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Project Report

Habitat Management of the Endemic and Critical Endangered Montseny Brook Newt (*Calotriton arnoldi*)

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Abstract: *Calotriton arnoldi* is an endemic amphibian inhabiting Montseny Natural Park and Biosphere Reserve (PNRBM), listed as "critically endangered (CR)" by IUCN. At the end of 2016, the Life Tritó del Montseny (LIFETM) project (LIFE15 NAT/ES/000757) was launched. The aim of the project was to promote around fifty actions to ensure the conservation of *C. arnoldi* and its natural habitat, and this entailed five strategic lines: (1) Increasing the scientific and technical knowledge with regard to *C. arnoldi*'s conservation status and its habitat management. (2) Expanding its geographic distribution. (3) Involving and engaging stakeholders in the conservation of the Montseny brook newt. (4) Eliminating or minimizing threats that exist in the riparian habitat. (5) Establishing proper legal coverage and defining long-term strategic planning. The successes and failures experienced throughout the process provide us with essential information that will enable us to develop an adaptive management of the habitat. In order to eliminate or minimize threats to the newt's habitat, some of the actions that are currently being carried out are: (a) Land acquisitions and land exchanges with private properties. (b) Land stewardship procedures, with two custody agreements being signed. (c) Reduction of water withdrawal with nine water catchments and distribution being remodeled. (d) Improvement of water treatments and storage by installing ecological wastewater treatment facilities. (e) Ensuring ecological connectivity and riparian forest restoration. Here, we present an evaluation of the actions carried out to improve the habitat of this species, including the necessary considerations for them to be implemented correctly and to be successful in a natural area, which is under public-private management.

Keywords: urodela; salamandridae; caudata; biosphere reserve; habitat restoration; species management; life project

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1. Introduction

One of the major environmental effects of anthropic activity is the alteration of the hydrological cycle, which directly affects the quantity and quality of freshwater supplies [1,2]. Water resources, usually scarce and unevenly distributed throughout the Mediterranean region, are highly susceptible to Global Change. In fact, a decrease in streamflow discharge in several Spanish rivers over the period 1921–1996 has already been reported [3].

Calotriton arnoldi is an aquatic newt described in 2005 [4] as endemic in the Montseny Natural Park and Biosphere Reserve, (PNRBM from now) and which has a fragmented distribution in two areas located on the Tordera river's eastern and western slopes (Figure 1). Both subpopulations are separated genetically and morphologically [5,6]. After this description, the conservation status changed due to the low number of populations and individuals, and thus, it is now listed as critically endangered "CR" [7]. *C. arnoldi* is only found at altitudes above 600 m asl in clean, cold and well-oxygenated streams surrounded by well-structured beech and oak forests. It is a small newt (maximum total length is 103 mm), the dorsum is dark, and the head is heavily flattened [4] (Figure 2).

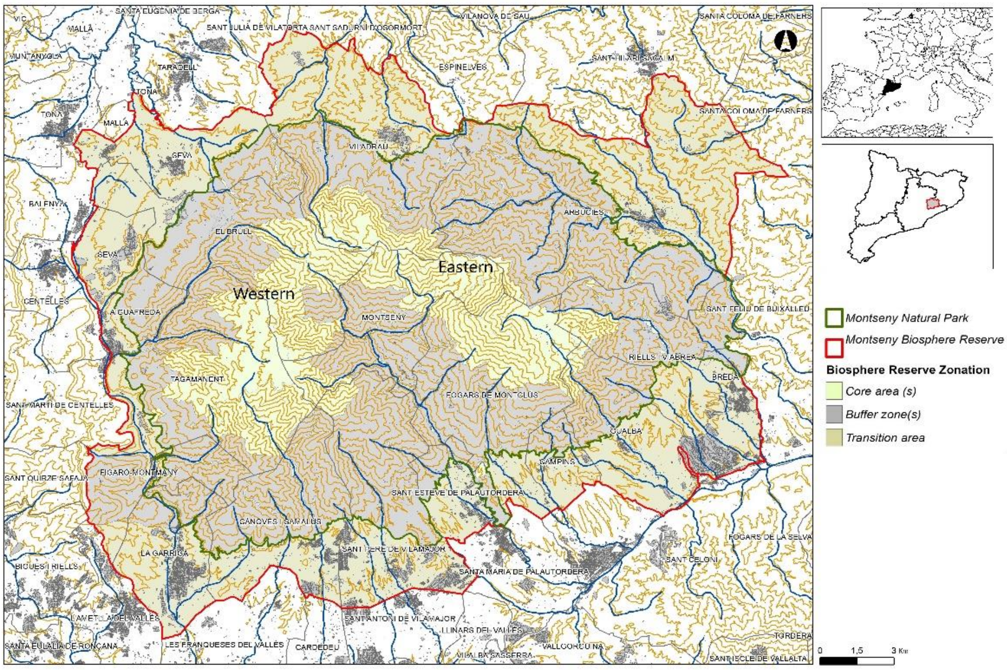

Figure 1. Location of the PNMRB and the two isolated subpopulations (Western and Eastern) of *C. arnoldi* in La Tordera river basin.

Figure 2. (**Right**) Male of eastern subpopulation. (**Left**) Male of western subpopulation.

With the elaboration of the PNRBM's Conservation Plan in 2011 [8], managers were able to strengthen the monitoring program for the Montseny brook newt and its habitat, perform population health surveys, increase and improve the breeding program's functionality and start the release of captive newts in uninhabited streams.

In 2016, the LIFE Tritó del Montseny project (LIFETM henceforth) (LIFE15 NAT/ES/000757) began with Diputació de Barcelona (DiBa), Diputació de Girona (DiGi), Generalitat de Catalunya (GC), Zoo de Barcelona (BCNZoo) and Forestal Catalana S.A. (FC) as partners in the project. The LIFETM provides the necessary tools for a suitable management of the habitat and the species so as to obtain the necessary data, and thus, plan for the future management of this species' populations and habitats (Supplementary Materials Table S1).

Surface water diversion [9] is one of the most dramatic and immediate threats to this species since large amounts of water are being extracted from PNRBM for human consumption and livestock along all the river secondary basins. It is likely that water overexploitation is a severe threat because of the species' ecological requirements [10,11].

In fact, the current environmental legislation is not being enforced efficiently with regard to the maintenance of ecological flows and their monitoring and this constitutes a significant difficulty when dealing with this threat [12].

Wood plantations [9] and, more specifically, those that consist of fast-growing allochthonous conifers, may be behind the waterflow reduction in the La Tordera basin. They require large amounts of water and take up 11.40 ha (12%) within the natural range of *C. arnoldi* [13]. Global warming and other severe climatic or weather events outside the natural range of variation [9] are other threats currently affecting the species. For instance, the beech (*Fagus sylvatica*) forest, an excellent habitat for *C. arnoldi*, has shifted upwards by 70 m at the highest altitudes (1600–1700 m asl) since 1945, and it is being replaced by a holm oak (*Quercus ilex*) forest at lower altitudes (800–1400 m) [14].

The recent appearance of an isolated focus of *B. salamandrivorans* very close to the *C. arnoldi* populations [15], forces us to be very strict with regard to preventive biosecurity measures.

