THE CURRENT DISTRIBUTION STAGE OF *Nymphaea lotus* L. var. *thermalis* (DC) Tuzson, A PLANT SPECIES THAT IS THREATENED WITH EXTINCTION IN EUROPE

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Abstract. The *Nymphaea lotus* L. var. *thermalis* (DC) Tuzson or thermal waterlily a thermophilic species is threatened with extinction due to the loss of thermal water in its natural habitat, namely the Lake "Ochiul Mare" of the "Peţea Creek" Natural Reserve. The taxon is related to the well-known species the Egyptian *Nymphea lotus*, but due to it long history of specific physiological and anatomical peculiarities it is considered as native for Romania. The overexploitation of thermal water in the past 30 years led to the cessation of its artesian flow inside the species habitat, causing the drying of the lake and implicitly the disappearance of this characteristic thermal ecosystem that is unique in Europe. Under these circumstances, the scope was to identify specimens of thermal waterlily that were spontaneously propagated in nature, outside the native range area. Based on the field research it was possible to determine and identify thermal waterlily specimens naturally spread, adapted and survived in the Hidişel creek bed, in a previously unreported area, thus occupying a new habitat that provides the species with conditions similar to those of the natural ecosystem. Thus, the occurrence of the thermal waterlily outside the reserve is a certainty, but not the survival of the species because in the new habitat it is subjected to a strong anthropogenic impact, being outside the protection area. Following the research performed both on the trail of the Peţea creek and on that of the Hidişel creek, the main tributary and water supplier of Peţea, approximately 166 stalks of *N. l* var. *thermalis* were identified. Although the trend of the thermal waterlily population that grow spontaneously outside its natural area is increasing apparently, the future of this species remains uncertain. Thus, the need for protection and rescue from extinction is a must and new in situ and ex situ conservation measures should be in place for the near future.

Keywords: *Nymphaea lotus* L. var. *thermalis*; endangered species; "Peţea Creek" Nature Reserve; ROSCI0098 Lacul Peţea.

INTRODUCTION

The rate of extinction of species at the global level is higher and the International Union for Conserving Nature is claiming that almost 100.000 species are at the edge of their survival [5]. However, not all species that should be monitored are already recorded by the IUCN portal. Thus, it is a great need to take care and action for covering as much as possible with protective measures all species that are threatened by new threats or risks for extinction. Several authors are asking if species will be able to develop surviving mechanisms to disperse and to persist in the same climatic niche under so many pressures [28]. For example, in Europe the specialists agreed recently that over 454 tree species are threatened with extinction due to major anthropic pressure and climate change [27]. Habitats as well are threatened with extinction [2] and in Europe relevant studies have been already published [8]. Romania is also facing the same threats, species and habitats being threatened with extinction as well. There is a great need to rethink our conservation strategies and try to couple *in situ* and *ex situ* conservation measures in more cost-effective possible ways [1].

The scope of this article is to reveal for the species *Nymphaea lotus* L. var. *thermalis* (DC) Tuzson new scientific evidences by which we can support that the species being under the threat of extinction due to the loss of its natural habitat may survive due to its incredible ability to disseminate in the same climatic niche by occupying new habitats.

The "Peţea Creek" Nature Reserve is located in Bihor County, the Băile *1 Mai* Resort, that belongs to the administrative territory of Sânmartin Commune (fig. 1 A, B). As a protected natural area of community importance, the reserve is an integral part of the European ecological network Natura 2000 (Natura 2000 site - ROSCI0098 Lacul Peţea), the name of this priority habitat is "Transylvanian hot-spring lotus beds", code 31A0* [14], while Law 462/2001 classifies this reservation into the IUCN category IV as a habitat / species management area [32, 35]. According to the national classification, the name of this unique habitat in our country is R2209 "Tertiary communities relict with *Nymphaea lotus* var. *thermalis*" [13], which until recently, was the only natural thermal ecosystem in Romania, with a well-defined specific physiognomy, currently being in an advanced state of ecological decline. It is already among the major looses of our country as well as of European importance.

The warm waterbodies of the Peţea creek hosted the edifying and characteristic species of this type of habitat: the thermal waterlily (*Nymphaea lotus* L. var. *Thermalis* (DC) Tuzson), this reserve being the only place in the world where this plant species spontaneously survived. Following numerous researches carried out over more than 90 years on the thermal waterlily, it was revealed that this is a species that survived glaciations, and in addition to this scientific curiosity it brings its own floristic value, being considered an endemism. Therefore, following the tenacity actions of the botanist Alexandru Borza, the thermal waterlily was declared a natural monument

since 1931 (Journal of the Council of Ministers no. 148), and a year later, the Peţea creek was declared a nature reserve [10, 11]. This thermophilic species took refuge during the Quaternary glaciations into the warm waterbodies of the Peţea creek together with *Microcolpia parreyssii* a gastropod relict (R.A. Philippi, 1847). Furthermore, these warm waterbodies became the distribution habitat area for thermal rudd (*Scardinius racovitzai* Müller, 1958), a subspecies of rudd.

In the last 200 years, there have been numerous debates among scientists about the taxonomic status of the *N. lotus var. thermalis* species, the thermal waterlily being considered by some researchers as a Teriary relict species [4, 12, 16, 24-26, 31] while other researchers on contrary based on recent molecular analysis proved that th species is not divergent of the Nile waterlily [17]. Beyond these debates, the importance of conserving this species remains vital as well as the ecological restoration of the original habitat and of the unique species community that was hosted [6, 7] and it is a species of community interest in the end for all Europe [17].

The huge anthropic pressure to which the reserve has been subjected in recent years, as a result of irrational and excessive use of the geothermal pool, has led to the cessation of the artesian flow of "Ochiului Mare", the only active spring of Peţea Creek. This caused the drying of the lake surrounding it, a lake that represented the native habitat area of species of conservative interest: *Nymphaea lotus* var. *thermalis* (DC) Tuzson, *Microcolpia parreyssii* (R.A. Philippi, 1847) and *Scardinius racovitzai* (Müller, 1958). The drying of the lake led to the degradation and then to the almost entire disappearance of the habitat code 31A0* (i.e. Transylvanian hot-spring lotus beds), as well as to its replacement with other communities of species, following the ecological succession [35].

