristics (fibre, protein and sugars content according to the plant phenological stage) and plant secondary compounds (condensed tannins, hydrolysable tannins, saponines aromatic compounds); Reared breeds; Preventive strategies of the predation activity by great carnivores (wolf); Organisation of an agro-forestry-pastoral farm by the means of Geographical Information Systems.
- Chemical and nutritional characteristics of livestock products obtained by the means of these techniques;
- Economic efficiency.

b) Open Access Scientific papers

The results of the survey and those of the trials carried out during the project will be published in at least 2 Open Access publications on national and international scientific journals; the results will be spread around as well in the form of scientific communication and/or posters in national and international meetings/congresses focused on the topics.

c) Development of an educational package for visits to the local HNV farms to observe and learn technics and results of sustainable livestock and "biodiversity-focused" production systems.

d) Development of an educational package for the students of the Technical Institute for the Agriculture in Sant'Anatolia di Narco (PG), aimed to increase the awareness on the environmental emergencies and on the possible sustainable and "biodiversity-focused" production methods.

Knowledge concerning extensive livestock production systems and their ecosystem services as instruments for safeguarding grassland biodiversity, achieved during the project Actions, can be considered fundamental in creating in the new generations the awareness of the role that can be played by pastoralism and other livestock extensive systems for the environment, the landscape and the human communities living in the Apennine. "Ad hoc" seminars on the subjects will be provided at the Agriculture Technical Institute in S. Anatolia di Narco, also as a stimulus towards the economic opportunities that pastoralism can play in mountain areas.

e) Knowledge and know-how are the basis for proper management of a farm and for the operators who gravitate around it. In particular, for agro-pastoral farms, knowledge on the correct management of animals (feeding and reproduction) and on the grazing strategies most suitable to the species bred and to the territory, become fundamental in order to recover lost knowledge (as in the case of pastoralism) and to acquire new ones (land management techniques). Courses will be organized providing theoretical knowledge on: the feeding behavior of grazing animals, their physiological aspects and rearing techniques; the main spontaneous forage plant species characterizing Annex I habitats and their botanical and chemical-nutritional characteristics; Annex II-IV plant species typically hosted in these habitats; practical lessons for planning the optimal grazing seasons and areas, on a cartographic basis; principles of grazing techniques. Finally, specific case studies will be addressed to allow the acquisition of skills in the solution of technical-practical problems that can occur in animal breeding.

### **Dissemination for restoration works in wetlands environments**

An information brochure (in English and Italian) will be printed in a limited number of hard copies (500), while an electronic version will be published and distributed on the project website, for the dissemination of action results for the restoration of aquatic ecosystems (actions A8-C11 and A9-C12). The aim of this sub-action is to disseminate the results of the habitat maintenance and restoration activities carried out by the project and to increase the awareness over wetlands environments, even the small ones, which represent a supporting network for the regional ecological connectivity for many vertebrate and invertebrate species.

For the sub-action a documentary video will be produced and promoted for educational purpose by means of different media and local broadcasting networks.

### **Dissemination for odonata (action C11)**

#### a) Dragonfly days

The aim of the activity is to promote knowledge, sensitivity and awareness towards the challenging issue of insect decline. In particular, a series of annual events (at least once a year) will be held for students, citizens and technical staff focused on dragonfly conservation, paying particular attention to providing both information on the biology and ecology of the species and key information for their taxonomic determination and conservation management. The events will be part of the national initiative of 'Dragonfly Days', coordinated by the Scientific Society Odonata.it. The present sub-action will continue from the first until the last year of the project. The events will be organized as workshops, field trips or lectures at the main wetlands included in the N2K network with the purpose to reach at least 30 attendees every year.

#### b) Scientific dissemination of odonata results

The results obtained by the field activities carried out on the three dragonfly species will be shared at national and at least 3 international scientific events, focused on the topic and will be published in at least 3 Open Access international scientific journals by the end of the project.

#### c) Brochure on wetlands and odonata species

With the aim to promote awareness of the conservation of dragonflies and their habitats in the Natura 2000 context, a booklet of about 120 pages (in English and Italian) will be printed in a limited number of hard copies (300), while an electronic version will be published and distributed on the project website. The booklet will be completed at the beginning of the fifth year of the project.

### **Communication campaign for amphibians and reptiles (action C17)**

#### a) Development of educational courses and activities for the use of the *Bombina pachypus* reproductive center.

Specific proposals will be developed to encourage the use of the center also by schoolchildren who will have the opportunity to get to know one of the most threatened species of Amphibians at Community level. The production of a 1.000-copies printed brochure is expected on the center's activities.

#### b) Dissemination publication on good practices for the management and conservation of habitats for amphibians.

The conservation of the Amphibians also passes through a correct management of the small wetlands present in agricultural areas, on the prairies or simply in private gardens. Small ponds, fountains, ditches and small streams are among the most important habitats to be protected in order to promote the conservation of amphibians and it is extremely important to inform citizens about the actions and good practices to be encouraged. 3.000 copies are expected to be printed.

#### c) Development of educational courses and activities for the use of the reproductive center of *Emys orbicularis*.

Specific proposals will be developed to encourage the use of the center also by schoolchildren who will have the opportunity to get to know one of the most threatened species of Amphibians at Community level. The production of a 1.000-copies printed brochure is expected on the center's activities.

#### d) Brochures aimed at users of the areas affected by the presence of *Vipera ursinii*. Being in Umbria the presence of *Vipera ursinii* limited to a few areas within the Monti Sibillini National Park



characterized, especially at certain times of the year, by a high tourist use is of fundamental importance for the protection of the species to inform users of the good practices that must be followed along the paths that affect the most sensitive areas.