The goals proposed by the LIFETM are: (1) Increasing the scientific and technical knowledge of *C. arnoldi*. (2) Expanding its geographic distribution. (3) Involving and engaging stakeholders in the conservation of the Montseny brook newt. (4) Eliminating or minimizing threats to the riparian habitat. (5) Establishing proper legal coverage and defining long-term strategic planning. This publication aims to be an operational paper for scientists and conservation managers. Fulfilling our objectives (Figure 3) will depend on how we think and act as project managers while being aware of what it means to solve problems through action-oriented approaches (Figure 3) [16].

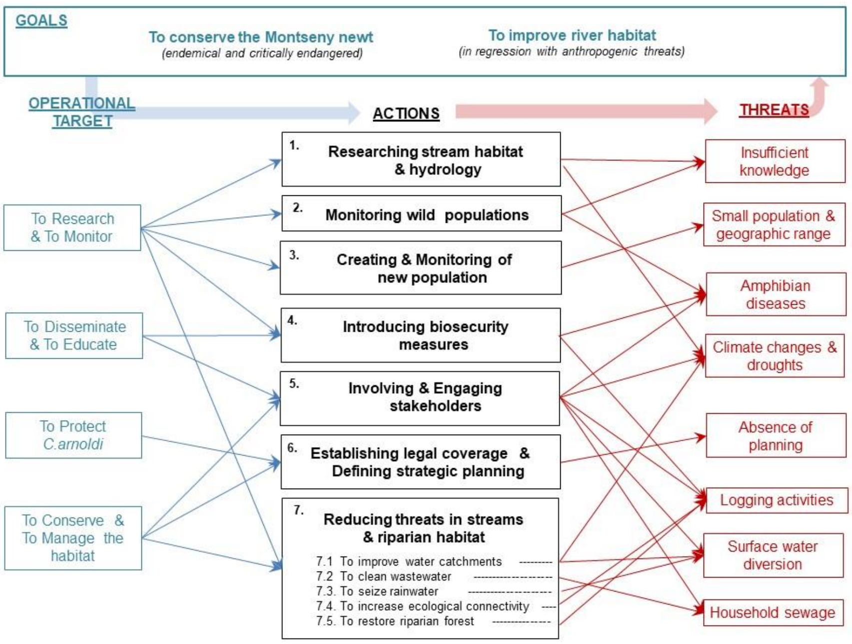

Figure 3. Goals, targets, actions and threats. Two big goals have been established. Of these, four operational targets are specified. To achieve these targets, seven groups of actions have been executed for the different threats to be addressed.

2. Materials and Methodology

Each stream's toponymic name and coordinates have been omitted and population codes are used in all sections in order to protect the species.

2.1. Monitoring Methodology

2.1.1. Researching the Stream's Habitat and Hydrology

A surface water hydrological monitoring network (http://www.lifetrivers.eu (accessed on 14 March 2022)) has been implemented to provide a continuous time series of the brooks. This network currently includes: 5 meteorological stations, 7 (U20-001-04 Data Logger Hobo®) water level sensors, 15 (UA-002-64 Data Logger. Hobo®) light and temperature sensors, 1 (Be-U-4 Hobo® optical base) to download data Logger data and Software (BHW-PRO-DLD Software HoboWare Pro® Windows®/Mac) for data analysis (Figure 4).

Figure 4. Network of monitoring abiotic variables. (**A**) Technicians downloading hydrological data. (**B**) Meteorological station located in eastern subpopulation area.

Stretches of 30 m were established in each brook to perform the hydrological description [17]. A Generalized Linear Model (GLM) with logistic function for each variable has been applied while the registered variables are as follows: Altitude, Slope, LBOM (leaf), FBOM, Bed structure (rocks, stones, pebbles, gravel, sand), Flow, Depth, Wet width, Maximal water speed, Stream structure (runs, falls, pools, dry stretches) and hydraulic status (dry, hyporheic, arheic, oligorheic with some subterranean flow stretches, oligorheic, eurehic, hiperheic).

To analyze the water's chemical components, a total of 59 samples were obtained (40 in streams without newts), and 12 chemical parameters were analyzed: pH, alkalinity, Na^+, K^+, Ca^{2+}, Mg^{2+}, NH^{+4}, NO_3^-, SO_4^{-2}, Cl^-, Cu and conductivity [17,18]. A geomorphologic, geotectonic and hydromorphologic study on the streams where *C. arnoldi* is located compared to where it is not present was conducted to outline the parameters that define the fluvial habitat of the newt. Sixteen streams were analyzed and in each river course, and there were 1, 2 or 3 control stations, resulting in a total of 29 sections of ten meters in length [19].

2.1.2. Monitoring of Wild Populations

In order to avoid the spread of emerging infectious diseases, biosecurity protocols were implemented [20,21] throughout the sampling. The materials on the field were disinfected by using Virkon before and after the visits for each population. Three kinds of surveys were designed to gather ecological data of all known populations.

Intensive Survey

In the chosen A2 stream, a 150 m stretch was selected and divided into sections of 10 m each. The surveys were performed from 2018 to 2020. The stream was regularly surveyed on a monthly basis and newts were actively searched for by moving upwards. Newts were marked by injecting a visible implant elastomer (VIE, Northwest Marine Technology, Inc., Anacortes, WA, USA) and using a visual code based on ten injection points: four in the abdomen, four in the legs and two in caudal region [22] (Figure 5). The captured individuals were either georeferenced, measured and sexed [23], or assigned to one of the four immature age-classes that were taken into consideration (larvae, metamorphic, juvenile and subadult). The sex ratio was calculated as the proportion of mature males in relation to the total number of adults [24]. However, immature individuals were not included in estimated population size models. To estimate population size, we assumed that populations were closed. This assumption is based on the fact that *Calotriton* newts are not particularly mobile organisms [25], and the selected stretch includes the upper limit for the presence of water and the lower limit for the presence of the species in this stream [22].

Figure 5. Marked individual with elastomer in caudal area.

The POPAN model estimator (adaptation of the Jolly-Seber model) was used to estimate population parameters using the MARK^{TM} software [26]. The selected model with lower Akaike value was φ (.) p (t) pent (t) N (Supplementary Materials Table S2), where apparent survival (φ) was considered to be constant over time, while the probability of capture (p) variable over time, and the probability of entry into the population by chance (pent) varied during the period in which they were studied.

Extensive Survey

C. arnoldi is currently known to be inhabiting eight streams, three in the eastern area and five in the western one. Field surveys were performed at night by a team that consisted of the same researcher and two rangers of the GC who are trained in the detection of the species. Samplings were performed either in spring and autumn (55.5%) or only in spring due to the severe droughts experienced in the autumn season. The amount of time spent was proportional to the length of the stream ($\bar{x}$: 973 m ± 95%CI: 140 m, Range: 150–1558 m) to maintain a constant survey effort. No active searching was performed and only specimens which were detected visually were taken into consideration. All observed newts were georeferenced and sexed where possible [23], and three individual classes were considered: adults, immature and larvae. We summarized the gathered information by computing two variables: the percentage of stream length inhabited by newts and the relative abundance of adults (number of adults/stream length) in relation to the survey numbers.

2.1.3. Creating and Monitoring New Population

Due to the species' critical situation shortly after their description, in 2007 a captive stock of newts was started by placing 20 newts from the two subpopulations in the facilities of the Wildlife Center Recovery of Torreferrusa (GC) [27].

The species' potential distribution area was modeled by employing the Maxent maximum entropy method [28]. The lack of biological and environmental representativeness in the available data was also taken into account when calibrating the models. Comparative multidisciplinary studies were performed between the streams which the newt inhabits and those which it does not. The plant structure, hydrology, trophic availability, geomorphology and the presence of predators, among others, were analyzed [17–19,29–32]. A New Populations Analyses Commission was created and worked in parallel with the experts' commission to decide, after several field surveys in optimal candidate streams, on how, where and when to release newts as well as the number and which age-classes should be released. A cost evaluation was also important to determine the project's effort capacity and the viability of its objectives.