At present, none of the three endangered taxa can be found inside the borders of the reserve, the species *Microcolpia parreyssii* (R.A. Philippi, 1847) being lost forever, *Nymphaea lotus* var. *thermalis* (DC) Tuzson survived as few specimens and *Scardinius racovitzai* (Müller, 1958) is surviving only in botanical gardens [33], aquariums or museums.

Predicting the ecological disaster towards which the "Peţea" Creek Reserve was heading, our team of researchers from the University of Oradea, started a series of studies in order to save the thermal waterlily from extinction. Thus, some of the scientific methods used for the conservation of the thermal waterlily involved plant biotechnology techniques, aiming to identify sterilization protocols for in vitro culture, the optimal composition of culture media for initiation and micropropagation (i.e. minerals and growth regulators formula) as well as the type of physical support for *in vitro* seedlings [3, 23], while other research tried to study the optimal conditions for thermal waterlily relocation into a new artificial thermal habitat [29].

In 2015, after the implementation of the project "Conservative management of Natura 2000 sites in the custody of the Museum of the Land of Crişuri", the field inventory of species was performed, which led to the identification and localisation (at that time) of a number of only 70 specimens of thermal waterlily, the population being present spontaneously in the Ochiul Mare (i.e. 33 mature specimens located into the basin of the lake), but also into the Peţea creek, after the confluence with Hidişel creek, in spots where the water had a slower flow - 37 copies [35].

At present, the *N. lotus* var. *thermalis* species is completely extinct from its native habitat („Peţea Creek" Nature Reserve), however, some sporadic occurrences have been recorded outside its distribution area, namely in the valley of Hidişel Creek, where a small number of scattered specimens has been identified. The possibility of self-maintenance of the population outside its native habitat, in the same climatic niche, and the probability of spreading is minimal, as thermal waterlily specimens occurring in the creekbed are vulnerable to high floods risks due to the late torrential rains claimed by climate change and also to anthropogenic pressures, as this area is outside of the Reserve. Therefore, mandatory requirements are species reintroduction into its naive habitat, monitoring and also the implementation of a responsible management plan for ensuring a safe habitat in which the plants can occur naturally. It should be noted that the small number of specimens determines a low degree of diversity and genetic variability, impacting the endangerment of its survival and the decline of the size of population.

Taking into account the advanced degree of ecological decline of its natural and native unique habitat, the high endangerment status of the species today, as well as the scientific knowledge that reports the sporadic occurrence of several thermal waterlily specimens in the neighbouring of its native habitat [35], updating the scientific data regarding its natural distribution in the field of specimens able to spontaneously survive outside protected area was raised. In this respect, the scope of this study is to present new scientific evidences regarding the extraordinary ability of the species to survive outside its native habitat but in the same climatic niche where it is able to disseminate and spontaneously grow. Also we are investigating the major threatens and risks for not taking immediately new protective and conservation measures for species and its natural occurring habitat protection.

MATERIAL AND METHODS

Study area. In order to identify and geolocate specimens of *N. lotus* var. *thermalis* that spontaneously occur in the wild, outside or inside their natural habitat (Ochiul Mare lake of the Natural Reserve "Peţea Creek" from Băile 1 Mai, Bihor county), filed missions were made, into specific sites of the creek beds of the

two creeks, in which local people signalled the presence of the thermal waterlily.

Thus, the Peţea creek was surveyed continuously along its entire length starting from inside the Natura 2000 site ROSCI 0098, while the Hidişel creek was also surveyed continuously from the entrance to the Băile Felix locality up to the confluence with the Peţea creek (fig. 1). It was covered a wide area of at least 10 m distance from each of the creek banks, left and right.

Field equipment. For specimens geolocation it was used a GPS equipment (Garmin GPSMAP 66i). During the research, the water temperature was timely measured, to this end, a dedicated equipment was used (WTW Multi 3630 IDS, equipped with TetraCon® 925 conductivity and temperature probe) and photographs were taken using a field camera (Ricoh WG-50).

Local people knowledge. From the start of these filed missions we collected information from local people of the villages Băile 1 Mai, Rontău and Băile Felix. The communication was on phone, and directly before starting the missions

Field missions. Ten field missions were conducted between August and October, 2019. This period was the best to ensure us to be able to screen as many as specimens in the wild due to the fact that during this period the thermal waterlily plants were in full process of anthesis.

RESULTS

Hidişel creek is a left tributary of the Peţea creek [35], into which it flows within the Sânmartin commune GPS: 47°00'20.4"N 21°58'47.2"E (47.005663, 21.979774). This is a non-thermal tributary creek, but has a higher waterbody temperature compared to other creeks due to discharges of thermal wastewater coming from the resort area (i.e. hotels, guesthouses and houses located in the neighborhood area of the Baile Felix resort). Consequently, the species *N. l.* var. *thermalis* has found a suitable habitat for its *in situ* survival inside the bed of this stream, however in a small number of specimens that have been counted. Hidişel creek is affected by anthropic factors (i.e. constructions and human other activities), the natural habitats being modified by the inhabitants who built dams along the creek, artificial banks, which extract sediments and water from the creek, and modifying the structure of the creekbed. Also, Hidişel creek is the main pollutant of the Peţea creek, as it is discharging large amounts of organic matter of anthropic origin, on the route of the two creeks being identified several pipes discharging sewage waters [35]. All these factors dramatically affect the natural equilibrium of the two streams, as well as the major habitat characteristics for plant and animal species, including the thermal waterlily, which even it is in an ecological decline still occur spontaneously in the waters of the both two streams.

Specimens of thermal waterlily were identified in 5 stream segments, out of which segments I, II and III were located on the route of the Hidişel creek, starting from the exit of the Băile Felix resort up to its confluence with the Peţea creek, while, segments IV and V were located into the Peţea creek after the confluence with the Hidişel creek (fig. 1C). In the segments I-III marked on the map in fig. 1C, the identified specimens are spontaneously occurring outside the Reserve area ("Pârâul Peţea" Reserve).