### **Communication campaign for bats (action C16)**

a) Disclosure of the recovery activities of the disused Enel masonry substations for the construction of refuges and publication of the results obtained. To encourage awareness among the population, the 15 Enel masonry substations that will be transformed into shelters for chiroptera fauna will be equipped with as many information panels on the importance of the conservation of the roost, with particular reference to those used by anthropophilic species.

b) Disclosure of regulatory activities for access to cavities of chiropterological interest. An information panel is expected to be installed near the caves subject to intervention, illustrating the importance of the cavity for Chiroptera and the regulation of access.

c) Communication campaign on Chiroptera articulated in:

Meetings with schools with construction and installation of bat boxes at school buildings. The purchase of materials for the construction of 300 bat boxes is expected.

Realization of a popular notebook on the relationship between man and Chiroptera printed in 3.000 copies.

Organization of 5 bat nights

### **Dissemination campaign for fish species (action C18)**

During the project, the development of educational courses and activities for the fruition of the Borgo Cerreto Ichthyogenic Center and the green classroom by students and citizens will be envisaged.

Explanatory panels will be created to be installed along the Nera river and its main tributaries, with the aim of illustrating the recovery project for Mediterranean trout and indigenous crayfish.

a) 10.000 brochures will be made to be distributed together the fishing record books issued by the Region of Umbria to sport fishermen; in these brochures the Mediterranean trout will be described from a morphological point of view, and the elements useful for its recognition will be provided, indicating the best practices to follow during the fishing activities.

b) Proposals will be made to encourage the fruition of the Ichthyogenic Center of Borgo Cerreto and the green classroom, and make students aware of the importance of river ecosystems and promote knowledge of the characteristics of aquatic organisms. The production of an informative brochure printed in 1.000 copies on the activities of the Ichthyogenic Center of Borgo Cerreto is expected.

c) Three territorial meetings (start, half, end of the project) will be organized with the fishermen's associations, managing bodies, local associations, trade associations and public administrations to encourage the participation of local stakeholders: during these meetings the problems related to the presence of alien brown trout will be illustrated, and the activities implemented in actions C13 and C19 will be disclosed, also through visits to the areas of intervention.

### **Enhancement of SUNLIFE portal phase III: start-up of the IAS section – Update of information and distribution data (action C19)**

The information and distribution data for each identified IAS will be updated or inserted in the SUNLIFE portal, also in coordination with other already existing dedicated offices and tools (e.g. <http://www.portaledisinfestazione.org>). This can contribute to increase citizen awareness about the relevance of this issue, and can have important implications for containing the spread of IAS. In many cases, in fact, the propagation of IAS can be due to the release by citizens who are not aware of the consequences that such behaviour can lead. Citizen involvement can also play a useful role in the early identification of a recently introduced IAS.

### **IAS Pet species dissemination**

At the same time and following the eradication interventions of the alien tortoises, with particular reference to the areas where the reintroductions of *Emys orbicularis* are present, it is planned to carry out dissemination actions to inform the citizens about the management of the company's animals through the production of a brochure which indicates good animal management practices

in general and with particular reference to the most problematic species such as alien tortoises. The brochure will be made in 10.000 copies.

### **Dissemination of results of action related to the conservation actions for wolf and brown bear**

a) A technical report on the wolf census protocol developed and applied in action C10 will be produced and disseminated among public bodies interested in wolf conservation and management. The protocol will be cost effective in order to give a feasible option of population size estimate that will help to develop a realistic data base in the Apennine region that is now lacking this instrument. This protocol will be discussed in technical and scientific meeting and presented to relevant stakeholders.

b) The results of wolf population estimate and genetic integrity will be presented in public meetings with stakeholder (at least four in the different areas of the region) in order to guarantee the widest and more transparent information among stakeholders and public opinion in general, taking into account that the lack of correct information is one of the main problem in wolves conservation in the nation.

c) Following the training of inspectors able to be responsible of the control and certification of the damage caused by large carnivores a practical guide will be produced and spread by the help of agricultural association in order to make livestock breeder aware of the basic knowledge need to determine the predator responsible of a kill.

d) Following the action on wolves damage prevention for livestock a practical leaflet on the best prevention means will be produced in order to spread good practices on this problematic issue among livestock breeder with the help of the main agricultural associations, moreover meetings with the livestock breeder representatives will be organized.

### **Dissemination of the results related to birds conservation (action C9)**

a) Following the Promotion of good practices for the reduction of pesticides and the analyses of the outcome of different use of pesticides in agriculture by species richness, diversity, rarity index, conservation value index, community Specialization Index - CSI, specific abundance a series of meetings will be organized at a regional level to present the outcome of different practices and to explain to public opinion the advantages of a wise use of agrochemicals in promoting biodiversity.

b) A technical report on birds diversity in relation to different practices will be also produced and spread among stakeholders to reinforce the message related to good practices in farmlands management.

c) A map of the intervention areas on the action promoted in the safety of high-voltage power lines for Birds together with a leaflet on the advantages for bird conservation of such initiatives will be produced and spread with the help of animal protection association among all potential subjects interested in landscape planning and among general public to raise the awareness on this problem and its solutions.

#### *Reasons why this action is necessary*

The action is required to ensure the dissemination of operational efforts and the results achieved in the interventions aimed at maintaining and improving the conservation status of habitats and species of Habitat and Birds Directives. Furthermore, thanks to the initiatives and the materials produced, we will be able to spread awareness among the citizens about the problem represented by the IAS.

#### *Cost estimation:*

**280.075 €**