New population surveys employ a similar methodology to that of an intensive survey. All of the newts released were taken from breeding centers. Prior to this, the newts were sexed, measured, weighted and marked using Trovan Ltd. microchips. Release points were marked with iron flags, while GPS coordinates were also registered for each point. Once the specimens were released, two active surveys per year were carried out.

2.2. Management Methodology

2.2.1. Introducing Biosecurity Measures

To control and monitor emerging diseases affecting amphibians and the Montseny brook newt, a Biosecurity Commission was created with three main purposes: (i) establishing the protocols to be followed in all the activities carried out in the PNRBM (educational, economic, sports, leisure, etc.) [20,21]; (ii) setting up space and time monitoring processes to determine the presence or absence of pathogens in the species' habitat as well as a security perimeter; and (iii) training of PNRBM workers and raising users' awareness of biosecurity protocols (Figure 6).

Figure 6. Biosecurity procedures to prevent the entry of pathogens in the *C. arnoldi* distribution area. The disinfection of all material and machinery with Virkon S is mandatory before and after all activities.

2.2.2. Involving and Engaging Stakeholders

Only 20% of the surface area of PNRBM is under public ownership. Meetings were arranged with forest owners to reach agreements between both parties. A commission was created to disseminate the LIFETM project among the locals and the general public. In the digital sphere, a website (https://lifetritomontseny.eu (accessed on 14 March 2022)), Youtube channel and Twitter were developed. In terms of educational actions, a travelling exhibition was created: "My name is *Calotriton* and I only live in Montseny" and the educational program "El Montseny a l'Escola" (http://www.elmontsenyalescola.cat (accessed on 14 March 2022)) includes supporting material for the educational community. BCNZoo also offers an educational program on *C. arnoldi*. The involvement of landowners was achieved through plenary meetings, and the mechanisms to involve landowners resulted in the signing of land stewardship and the purchase or exchange of land contracts and agreements.

2.2.3. Establishing Proper Legal Coverage

The Montseny brook newt was included in Annex IV of the Habitat Directive as a *Euproctus asper*. Letters were sent to the Spanish Government in order to pursue the explicit recognition of *C. arnoldi* in the Habitat Directive (92/43/CEE), ensure the conservation of its habitat and develop actions that improve its conservation status. I order to improve the river habitat, a protection zone was drawn, land was obtained, and a recovery plan is awaiting approval.

2.2.4. Eliminating or Minimizing Threats in Streams and Riparian Habitat

Prior to the launch of the LIFETM, the PNRBM Conservation Plan [8] analyzed the main threats, set by the Unified Classifications of Threats [9], and these are: (1) water management/use; (2) household sewage; (3) climate change and droughts; (4) logging roads; and (5) wood plantations. Specific actions were specified in order to reduce the negative impact on the species mainly caused by habitat anthropogenic changes. We grouped habitat restoration actions by using five typologies.

Reducing and Improving Water Catchments

We are promoting the removal of shallow water catchments, the remodeling of existing water catchments, the change from sprinkler irrigation to trickle irrigation and the utilization of rainwater. LIFETM provided the relevant legal information regarding water catchments to the landowners and sent a report to the Catalan Water Agency for their dismantling based on their negative impact.

Cleaning Wastewater

LIFETM improved wastewater that had been released into the streams which the species inhabits by employing tertiary treatment of wastewater from isolated houses and public amenities. After establishing primary and secondary treatment processes, we built artificial marshlands which consist of shallow ponds with gravel and aquatic vegetation (70% *Phragmites australis* and 30% *Iris pseudacorus*) which means that water emerging from the artificial marshland is discharged into the forest, where it is filtered below ground.

Seizing Rainwater

In order to diminish water over-exploitation, we are currently implementing two actions: rainwater harvesting and changing current irrigation systems. To collect water rain, we employed the catchment and conduction of run-off water, filtering it and storing it in tanks so that it can pumped and used for gardening purposes, to fill swimming pools and for livestock.

Increasing Ecological Connectivity

To increase the number of stream sections that have optimal connectivity for all aquatic organisms, LIFETM has removed forest tracks and rebuilt bridges of different types according to their level of use, while recovering the natural streambed to allow the movement of aquatic fauna along it.

Restoring the Riparian Forest

The restoration of the riparian forest was performed by removing allochthonous conifer plantations, carrying out slope stabilization and promoting the replanting of autochthonous species. Exotic conifers require huge amounts of water and, for this reason, the LIFETM cut out exotic conifers (*Pseudotsuga menziesii*, *Pinus ponderosa*, *P. sylvestris* and *Cedrus* sp). The open spaces created by this logging will be managed to naturally regrow an autochthonous forest with a high diversity of plant species (*Sambucus nigra*, *Alnus glutinosa*, *Fraxinus excelsior* and *Corylus avellana*).

3. Results

3.1. Monitoring Results

3.1.1. Stream Habitat and Hydrology

The long-term monitoring network of hydrological variables has been successful by means of 264 visits to 13 streams. This has provided information on temperature and flow discharge, along with the aquatic status for three years. Currently, this network is fully operational, and the first results have been published [32]. Results show that the brooks with newt populations have not completely dried out despite there being summer hydric stress (Figure 7). The pools and a little surface flow (less than 0.5 L/s) remain, even in the streams with the lowest discharge. Large variations in discharge have been recorded, from 0.5 L/s to 10 L/s, following major rainfalls, suggesting a possible direct relationship between waterflow and precipitation (r = 0.892; $p < 0.001$; $n = 27$), with catchment groundwater having a residual role on the brook flow. Data shows the basal flow rate for most of the streams is lower than 4 L/s, with values below 1 L/s in one of them [32]. There is a certain resilience in the flow at times when rainfall is scarce. Streams do not dry out completely because of the underground circulation of water through the fissured rocks is maintained.

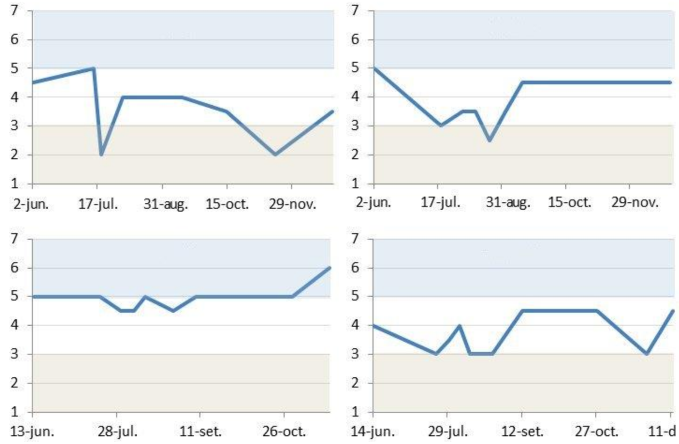

Figure 7. Hydrological temporal variation of four representative streams inhabited by *C. arnoldi*. Values of Y-axis: 1: dried; 2: Hiporheic; 3: arheic; 4; oligorheic whith streaches whith only subterranean running water; 5: oligorheic; 6: eurheic; 7: hiperrheic. Modified from [32].