Segment I. The first segment of the Hidişel creek, in which thermal water lilies were identified, is characterized by banks very rich in plant species, being mainly represented by the allogeneic species and the *Parthenocissus quinquefolia* (L.) Planch. (Virginia creeper), *Typha latifolia* L. (bulrush) invasive liana on the edge of the watercourse and on the shore and *Potamogeton nodosus* Poir. (longleaf pondweed), a predominant hydrophyte into the mainstream. The waterbody has an average depth for summertime of 20-25 cm and a relatively high flow rate. At this point, based on literature the water temperature range between 14ºC during the winter and 29ºC during the summer. The sediment of the creekbed was muddy. 6 specimens of thermal waterlily were identified in this area with exact geolocation. All specimens were found on the edge of the watercourse, in places where the water flow was very low, the sediment with a mud thickness of at least 5 cm and a water depth of 10-15 cm. The identified specimens were in their juvenile stage of development with a number of 3-5 elliptical, submerged, green leaves. GPS: 47°00'01.0"N 21°58'57.1"E (47.000267, 21.982517).

Segment II. The next Hidişel creek segment where the thermal waterlily was located is at the entrance of the Băile 1 Mai resort, a few tens of meters downstream of the first area, reason of which the studied area had characteristics similar to those described above. Thus, the banks were also rich in plant species, mainly invasive alien species of Asian origin *Polygonum cuspidatum* Siebold & Zucc., along with *T. latifolia*, and in some places *Sparganium erectum* subsp. *neglectum* (Beeby) K.Richt. and *Alisma plantago-aquatica* L. being encountered. The waterbody was 25-30 cm depth, but the flow was slower. The sediment of the creekbed was muddy, 4-6 cm thick, without gravel. At this point, the water temperature varies between 13.5ºC during winter and 28ºC during summer. In this area, 2 stalks of thermal waterlily were identified, the specimens being better developed compared to those identified not the previous segment, most likely due to better lighting of this area. They presented 4-5 green elliptical leaves, most of which were underwater. As in the case of the first segment, the two specimens were found near the creek bank, thus avoiding the main current of the stream. GPS: 47°00'06.2"N 21°58'57.3"E (47.001722, 21.982583).

Segment III. The third segment of the Hidişel creek in which specimens of thermal waterlily were identified, was located downstream at a distance of approximately 400 m from the previous segment,

before the confluence of Hidişel creek with Peţea creek (fig. 1C), and located next to a household, whose garden has an exit to the stream. On this section, the width of the creek bed was 5-5.5 m, being the largest throughout its length. Several dams are built from place to place, which has contributed to a significant reduction of the water flow rate and an increase in its depth (30-40 cm).

It appears that this calming area of the creek also had consequences on the increase in thickness of the sediment mud, exceeding in some places 15 cm in depth. In addition, the lighting of this segment is particularly good, lacking the trees and shrubs in the immediate vicinity of the banks. All these conditions appear to favour the spreading and occurrence of thermal waterlily. As a consequence, approximately 100 stalks of specimens have been identified in this area, the plants being vigorous, well developed, presenting numerous elliptical and cordiform leaves, including flowers and fruits. Downstream of the dams, into the water, next to the thermal waterlily, the *P. nodosus* species was well represented. In this area, the water temperature range between 13.5ºC during the winter and 27ºC during the summer. It should be noted that one of the banks was dammed, embedded into concrete, and the other had abundant vegetation, including trees, located at a distance of 3-5 m from the bank. Numerous wastes (used tires, bottles, etc.) were also identified in the creek bed. GPS: 47°00'19.0"N 21°58'48.2"E (47.005267, 21.980050).

Segment IV. The next Hidişel creek segment, in which specimens of thermal waterlily were identified,