The water temperature shows the expected annual cycle values and different patterns between the streams [17,32]. The recorded temperatures (Table 1) are within the preferred temperature values (Tp) described for the species (Males, $\bar{x}$: 14.98 °C, Range: 11.7–19.48; Females, $\bar{x}$: 17.51 °C, Range: 15.22 to 20.88) [33].

Table 1. Water surface temperature of some streams which *C. arnoldi* inhabits or where it is absent. Max: Maximal year temperature. Min: Minimal year temperature. Ampl: Average of daily thermic amplitude. Values are expressed in degrees centigrade. Data resumed from [17].

Inhabited	Max	Min	Ampl	Absent	Max	Min	Ampl
B5	17.5	2.52	3.41	Stream 1	17.92	3.26	2.99
B1	15.2	7.38	2.07	Stream 2	17.35	4.10	2.61
B2	20.3	1.76	5.06	Stream 3	16.76	3.05	3.48
A1	14.6	2.09	6.28	Stream 4	17.46	1.33	7.24
B4	16.8	1.76	3.43	Stream 5	16.37	0.35	9.11
A2	17.1	3.58	4.76				
A3	16.7	2.73	6.90				

No significant differences in the habitat's hydromorphology have been found between brooks which are inhabited by the newt and those where the newt is not present (Figure 8). The application of a Generalized Linear Model (GLM) with logistic function for each variable does not show a significant model (chi-square test $p > 0.05$) for all cases [17]. The Montseny's headwater streams are characterized by low mineralization (Table 2). Ammonium has been undetectable in many cases; therefore, it will not be taken into consideration in the statistical analyses [18]. The two groups of streams do not show any significant differences (Figure 8).

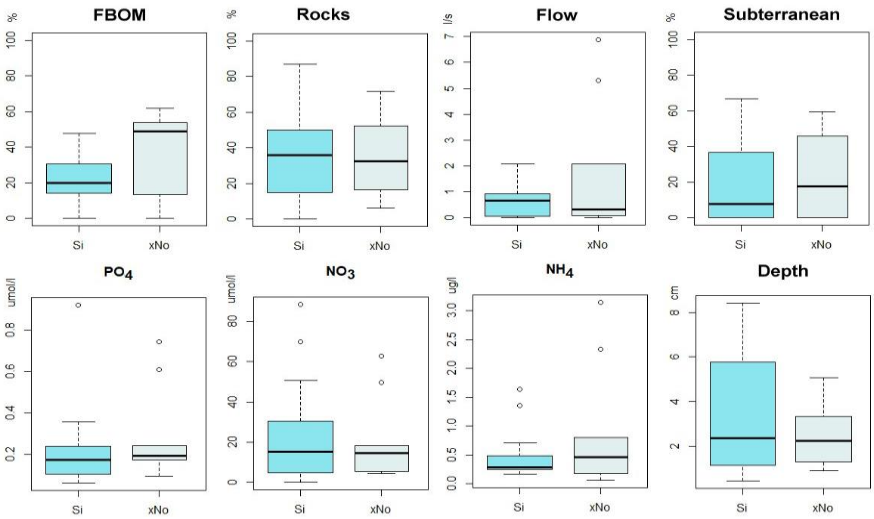

Figure 8. Some hydrolomorphological variables analyzed. FBOM: fine organic matter (expressed as percentage of the riverbed where this matter is found). Si: Streams with presence of newts. xNo: Streams with absence of newts. Depth: Average of Depth in the center of streambed. All comparisons are nonsignificant.

Table 2. Chemical composition of the water in the streams inhabited by *C. arnoldi* and streams were *C. arnoldi* is absent. (Cond): Conductivity in $\mu S/cm^{-1}$; (Alc.): Alkalinity. Na^+, K^+, Ca^{2+}, Mg^{2+}, $NH_4{}^+$, $NO_3{}^-$, $SO_4{}^{-2}$ and Cl^- in $\mu eq/L^{-1}$. Data from [32].

	n	Cond $\bar{x}$	Cond Std	pH $\bar{x}$	pH Std	Alc. $\bar{x}$	Alc. Std	Na^+ $\bar{x}$	Na^+ Std	K^+ $\bar{x}$	K^+ Std	Ca^{2+} $\bar{x}$	Ca^{2+} Std
Absent	40	78.5	41.4	7.4	0.4	520	334	237.1	82.2	10.1	5.8	410.3	295
Inhab.	19	63.5	11.8	7.3	0.1	342	97.1	227.9	44.1	8.2	3.1	264.6	58.3

	n	Mg^{2+} $\bar{x}$	Mg^{2+} Std	$NH_4{}^+$ $\bar{x}$	$NH_4{}^+$ Std	$NO_3{}^-$ $\bar{x}$	$NO_3{}^-$ Std	$SO_4{}^{-2}$ $\bar{x}$	$SO_4{}^{-2}$ Std	Cl^- $\bar{x}$	Cl^- Std
Absent	40	193.1	104.7	0.2	0.5	17.4	25.7	121.7	72.2	118.8	85.9
Inhab.	19	175.5	34.8	0.1	0	17.8	23.6	136.8	57.7	108.3	14.3

The dense surrounding vegetation mainly consists of deciduous trees, which lead to a significant input of fallen leaves in the riverbed. This is the main organic input in the stream system, as the autochthonous primary production is really low (Chlorophyll a concentration under 60 $\mu g/cm^2$), due to low sunlight penetration. Relatively high concentrations of large organic matter (LBOM) and fine organic matter (FBOM) were recorded. Vegetation cover plays an important role in water temperature, preventing an increase where there is a greater degree of cover.

The flooding waters have extremely low conductivity (Table 2) and are poor in inorganic nutrients. The geomorphological data of the torrents [19] indicate that they have exclusive geological and hydrological characteristics. The presence of large blocks and colluvium that form morphological gaps and fissural and cavernous porosity, leading to subway water circulation and the presence of porosity between the blocks, determine the presence of newt. There is a preference for areas with a large accumulation of blocks, with at least 2 levels of stratification, and with large blocks without imbrication as well as a high occupation of the bed. Areas where the bed with rocky substrate is occupied by blocks that form cavities of decimetric order, together with the presence of fracture fissural cavities, may determine the presence of newts [19].

3.1.2. Monitoring of Wild Populations

Intensive Surveys

In the surveyed stream (A2), the largest number of specimens were caught in spring, mainly May–June (Figure 9A). Stream surface groundwater activity decreases during the cold period from January to March and quickly during the summer (July and August), coinciding with the torrent's dry period. These data are in accordance with those provided by different studies [10,18,34]. There is also a significant increase in activity in autumn and early winter [22]. The species has the tendency of inhabiting underground aquifers, between the cracked rocks in the stream, and this influences the timing of surveys because during dry periods, surface activity is almost non-existent.

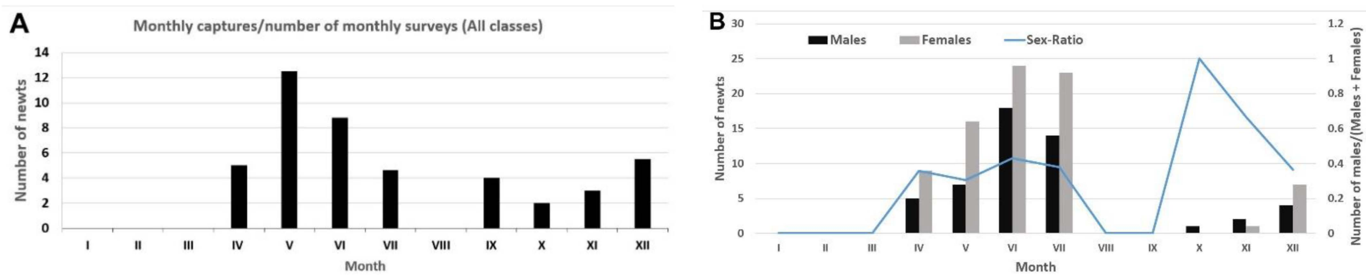

Figure 9. (**A**): Number of individuals from 2018 to 2020 in the A2 stream, captured by surveys in relation to number of surveys for each month. (**B**): Monthly Sex-Ratio in the A2 stream and absolute number of males and females captured each month.

The sex-ratio average obtained (♂♂/(♂♂+ ♀♀)) is favorable to females (0.42), even throughout the year (Figure 9B). These observations therefore match [4,11], which found that sex-ratio is clearly favorable to females in the eastern and western nuclei. After two years, the average displacement is 7.05 m. This mostly consists of males, whose preference is to go upstream.

In the A2 stream, 70 newts were captured, marked and released from 2019 to 2020, and the recapture rate reached 34.3%. Their estimated survival is very high (φ = 0.9984 CI95%: 0.9965–0.9994) and the average density of newts was 0.63 newts/m (CI95%: 0.54–0.79).

Extensive Surveys

All eight populations were monitored by means of extensive surveying (Table 3). The number of observed newts per number of nights spent in stream sections for the period 2010 to 2020 and the samplings during this period showed a decline after there had been increases for two years (Figure 10A). This trend was more marked in eastern populations, which exhibited larger numbers of newts than in western ones.

Table 3. Length of the streams where wild populations of Montseny brook newt were located. Potential: potential inhabited total length of stream (in m). Inhabited: Estimated inhabited lengths (in meters). I/P: percentage of occupied length (indicator of relative stream occupancy). A codes: Eastern populations. B codes: Western populations.

Population Code	Potential	Inhabited	% I/P
B1a	821	83	10.11
B1b	1117	587	52.55
B2	895	475	53.07
B3	932	107	11.48
B4	2220	782	35.23
B5	804	654	81.34
A1	3150	697	22.13
A2	2250	178	7.91
A3	1180	34	2.81
Western Range	6789	2688	39.59
Eastern Range	6580	909	13.82

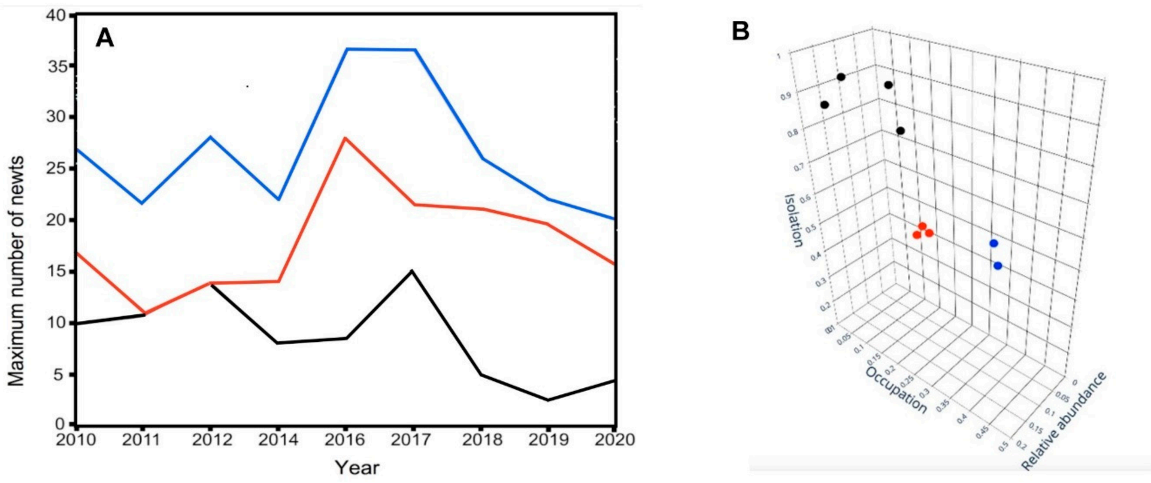

Figure 10. (**A**): Maximum number of newts observed at night for every year. Lack of data during 2013 and 2015 is due to the severe drought affecting the populations that prevented us from gathering enough data (black: eastern populations; red: western populations; blue: total). (**B**): Tridimensional plot of the values for each Montseny brook newt population on three indicators: relative abundance (number of newts/m). occupation (percentage of stream occupied by the population) and isolation (dendritic index between pairs of geographically close populations). Black dots show the tridimensional position of populations with high risk of extinction given their high levels of isolation and low percentage of stream occupation. The best combination of favorable indicators, which are high connectivity, stream occupation and relative abundance, are found only in two populations marked with blue dots, whereas three populations (red dots) showed intermediate risk of extinction.

The tridimensional space generated by the three indicators provides a clear picture when assessing the vulnerability of *C. arnoldi* populations (Figure 10B). Four populations are very isolated from the neighboring ones and in total, two of them occupy a very small proportion of the stream and yet, quite remarkably, relative abundances are high in all cases, which indicates a strong concentration of newts in small sections. The other four populations seem to be in a better situation from a conservation standpoint and show better connectivity and extensive occupation of the stream. However, and perhaps quite strikingly, only two of them additionally have a large relative abundance. Therefore, this ideal combination is found in 25% of the populations and both belong to the western area.

3.1.3. Creating and Monitoring New Populations

The field studies [17–19,29–32] confirm that streams where *C. arnoldi* is absent have similar biotic and abiotic characteristics when compared to streams where the newt is present. Table 4 shows the new populations created prior and during to the LIFETM.

Table 4. New populations established in the *C. arnoldi* potential area of distribution. Only population codes are written because of conservation policies for this CR species. Range: slope location in La Tordera river basin. First release: year when first release was made. Last released: last booster. Last recapture: year when *C. arnoldi* was captured in the stream. Property: Land property where the new population and stream stretch are located.

Code	Range	Number of Released Newts	First Released	Last Released	Last Recapture	Property
A4	Eastern	166	2011	2014	2016	Private
A5	Eastern	63	2020	2020	2020	Public
A6	Eastern	127	2014	2020	2021	Public
B6	Western	436	2010	2020	2021	Private
B7	Western	106	2014	2015	2019	Private
B8	Western	261	2019	2021	2021	Public
B9	Western	267	2021	2021	—-	Public

The average displacement of newts throughout 2019 in B8 stream was 2.44 m downstream. However, displaced recaptured newts move in average 14.75 m downstream and 7.44 m upstream. Despite wild *C. arnoldi* specimens showing very little mobility and dispersal capacity in natural habitats [35,36], it is expected that newts that were born in captivity and released into a stream will disperse and explore more while seeking suitable microhabitats. However, the results do not confirm this (Figure 10B). The dispersion observed in the first recapture in the B8 stream compared to the estimate for the wild population A2 within two consecutive captures is not significant (t-test: -1.21; df: 56; $p = 0.229$). Preliminary results indicate that the population is well established. The distribution of recaptures in the new population B8 is very similar to the distribution of recaptured individuals in the A2 population (Figure 11A). Figure 11B shows the dispersion of the recaptured specimens relative to their release point.