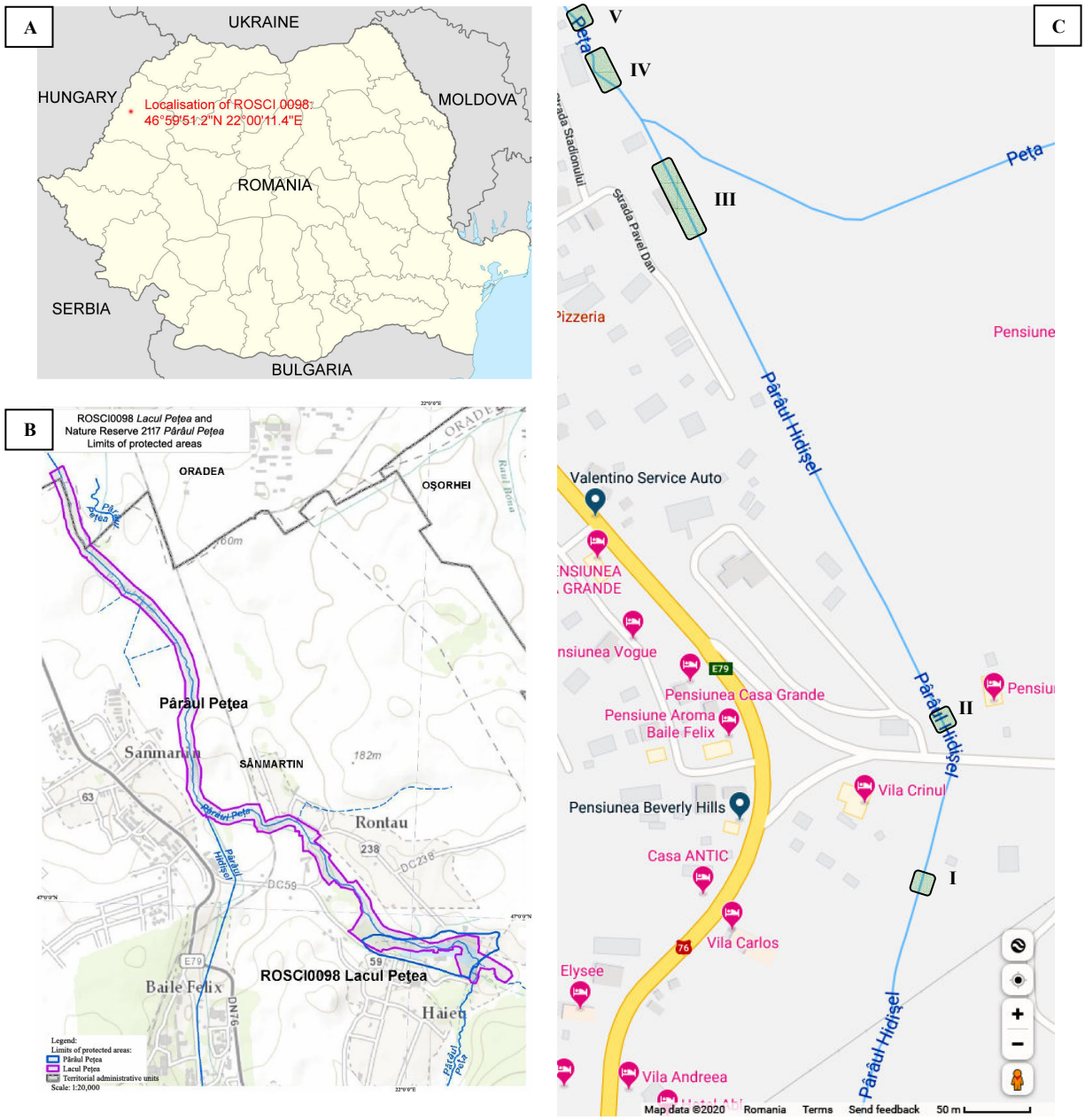

Fig. 1. The location of the Natura 2000 site ROSCI0098 „Peţea Lake" (A), of the nature reserve 2117 „Peţea Creek", highlighting the two creeks where the studies were carried out (B) during the summer of 2019 (map taken from ***1, 2016); the location of the creek segments where the last stalks of *Nymphaea lotus* var. *thermalis* (C) (map taken and modified from maps.google.com).

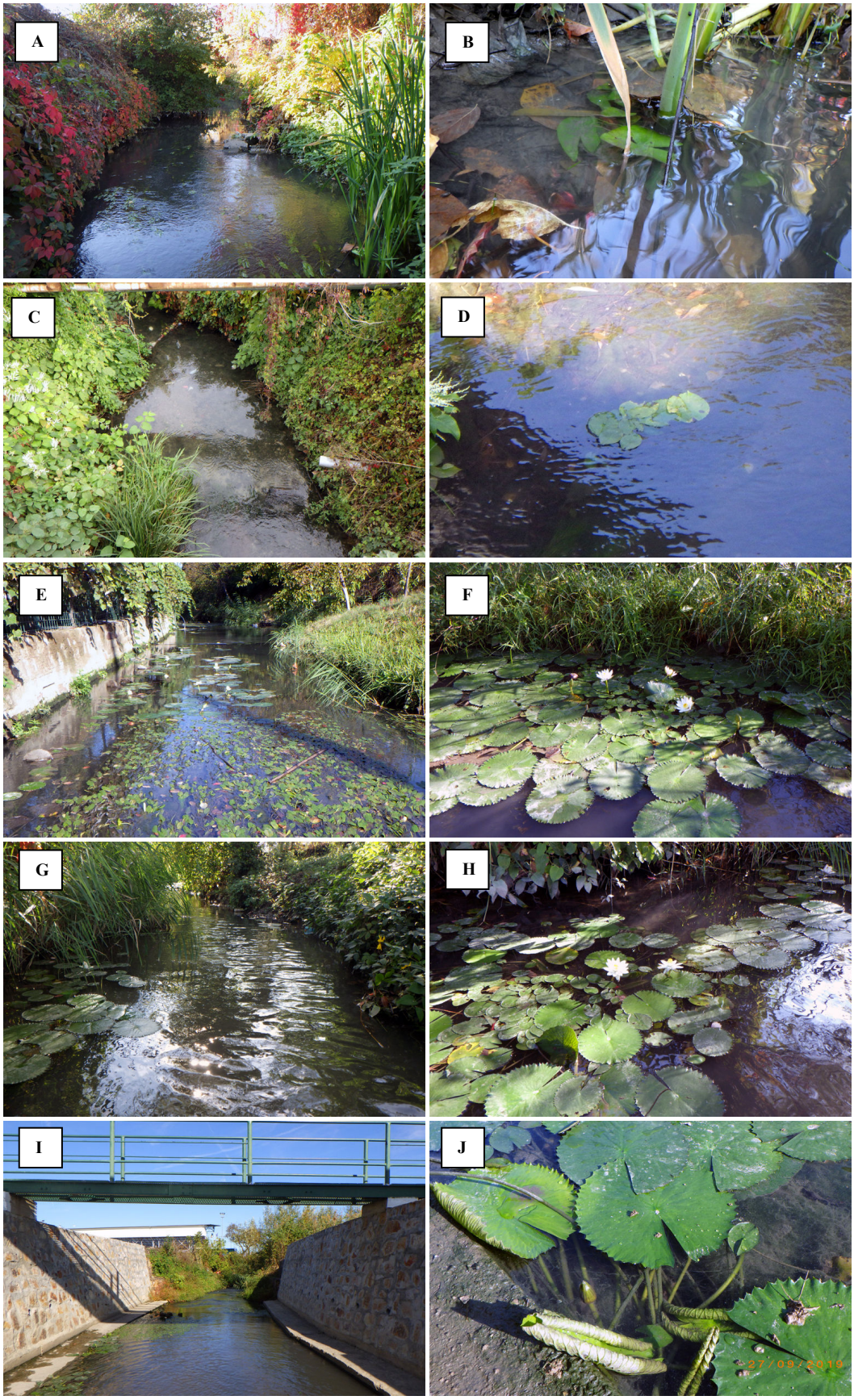

Fig. 2. General views of the creek segments (left column) and stalks of *N. l.* var. *thermalis* identified in each of these segments (right column) during 2019.

was located downstream of the confluence of the Hidişel creek with the Peţea creek (fig. 1C). On this section, the banks were full of grassy and woody vegetation, predominating the *Ailanthus altissima* (Mill.) Swingle (tree of heaven), *Robinia pseudoacacia* L. (Black locust), *Salix x fragilis* L. (crack willow), *Polygonum cuspidatum* Siebold & Zucc., *Parthenocissus quinquefolia* (L.) Planch., *Phragmites australis* (Cav.) Trin. ex Steud. (common reed) species, while in the creekbed, in the parts with shallow water and high current, the species *P. nodosus* was well represented. In general, in this area, the water was 25-40 cm deep, smooth flow (low flow rate), and the sediment was muddy, without gravel or sand. The water temperature in this place varies between 13ºC during the winter and 27ºC during the summer. The light intensity of the creek is medium (bellow 800 lx). There is an alternation between the areas not covered by vegetation and those partially covered. Thermal waterlily stalks have been identified (about 50 specimens) generally in the areas of the creek that were highly illuminated and with a slow flow rate. The plants were well developed, having many large, cordiform leaves, but also buds, flowers and fruits. GPS: 47°00'21.7"N 21°58'45.4"E (47.006033, 21.979283).

Segment V. The last stream segment of the Hidişel creek, where specimens of thermal waterlily were identified, was the sector V (fig. 1C), located next to the Hydrometric Station from Sânmartin commune. In this area, the banks of the Peţea creek are regularized by damming and the characteristic vegetation only sporadically occurred. In this place, the water of the stream had small depth (10-15 cm) and low flow rate due to the width of the creekbed, which is of 4.5-5 m. In the parts of the creekbed with a slower flow and in the vicinity of the thermal waterlily stalks, phytocenoses of the *Potamogeton nodosus* species together with filamentous algae of *Spirogyra sp.* were found. In the middle of the water stream, the predominantly sandy sediment becomes partially muddy towards the edge of the creekbed, where the flow rate of the stream has been halved.

The water temperature varies between 13ºC during the winter and 26ºC during the summer, the lighting being maximum due to the lack of vegetation on the banks (up to 10 000 lx during the clear sky). The waterlily plants identified in this area have been relatively well developed, having numerous large, cordiform leaves, but also others elliptical, and all with short petioles. Moreover, 8 identified specimens had buds and flowers, their number being lower compared to those residing the segments III and IV. GPS: 47°00'22.9"N 21°58'44.7"E (47.006356, 21.979081).

DISCUSSIONS

Although the Băile Felix and Băile 1 Mai resorts own their fame to the thermal waters, the expansion of the two resorts has had and has a significant impact on the environment, which has intensified in the last two decades, when the landowners appeared during this period and started the over-exploitation of the geothermal water pool which is essential to ensure the survival of the "Peţea Creek" Natural Reserve. Under these conditions, in recent years, the local landowners have come to perceive the reserve as an obstacle, claiming constructions supports to authorities for their future development without taking into account other sustainable development solutions in order to preserve the Reserve of thermal waterlily. Moreover, the over-drillings (i.e. legally or illegally) of the thermal water pool performed to supply the resort areas and land use change, disrupted the natural underground circuit of thermal waters. The consequences of these actions decreased or even stopped the water flow of the sub-thermal mesothermal springs in the „Ochiul Mare" lake, with direct effects on the water level into the Peţea creek waterbody and accelerating the clogging and cooling of the waterbody. Finally, these actions lead to the drying of the main native habitat and historically recognized as the major habitat of the thermal waterlily, the "Ochiul Mare" lake.

The biologist Anna Marossy has predicted and warned authorities about the countless times regarding the risks threatening the reserve. She clearly indicated direct causes of the disaster, namely: the over-exploitation of thermal water pool through legal and illegal drilling, deforestation in the area causing a too rapid water flow in various valleys and channels [19]. Linc *et al.*, (2013) [18] stated that the lake is subject of an accelerated clogging process due to the fact that from the agriculturally cultivated slopes that define the creek basin, significant quantities of sediments reach the lake together with numerous pollutants used for agricultural crops (pesticides, insecticides, chemical fertilizers). All this has a strong impact on the biodiversity of the area. We add here the land use change under different forms such as: overlapping urban areas with non-urban areas, expansions of resort areas into the wild areas, expansion of constructed urban areas into the non urban areas as well as of agricultural areas into the wild areas. We add herewith the dams construction areas along the creek stream is another threat that fragmented the riparian habitats. All these aspects are in contradiction with the European regulatory framework for Natura 2000.

Cumulatively, all these aggression factors determined that, in December 2011, with the arrival of winter and the use of thermal water for heating installations, the water level and temperature in "Ochiul Mare" to drop drastically [3], and the reserve was saved by the supply of water brought by means of a hose from a nearby drilling. Specialists and custodians from that date of the reserve management unit (i.e. The Museum „Ţării Crişurilor") explained that the situation is due to illegal thefts of thermal water, accusing over 100 illegal drillings [34].

The situation encountered in December, 2011, repeated again in the same month of the next year,

when in two days after the installation of the cold time (correlated with the start of home heating based on thermal water), the water pressure dropped dramatically, so that it stopped the flow of the mesothermal sub-lacustrine springs of the "Ochiul Mare" lake. As a result, the lake dried up again almost completely on December 7, 2012, when, from the surface of 1 hectare of the lake, only 10 sqm of water cluster remained and having freezing limit temperature. The inevitable occurred on December 8 and 9, 2012, when the bottom of the lake froze. The spring of 2013 was a bit wetter, so the water level in the lake rose a little bit due to the rains and melting snow, but the summer and autumn that followed were droughty. Unfortunately, due to the drastic decrease of the water column in the lake and its freezing, the rhizomes of thermal waterlily did not survive. Therefore, 2013 was the first year without thermal waterlily specimens in the reserve, the lake having the appearance of a muddy puddle. The sub-lake spring from "Ochiul Mare" (the main source of thermal water supply of the lake [30]) ceasing its activity 10-11 months a year, the lake dried up, fact that led to the almost complete disappearance of the habitat 31A0* and to its replacement by dominant communities of invasive species of *Typha latifolia* and *Phragmites australis* [35]. Under these conditions, the main species of conservative interest, *Nymphaea lotus* L. var. *thermalis* (DC) Tuzson - the edifying species of this type of habitat, *Microcolpia parreyssii* (R.A. Philippi, 1847) and *Scardinius racovitzai* (Müller, 1958) have disappeared forever from their natural area - Lake Ochiul Mare from the "Peţea Creek" Reserve.