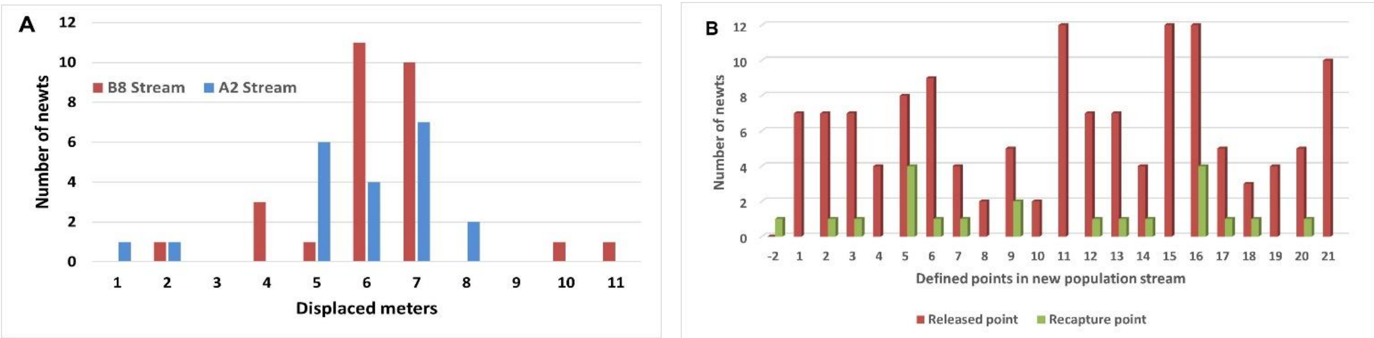

Figure 11. (**A**) Displacements observed for the newly released individuals in a newly created population (A8) in the first year and in the wild population (A2). Horizontal axis represents the number of displaced meters from release to recapture. The positive values indicate upstream displacements and negative values indicate downstream displacements. (**B**) Distribution of newly released newts in the stream B8 and the recaptures during the first year after release. Red bars indicate the release point and green bars the recapture point.

3.2. Management Results

3.2.1. Biosecurity Measures

Throughout 2018, several theoretical-practical training sessions were held for workers and users of the DiBa. At the same time, two biosecurity protocols were developed to be applied in the activities. An extensive one that would cover all LIFETM activities and those that would be developed in the PNRBM (i.e., fishing, surveys, works, etc.), while another protocol focused on educational activities to be carried out in the aquatic environment, aimed primarily at nature schools and educational centers that carry out activities in the PNRBM [20,21]. In 2021, information posters were placed to increase awareness of PNRBM's ban on bathing in rivers and torrents.

3.2.2. Involving and Engaging Stakeholders

The negative effect of visiting the riverside habitat and the dangers associated with the unintentional introduction of infectious diseases by amphibians (Bd, Bsal, Ranavirus, among others) have been highlighted. People who want to see the Montseny brook newt are advised to visit the BCNZoo and CRPS breeding centers. With the aim of disseminating the project, a variety of informative material has been published (Table 5). The monthly number of visits to the website increases with time and are correlated with the number of published news each month (R = 0.670; DF = 40; $p < 0.001$).

Table 5. Dissemination actions and informative material made during LIFETM. (En. Ct. Cs): English, Catalan and Castilian languages. (*): Data from 01/01/2017 to 12/31/2021. (**): Data from 1 May 2018 to 31 December 2021.

	Number of Items	Number of Media or Sites	Number of Receptors/Visitors
Permanent Exhibitions			
Informative panels	8	8	Visitors of PNRBM, BCNZoo, CRFPS and CRFTF.
Temporal Exhibitions			
My name is *Calotriton* and I only live in Montseny	8 panels	23	20,712 schoolchildren/individuals **
Photography (Iñaki Relanzón)	1	1	584 (Web)
Press			
National Press (Spain)	182	18	(55 Written press and 127 digital press) *
National Press (Catalonia)	182	39	(55 Written press and 127 digital press) *
Newsletters	11	3 (En. Ct. Cs)	234 registered *
Information leaflet		3 (En. Ct. Cs)	16,500 unities
Web			
http://www.lifetritomontseny.eu (accessed on 14 March 2022)	125	1 (En. Ct. Cs)	>74,000 visits (9066 users) *
Video capsules	8	1 (En. Ct. Cs)	3163
YouTube Chanel	166	1	12,705 (including video capsules)
Education			
El Montseny a l'escola	3	18	1372 pupils (2021/22)
Workshops	1	1	224 children
BCNZoo	1	1	13,800 schoolchildren/individuals *

The exhibition "My name is *Calotriton* and I only live in Montseny" has travelled to the 18 municipalities that make up the PNMRB. From September 2021, the exhibition will continue to travel around the Library Network of the Diputació de Barcelona. The educational program "El Montseny a l'escola" (http://www.elmontsenyalescola.cat (accessed on 14 March 2022)) has allowed 2450 pupils to participate in the didactic material and 224 children in the workshop to make a plaster newt (Table 5). The new education and breeding center at BCNZoo has become the benchmark facility for the Montseny brook newt and has received almost 500,000 visitors since June 2018. A total of 13,800 people (mostly school groups) have carried out activities on the Montseny brook newt, guided by the BCNZoo educational staff.

Seventy meetings have been held with forest owners. This has led to the involvement of nine estates which the newt inhabits in the Montseny area. Three land stewardship contracts have been signed with local landowners who have *C. arnoldi* in their properties while contracts have been exchanged with two more. The project has been disseminated in 46 conferences, presented at local, national and European technical conferences and congresses (4945 attendees in total). Three hydrology conferences have been held with the participation of 20 scientists from 14 different institutions [37,38]. With regard to foresters and forest managers, a Manual of Good Environmental Practices has been developed [39].

3.2.3. Establishing Proper Legal Coverage

Habitat Directive technicians, from the EU and Spain Government, established that since *C. arnoldi* is a split of the *Euproctus asper*, included in the Annex IV of DH 92/43/CEE, E. asper would be divided into two species, *C. arnoldi* (Code 6920) and C. asper (Code 6944), to prepare the sexennial reports (Art. 17 of DH). A technical document on the recovery plan has been developed, and it is expected to be formally approved before the end of LIFETM in 2022. To improve the protection of the streams where *C. arnoldi* lives, work is being done on two strict protection measures within its distribution area. (i) Supervised management zone has been established on private land and this involves a land stewardship contract to manage habitat conservation targets. These zones have a width of between 50 m and 100 m on each side of the streams. (ii) The nature reserve zone is a legal figure where exploitation of natural resources is banned. As part of the drafting of the PNRBM's protection plan and within the framework of new legislation for the Montseny Natural Park (D.127/2021), it is expected that most brook newt populations will be included inside this zone. Due to the fact that the majority of the newt populations are on private land, 87 ha with brook newt populations have been purchased.

3.2.4. Eliminating or Minimizing Threats in Streams and Riparian Habitat

Reducing and Improving Water Catchments

During the last four years we have removed six water catchments directly affecting populations of *C. arnoldi* and these were entirely financed by the landowners to avoid future fines. From June 2020, the owners of legalized water catchments must ensure sustainable water use and the maintenance of ecological water flow (based on legislation 2000/60/CE. DL. 3/2003. D. 380/2006 and D. 1/2017). The LIFETM advised and promoted the modification and improvement of four legalized catchments. In order to do so, mechanisms avoiding water extraction when water tanks are full were installed. In addition, distribution boxes were installed to restrict the amount of water available for exploitation and which is legally allowed (Figure 12).