Given that the natural habitat of *Nymphaea lotus* var. *thermalis* (DC) Tuzson species was subjected to a high anthropogenic pressure, degrading rapidly, the thermal waterlily was forced to adapt to new habitats that would provide surviving conditions similar to those existing into its native habitat, the main one being the high waterbody temperature (over 25°C during the vegetation period). Regarding the optimal life requirements of the thermal waterlily, Olteanu - Cosma (1959, 1977 and 1991) [20-22] stated that for a good development, this plant species needs water with a temperature ranging between + 20 °C and + 34 °C, with a water depth ranging between 45 - 50 cm, good sun exposure, substrate rich in sludge, low concentration of $CaCO_3$ and low alkaline pH. Some of the abiotic factors of the plant in its native habitat are met by the Hidişel creek which, although not a creek with thermal springs, has the characteristics of such a creek in terms of its water temperature, as a result of thermal water discharges provided by the resort areas of the Băile Felix. The sub-mesothermal waters of the Hidişel creek, feeding the Peţea creek, provides conditions for hosting this thermophilic species, a fact proved on the occasion of the study necessary to draw up the Management Plan of the Peţea Creek Reserve [35], when several specimens were identified outside Ochiul Mare Lake; according to it, in 2015, it was reported the existence of several specimens of thermal waterlily in the basin of the lake "Ochiul Mare" inside the reserve, which does not happen today, due to the cessation of operation of the sub-lake spring.

During 2019, the entire Hidişel creek was surveyed on foot, and then the Peţea creek, after the confluence with Hidişel creek, in order to identify and locate in the field the specimens of thermal waterlily. All the areas in which this plant was identified presented similar physico-chemical characteristics, benefiting from strong lighting throughout the day and small temperature variations from one area to another. Also, in the areas where the waterlily was present, the water had a low flow rate, which favoured sedimentation and the deposition of a sediment layer of mud sufficient for the survival and good development of the plant. Although the areas of which the plant was encountered presented similar conditions, in segment III belonging to the Hidişel creek, an increased number of specimens of thermal waterlily was recorded, much higher than the other creek segments. This fact is explained by the anthropic intervention into the creekbed as well as on its banks due to anthropic interventions that changed the characteristics of the creek (i.e. in the creekbed being built a series of waterfalls and dams that slowed down the water speed, and on the banks the fragmentation of riparian habitats by vegetation removed). These actions determined the strong illumination of the water lustre throughout the day, and creating favourable conditions for the spreading and occurrence of thermal waterlily specimens. However, along with the thermal waters that flow into the Hidişel stream, important pollutant quantities of anthropic origin arrive [35]. Despite these shortcomings, the thermal waterlily was able to survive spontaneously and furthermore, to proliferate into these waterbodies. It is relevant to mention that the annual temperatures of the creeks are below (those of the lake "Ochiul Mare" belonging to the Peţea Creek Reserve. Moreover, these waters are loaded with organic pollutants of anthropic origin. Finally, it appears that these new habitats are providing the main needed biotic and abiotic factors range needed for the surviving of the species. A melange of natural and anthropic factors appears to work for the survival of the species that is threatened to extinction.

The cessation of the sublake spring to fed the Ochiul Mare lake from the "Peţea Creek" Natural Reserve from Băile 1 Mai [9], caused the gradual decline of the unique thermal ecosystem in our country, leading to the extinction of species of community interest for whose protection this reserve was established (*Nymphaea lotus* L. var. *thermalis* (DC) Tuzson - the edifying species of this habitat type, *Microcolpia parreyssii* (RA Philippi, 1847) and *Scardinius racovitzai* (Müller, 1958). Thermophilic species *N. l.* var *thermalis* is today threatened with extinction from the country and it can be considered as extinct for the Ochiul Mare lake inside the Natura 2000 site ROSCI0098 "Peţea Lake" where no specimens of

thermal waterlily were recorded, due to the fact that the Peţea creek is completely depleted of any flow of thermal water to the confluence with the Hidişel creek. However, in the vicinity of the Reserve still survived specimens that spread and occurred spontaneously in the wild, being those in the creekbeds of the Hidişel and Peţea streams, immediately after the confluence of the two creeks. Therefore, it is necessary to urgently implement programs for intensive propagation of the existing thermal waterlily specimens, developing re-introduction actions of the species, monitoring all specimens that survived *ex situ*, outside the Reserve area, in order to stabilize and increase the number of specimens of the population and to ensure the maintenance and possible increase of genetic variability of the species. There is a great need to cooperate with all decision factors for ensuring the survival of this unique species in our country, to restore its unique historical native habitat for Europe and at the World level, which is threatened today with extinction due to a high anthropogenic pressures. Moreover, it is also necessary to adopt fundamental methods to revitalize and restore the entire thermal habitat of the ROSCI0098 Peţea Lake community site, by regulating the rational use of geothermal water, by implementing specific systems for recycling and treatment of the sewage wastewater, but also by re-evaluating the exploitable thermal water reserves. Information campaigns regarding the negative anthropogenic impact on the reserve and on its specific species could contribute to raising awareness among members of the local community and tourists about the effects of their action and raising awareness and manifesting their civic spirit [15]. Accessing non-reimbursable funds, the realization and implementation of projects with the help of the European Union to local authorities, promoting the values that the reserve offers, could be steps towards the ecological restoration of the reserve, along with a drilling in the site to refuel and to fill the "Ochiul Mare" lake with thermal water, but also to build and maintain an artificial pool with earth banks along Hidişel creek, which will be supplied with thermal water coming from Băile Felix, where the specimens of thermal waterlily can be translocated and monitored.