Figure 12. Installed mechanisms which regulate water extraction (**A**) and distribution boxes (**B**) restricting the amount of legally allowed water for exploitation.

Cleaning Wastewater

One tertiary treatment has been installed (Figure 13) and when the water is discharged it is in good condition: pH (6.7) and optimal DQO (74 mgO_2/L).

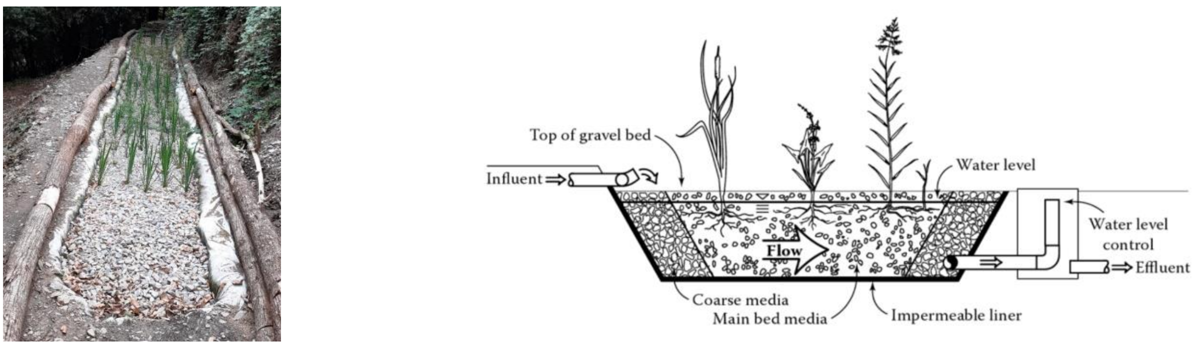

Figure 13. (**Left**) Artificial marshlands formed by shallow ponds with gravel and aquatic vegetation (70% *Phragmites australis* and 30% *Iris pseudacorus*). (**Right**) Schematic draft with permission from [40]. Copyright 2006 by the Water Environment Research Fundation.

Seizing Water Rain

The LIFETM has removed drip irrigation in an area of 6500 m^2 which produces 60,000 plants with a mean water consumption of 20,000 m^3/year [41]. Currently, this area is irrigated by employing 65,000 droppers therefore leading to, at least theoretically, a reduction in water use of 11,700 m^3/year. The LIFETM is promoting rainwater harvesting by reconditioning roofs and roads that cover 5830 m^2 in total. This adaptation may retrieve 4110 m^3/year to tanks, which have been installed in four properties at lower altitudes while owners also use electric or solar-powered pumps to move water to other tanks placed at higher altitudes. Overall, we have installed 14 tanks which has led to a storage of 895 m^3 in total, which will result in economizing 4000 m^3/year of water from the streams inhabited by Montseny brook newts. As an example, for a plant nursery where rainwater is collected and stored in four tanks (total capacity 314 m^3), the estimate is that there will be complete hydric autonomy for 6–8 months per year [41]. Another practical implementation was carried out in a camping area, where we estimate that the system may meet all of the camping area's water demands [42].

Increasing Ecological Connectivity

The LIFETM removed eight forest tracks and built fifteen bridges (Figure 14) of different types according to their level of use, while recovering the natural streambed to allow the movement of aquatic fauna along it. The most relevant work was done in two of the eastern streams, which had sections with under 50 lineal meters of connectivity and this was improved to more than 6 km without any barrier.

Figure 14. Modified bridges to recover the natural stream bed and to allow the displacement of aquatic fauna along them. Left images: before recovering structure. Right images: the same locality after recovering structure.

Restoring the Riparian Forest

About 700 m^2 of riparian forest has been recovered and the growth of autochthonous species has been encouraged (*Sambucus nigra*, *Alnus glutinosa*, *Fraxinus excelsior*, *Corylus avellana*) [39]. More than a thousand meters of barriers were installed to reduce the discharge of erosion sediments by using bioengineering techniques. The LIFETM has already cut down 3.7 ha of exotic conifers, thereby promoting the natural regrowth of an autochthonous forest.

4. Discussion

4.1. Stream Habitat and Hydrology

In PNRBM the holm oak forest appears to be expanding upwards at the expense of beech forests and heathlands [14], which may contribute further to a reduction of the streamflow. Thus, both climate and vegetation cover changes may threaten *C. arnoldi* populations. [43] conclude that in the Montseny Massif, the frequency and length of low streamflow events will increase dramatically. Montseny brook's hydrology is highly dependent on precipitation, and therefore its evolution is linked to climate change and the predictions have been ominous thus far [12,44,45]. The presence of blocks and fractures in the stream bed result in a continuous underground runoff flow that ensures the persistence of newts during dry seasons in which surface water flow disappears [17,19]. The data obtained indicate that the populations of *C. arnoldi* are resilient in a hypogean habitat during dry summer periods, but the current trend is towards population decline [7,11,34,46].

Over the previous 50 years, a decline in river discharges has been observed in Spain and it has been shown that this decline was due to an expansion of shrubs and forests in formerly cultivated areas [43,47–49]. A simplistic conclusion would be that it is beneficial to remove all forests, but this statement is completely misguided because vegetation recovery is generally thought to provide important environmental benefits, such as increases in carbon sequestration [50], and is thought to be the best alternative for nature conservation and biodiversity purposes [51]. Moreover, forests noticeably reduce runoff coefficients and slightly decrease annual flooding, a fact which can be explained by the effects of rainfall interception and forest water consumption that reduce soil water content and limit catchment hydrological responses [47,51]. Changing water availability and demand will require the application of innovative land- and water-management strategies, as well as forest management within the watershed scale, and water use and management [12]. A reduction of between 50% to 44% in relation to the current catchment has been achieved [40], and in some cases, the water automation will possibly last the 12 months of the year thanks to rainwater harvesting [41].

As previously mentioned, within the species' potential distribution area in the upper La Tordera river basin, there are no differences between the torrents where the species lives and where it is not currently found. Therefore, the most plausible hypothesis is that land use in the last 150 years may have caused the rarefaction and disappearance of the species in many torrents (Mining, carbonization, wood extraction, the transformation of forest into pastures with the consequent increase in the insolation of torrents and the contribution of sediments and chemical pollutants).

4.2. Monitoring of Wild Populations

Despite the species' very small geographic range, its distinct populations experience environmental spatial heterogeneity [25,52], which may be behind the differences we found in their demographic parameters [25,46]. The extensive survey highlights that the estimated survival rate is lower in the eastern population than in the western population, whereas population size follows the opposite pattern. One potential explanation is that larger population density increases competition for resources which leads to higher mortality [53]. Firstly, newts spend most of their time within the interstitial hypogeum environment of the stream bed. Secondly, we avoided removing rocks to locate newts to limit the impact of the study, but this is likely to have decreased capture and recapture probabilities.

The intensive monitoring of one eastern population through active search [22,35] resulted in higher population density values when compared to an extensive survey [46] due to the higher number of captures and recaptures. This could suggest that an active search by lifting stones is a more suitable methodology for demographic studies. While this may be the case, the effort required to carry it out for all populations makes it unaffordable for the LIFETM and therefore, it is better to use the suggested methodology for extensive monitoring to estimate global trends. However, the demographic data obtained with intensive monitoring are essential to estimate the ecological parameters needed for modeling new populations and to estimate the probability of survival and the carrying capacity of the streams selected to create it [13].