A first step towards the conservation of *Nymphaea lotus* var. *thermalis* (DC) Tuzson species consisted of starting a research program as a cross-border project ROHU29 [36], a project through which the conservation of thermal waterlily in the laboratory was considered and which, starting with 2020, also tests the conservation of the species in nature, in artificial environments that mimic natural conditions. But these steps need to be complemented and improved by similar ones, which requires further research.

Although field studies have led to the identification of a number of approximately 166 specimens of thermal waterlily spontaneously grown in the creekbeds of the two investigated streams, their number being higher than that reported by the study conducted in 2015 [35], when 70 stalks of thermal waterlily were identified (33 in Peţea Lake and 37 in Peţea creek, after its confluence with Hidişel creek), the risk of loosing the *N. l.* var. *thermalis* endemic species from the natural environment remains alarmingly high, the plant being vulnerable to pollution, floods, but also anthropogenic factors. The ecological restoration of the reserve would increase the chances of survival of the thermal waterlily, given that the literature confirms that the seeds of this species are particularly resistant, having great vitality, resisting drought for years and germinating even after 20 years [20, 21].

The results of this research reveal and prove the great ability of the species *Nymphaea lotus* L. var. *thermalis* (DC) Tuzson to adapt for surviving being able to disperse and occupy similar habitats in the same climatic niche. The historical native habitat can be restored and there is still time to recover an extinct habitat for Romania as well as for the European countries. Dramatic pressures of anthropic nature as well as due to climate change effects are acting already in its entire climatic niche and therefore urgent needs for action are to have the chance to ensure the long-term survival of the species and its natural and historical habitats.

Acknowledgments. This study was carried out within the ROHU29 project (*Conservation and protection of ecosystems endangered by lack of thermal and freshwater in crossborder area*).

REFERENCES

[1] Antofie, M.M., (2012): Conservarea biodiversităţii - consideraţii politice şi legislative. Lucian Blaga University Publishing House, 275 p.

[2] Bland, L.M., Nicholson, E., Miller, R.M., Andrade, A., Carré, A., Etter, A., Ferrer-Paris, J.R., Herrera, B., Kontula, T., Lindgaard, A., Pliscof, P., Skowno, A., Valderrabano, M., Zager, I., Keith, D.A., (2019): Impacts of the IUCN Red List of Ecosystems on conservation policy and practice. Conservation Letters, 12(5): e12666.

[3] Blidar, C.F., Tripon, I.M., Ilea, C., (2019): *In vitro* conservation of genetic resources of *Nymphaea lotus* var. *thermalis* (D.C.) Tuzs., an endangered plant species. Romanian Biotechnological Letters, 24(3): 448-457.

[4] Borsch, T., Hilu, K.W., Wiersema, J.H., Lohne, C., Barthlott, W., Wildes, V., (2007): Phylogeny of *Nymphaea* (Nymphaeaceae): Evidence from substitutions and microstructural changes in the chloroplast trnT-trnF region. International Journal of Plant Sciences, 168(5): 639-671.

[5] Brooks, T.M., Pimm, S.L., Akçakaya, H.R., Buchanan, G.M., Butchart, S.H., Foden, W., Hilton-Taylor, C., Hoffman, M., Jenkins, C.N., Joppa, L., Li, B.V., Menon, V., Ocampo-Penula, N., Rondini, C., (2019): Measuring terrestrial area of habitat (AOH) and its utility for the IUCN red list. Trends in ecology & evolution, 34(11): 977-986.

[6] Burescu, P., Csep, N., Tofan, T., (2002): La végétation du lac à nénuphar thermal de Băile 1 Mai - Oradea. Proceedings of the Symposium „Studies of the Biodiversity - West Romania Protected Areas". Orizonturi Universitare Publishing House, pp. 98-102.

[7] Burescu, P., Cheregi, V., Csep, N., (2004): Vegetaţia lacului cu nufăr termal de la Băile 1 Mai - Oradea. Analele Universităţii Craiova, Facultatea de Horticultură, 7: 149-153.
[8] Chytrý, M., Hájek, M., Kočí, M., Pešout, P., Roleček, J., Sádlo, J., Šumberová, K., Sychra, J., Boublík, K., Douda, J., Grulich, V., Härtel, H., Hédl, R., Lustyk, P., Navrátilová, J., Novák, P., Peterka, T., Vydrová, A., Chobot, K., (2019): Red List of Habitats of the Czech Republic. Ecological Indicators, 106: 105446.
[9] Cohuţ, I., Paal, G., (2017): Recviem pentru Rezervaţia Naturală „Pârâul Peţa" de la Băile 1 Mai. Nymphaea Folia naturae Bihariae XLIV, pp. 77-92.
[10] Danciu, V.M., (2004a): Aspecte privind starea calităţii apei la rezervaţia naturală „Pârâul Peţea" de la Băile 1 Mai. Oradea, Nymphaea, 31: 35-53.
[11] Danciu, V.M., (2004b): Rezervaţia naturală de la Băile 1 Mai la începutul mileniului III. Nymphaea, Oradea, 31: 111-131.
[12] Diaconeasa, B., Popa, D., (1964): Problema relictă a lotusului (*Nymphaea lotus* L. var. *thermalis* (DC) Tuzs.) şi a lacului termal de la Băile 1 Mai, în lumina analizelor microstratigrafice. Contribuţii Botanice, pp. 135-140.
[13] Doniţă, N., Popescu, A., Paucă-Comănescu, M., Mihăilescu, S., Biriş, I.A., (2005): Habitatele din România. Tehnică Silvică Publishin House, Bucharest, 442 p.
[14] Gafta, D., Mountford, J.O., (2008): Manual de interpretare a habitatelor Natura 2000 din România. Risoprint Press, Cluj-Napoca, 101 p.
[15] Ilieş, D.C., Buhaş, R., Ilieş, A., Morar, C., Herman, G., (2015): *Nymphaea lotus* var. *thermalis* (Pârâul Peţea Natural Reserve), brand near extinction of the Băile Felix – Băile 1 Mai (Romania) SPA Tourism System. GeoJournal of Tourism and Geosites, 15(1): 107-117.
[16] Jalas, J., Suominem, J., (1989): Atlas Flora Europaeae. Vol. 8. Nymphaeaceae to Ranunculaceae. Biological Society of Finland, Vanamo, 261 p.
[17] Laczko, L., Lucacs, B.A., Mesterhazy, A., Molnar, V.A., Sramko, G., (2019): Is *Nymphaea lotus* var. *thermalis* a Tertiary relict in Europe? Aquatic Botany, 155: 1-4.
[18] Linc, R., Dincă, I., Bucur, L., (2013): Intruziunea umană în areale protejate din judeţul Bihor. Rezervaţia Naturală Pârâul Peţa. pp. 627-637. In: Vert, C. (ed.): Conferinţa Naţională a Societăţii de Geografie din România, ediţia a X-a, cu tema „Geografia - ştiinţă a întregului teritorial", 22-24 mai, 2013, Timişoara, Eurobit Press, Timişoara.
[19] Marossy, A., (1976): Factori antropici ce au modificat echilibrul ecologic al rezervaţiei naturale „Pârâul Peţea". Nymphaea, Oradea, 4: 261-272.
[20] Olteanu-Cosma, C., (1959): Biologia şi ecologia plantei *Nymphaea lotus* L. var. *thermalis* (D.C.) Tuzs. de la Băile 1 Mai Oradea. Ocrotirea naturii, 4: 63-88.
[21] Olteanu-Cosma, C., (1977): Biologia nufărului termal *Nymphaea lotus* L. var. *thermalis* (D.C.) Tuzs., de la Băile 1 Mai – Oradea. Nymphaea, 5: 365-380.
[22] Olteanu-Cosma, C., (1991): Nufărul termal de la Băile 1 Mai (Oradea) solicită ocrotire. Muzeul Ţării Crişurilor Oradea, 35 p.
[23] Oneţ-Tripon, I.M., (2014): Iniţierea de vitroculturi de *Nymphaea lotus* L. var. *termalis* (D.C.) Tuzs., în vederea refacerii populaţionale din cadrul Rezervaţiei Naturale „Pârâul Peţea". Dissertation Thesis, University of Oradea, Romania, 99 p.
[24] Pax, F., (1905): Die fossile flora von Ganocz bei Poprad. Beiblatt Zu den Novenytani Kozlemenyek, 4: 19-59.
[25] Pop, E., (1976): Specii relicte în flora României. pp. 106-111. In: Beldie, A., Morariu, I. (Eds.): Flora Republicae Socialisticae Romania, Vol. 13. Editio Academiae Republicae Socialisticae Romania.
[26] Poczai, P., Matyas, K.K., Szabo, I., Varga, I., Hyvonen, J., Cernak, I., Gorji, A.M., Decsi, K., Taller, J., (2011): Genetic variability of thermal *Nymphaea* (Nymphaeaceae) population based on ISSR markers: implications on relationships, hybridization and conservation. Plant Molecular Biology Report, 29: 906-918.
[27] Rivers, M.C., Beech, E., Bazos, I., Bogunić, F., Buira, A., Caković, D., Carapeto, A., Carta, A., Cornier, B., Fenu, G., Fernandes, F., Fraga, P., Garcia Murillo, P.J., Lepší, M., Matevski, V., Medina, F.M., Menezes de Sequeira, M., Meyer, N., Mikoláš, V., Montagnani, C., Monteiro-Henriques, T., Naranjo Suárez, J., Orsenigo, S., Petrova, A., Reyes-Betancort, J.A., Rich, T., Salvesen, P.H., Santana López, I., Scholz, S., Sennikov, A., Shuka, L., Silva, L.F., Thomas, P., Troia, A., Villar, J.L., Allen, D.J., (2019): European Red List of Trees. Cambridge, UK and Brussels, Belgium: IUCN, viii + 60 p.
[28] Román-Palacios, C., Wiens, J.J., (2020): Recent responses to climate change reveal the drivers of species extinction and survival. Proceedings of the National Academy of Sciences, 117(8): 4211-4217.
[29] Söllösi, R.S., (2015): Popularea unui habitat termal artificial cu specia *Nymphaea lotus* L. var. *termalis* (D.C.) Tuzs. Oradea. Dissertation Thesis, University of Oradea, Romania, 83 p.
[30] Şoldea, V., (2003): Peţa şi nufărul termal. Universităţii din Oradea Press, 98 p.
[31] Tutin, T.G., Webb, D.A., (1992): *Nymphaea* L. pp. 246-247. In: Titin, T.G., Burges, n.a., Chater, A.O., Edmondson, J.R., Heywood, V.H., Moore, D.M., Valentine, D.H., Walters, S.M., Webb, D.S., (Eds.): Flora Europaea. Volume 1: Psilotaceae to Platanaceae. 2nd Edition. Cambridge University Press.
[32] www1: www.iucn.org accessed in February, 2020.
[33] www2: http://www.magazinsalajean.ro/divertisment/lacul-cu-nuferi-termali-de-la-1-mai-reconstituit-in-incinta-gradinii-botanice-jibou accessed in February, 2020.
[34] www3: https://www.ebihoreanul.ro/stiri/ultima-or-31-1/rezervatia-din-baile-1-mai-distrusa-de-hotii-de-apa-geotermala-99620.html accessed in February, 2020.
[35] ***1 (2016): Plan de Management al Sitului de Importanţă Comunitară ROSCI0098. 233 p. (http://www.mmediu.gov.ro/app/webroot/uploads/files/2016-04-11_PM ROSCI0098_Petea.pdf accessed in February, 2020).
[36] ***2 (2018-2020): Conservation and protection of ecosystems endangered by lack of thermal and freshwater in crossborder area – ROHU-29 (AQUARES). Link: https://aquares.ro/ accessed in February, 2020.

Received: February 29, 2020
Accepted: July 21, 2020
Published Online: July 29, 2020
Analele Universităţii din Oradea, Fascicula Biologie
http://www.bioresearch.ro/revistaen.html
Print-ISSN: 1224-5119
e-ISSN: 1844-7589
CD-ISSN: 1842-6433
University of Oradea Publishing House