Prey availability [29] in streams running over beech forests may be greater, as it was also recorded for the sister species *Calotriton asper* [54,55]. The eastern stream also has greater availability of interstitial microhabitats as refuges, and these differences may altogether result in larger population densities in the eastern population [34,35,46]. These results match those provided by [46], which highlight that the differences in *C. arnoldi*'s habitat would explain the differences in demographic parameters between the two analyzed Montseny brook newt populations. [56,57] concludes that the Pyrenean brook newt's presence in streams seems to depend both on the morphology of the stream and on introduced or translocated fish density.

4.3. Creating and Monitoring New Populations

One of the LIFETM's main successes has been to work with multidisciplinary teams and the creation of committees based on expertise, working together to evaluate the hydrological dynamics and actions to create new populations [16,58].

In 2021, the presence of newts has been confirmed in 3 of the 6 new established populations (Table 4) despite the intense drought suffered throughout the year. The adjusted demographic data obtained in the field studies with regard to the wild populations [22,34,35] have allowed us to obtain very precise theoretical models [31]. However, it is still too early to say that the models developed for the creation of new populations are actually fulfilling their function and that the newly created populations are actually establishing themselves.

The new population established in B8 shows similar dispersal characteristics to the wild population in A2. Thus, it cannot be confirmed whether previous life in captivity increases dispersal, searching for optimal habitats in a new environment. It could indicate a good choice of a torrent for the creation of a new population [36]. On the other hand, the weight of the recaptured individuals after one year in relation to the time of release gives an average weight gain of 1.36% [35].

4.4. Biosecurity Measures

Biosecurity measures and protocols have clearly proven their effectiveness. Despite the few cases detected of Bd, in one pool it was caused by the illegal release of a non-native amphibian species of the PNRBM (far from *C. arnoldi* distribution area). None of the three diseases surveyed (Bd, Bsal or Ranavirus) has been detected in any *C. arnoldi* wild or newly created populations. This indicates that our methodology with regard to biosecurity protocols is the appropriate one to prevent the entry of pathogens into the species' wild habitat. The location of Bsal in a nearby area outside the PNRBM [15] requires us to be extremely strict when implementing biosafety protocols in the basin inhabited by *C. arnoldi*. On the other hand, there are no data on the effects that Ranavirus has on *C. arnoldi*, and its recent expansion in the north of the Iberian Peninsula means that prevention is of the utmost importance [59].

4.5. Involving and Engaging Stakeholders

The actions proposed in LIFETM have in many cases exceeded the capacity of the project itself, mainly due to problems that have arisen in the management of *C. arnoldi*'s habitat, which is mainly located on private land. The species' recovery plans carried out, as well as the reference guides on species recovery plans, highlight the importance of public property for the management of the habitat and the species [58]. However, land purchase or exchange and stewardship agreements as well as the actions related to the efficient use of water resources, although incomplete, can be regarded as another success and as a good result for the work carried out as part of the project.

4.6. Establishing Proper Legal Coverage

Genetic results indicate that there is an ancient isolation between the eastern and western subpopulations. For this reason, the LIFETM has treated both subpopulations as two distinct units both in terms of population management, breeding centers and the creation of new populations. However, if Global Change endangers the species in its natural area in the future, there would be the possibility of creating hybrid populations because what would really be endangered is the species itself. This is an objective not raised in the LIFETM, but should be discussed in an experts committee, in order to reach a consensus and it would require the GC's approval. It is the administration responsible for the species conservation, as the Recovery and Captive Breeding Plan prepared by them raises the treatment of the two subpopulations as differentiated evolutionary units [27].

4.7. Eliminating or Minimizing Threats in Streams and Riparian Habitat

New population hydrological and expert commissions are a very effective way to make a threat assessment like TRA (Threat Reduction Assessment) [60]. Furthermore, the long-term monitoring of this species is mandatory in order to provide managers with useful information to repeatedly evaluate the state of conservation and threats of *C. arnoldi* populations. However, the detectability of *C. arnoldi* greatly depends on the occurrence of environmental conditions such as temperature and hydroperiod. For these reasons, only the persistent absence of observation year after year during a long period of sampling can be understood as evidence of population extinction.

The restoration of the riparian habitat is a job that does not yield immediate results. To determine how and where the interventions are working, threat reduction assessments should be performed to measure the conservation project's success [60]. However, some actions such as creating barriers against erosion and the removal of barriers that cross streams have been effective. For example, the intensive study of the A2 wild population [35] revealed that the bridge that crossed the population exerted a barrier effect for the connectivity between both sides of the torrent and produces the connectivity of both populational subgroups.

Changes in temperature or precipitation volumes have the potential to produce shifts in phenology and the timing of reproduction [61,62]. The models of climatic change [12,45] for the Mediterranean area forecast more irregularity in precipitation patterns, specifically more rainfall in autumn than in spring, and concentrated over a few days in some years while in others, there will be heavy droughts. Air temperature is rising [63] and the Montseny massif in particular experienced an increase of around 0.3 °C per decade in the second half of the 20th century [12]. A temperature increase can lead to enhanced evapotranspiration [63], which eventually will result in reduced streamflow. Besides climate change, landscape cover type has an influence on water resources as well. Several catchment studies have shown that an expansion of forest cover leads to a decrease in the water yield [64,65]. A reduction in tree canopy cover shows an increase in annual water yield, higher for conifers than for deciduous hardwood forests [3]. Trees intercept large quantities of rainfall, from 18% (scrub) to 32% (forest) vegetation, which has great repercussions on vegetation cover and the availability of water resources [66,67].

The maintenance of riparian habitat has been described as one of most important action to maintain *C. arnoldi* populations. However, data published on the effects of global warming predicts an increase in slow flows and drought periods [43]. Similar results highlighted the importance of flow intermittence on *Calotriton asper* distribution, with a decrease in habitat suitability when the frequency of zero-flow events increases [57].

The SWOT (strengths, weaknesses, opportunities and threats) of LIFETM is showed in Supplementary Materials Table S3.

4.8. Management Proposals

- Research and monitoring should be promoted in order to manage the natural heritage and resources with technical and objective criteria. However, for effective conservation to be achieved, collaboration from all the participants is needed (researchers, managers, stakeholders).
- Changing water availability and demand and mitigation strategies to improve the management of water resources will need to continue being implemented. This requires the application of innovative land- and water-management strategies on the all-watershed scale.
- Open spaces such as pastures and grasslands should be recovered in the river basin, promoting a landscape with a diversity of habitats that respect the riparian and autochthonous forests on erodible and sloping soils.
- It is highly recommended to promote the rewilding of habitats in order to have an old and mature forest, between pastures and grassland, mainly because tree transpiration is sometimes much higher in younger than in older trees.
- Coniferous plantations, especially of fast-growing species (such as *Pseudotsuga menziesii*, *Pinus radiate*, *P. ponderosa*, *P. nigra* or *P. uncinate*), must be removed in the protected areas, in order to increase the streamflow where *C. arnoldi* is present, because the largest water yield increases appear after removal of conifer forest.
- To conclude, the intensive human exploitation of natural resources in the Montseny is a threat to the survival of the *C. arnoldi*. Therefore, we should reduce water abstraction from the streams, and we should restore the diversity of natural habitats, especially by promoting meadows, respecting the natural evolution of the forest and encouraging the development of the autochthonous riparian forest.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/land11030449/s1. Table S1: Actions and objectives of LIFETM. Table S2: POPAN estimators and additional results. Table S3: SWOT (strengths. weaknesses. opportunities and threats).

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