Ecophysiological characteristics of aquatic carnivorous plants: A review

Lubomír Adamec[*]

Abstract

The contribution discusses an outstanding group of aquatic macrophytes: aquatic carnivorous plants. Eight species of aquatic carnivorous plants grow naturally in the Danube basin. Most of them are strongly or critically endangered species in single countries, and exhibit a gradual decline. The basis for their protection is detailed knowledge of their biology (i.e. ecological characteristics of their habitats, their ecophysiology including photosynthesis, mineral nutrition) that is summarised in the present contribution.

Introduction

About 50 species of the genera *Aldrovanda* (waterwheel plant, Droseraceae) and *Utricularia* (bladderwort, Lentibulariaceae) are submerged aquatic or amphibious carnivorous plants (Juniper et al., 1989; Taylor, 1989; Guisande et al., 2007). Unlike the majority of aquatic non-carnivorous plants, all aquatic carnivorous species are strictly rootless and, therefore, they can take up mineral nutrients for their growth from the ambient medium and from captured prey *only* via their shoots. Nevertheless, they fulfil all three principal functional criteria generally placed on carnivorous plants (Adamec, 1997a; Rice, 2011). Traps of aquatic species exhibit rapid movements, which are among the fastest within the plant kingdom, and represent fascinating objects for biological studies (Juniper et al., 1989; Vincent et al., 2011a).

On the territory of all the Danube River countries (Germany, Austria, Slovakia, Hungary, Serbia, Bulgaria, Romania) or countries within the watershed of the Danube River (Czech Republic, Slovenia, Croatia), eight aquatic carnivorous plant species occur which are possibly distributed over the major part of Europe (see Figs 1–3). These species are: *Aldrovanda vesiculosa* L., *Utricularia australis* R.Br., *U. vulgaris* L., *U. intermedia* Hayne, *U. ochroleuca* s. str. Hartm., *U. stygia* Thor, *U. minor* L. and *U. bremii* Heer (Casper and Krausch, 1981; Thor, 1988; Taylor, 1989). Taxonomically, the newly described species, *U. stygia* Thor,

---

[*] Corresponding author: adamec@butbn.cas.cz

may not be accepted by all national European floras as it was separated from a rather polymorphic taxon, *U. ochroleuca* s. lato Hartmann as late as in 1988 by Thor (1988; see also Płachno and Adamec, 2007). What is common to all these species? Although they commonly follow the streams of larger rivers including the Danube River, they never grow in running waters. They are common only in standing waters such as backwater pools, oxbows, in river floodplains or in larger river basins mainly in lowlands. The other habitats are associated more with peatbogs and fens. All these European species grow in dystrophic (humic) wetlands which have usually greatly deteriorated or even vanished from some parts of Europe during the last 150 years (e.g. Adamec, 1995). Consequently, out of the 8 species, most species have become strongly or critically endangered or even extinct (particularly *Aldrovanda vesiculosa*; Adamec, 1995) in most of the Danube River countries. These carnivorous species represent a specific ecophysiological group with peculiar traits (e.g. Adamec, 1997a; Guisande et al., 2007; Ellison and Adamec, 2011). Yet as compared to their terrestrial counterparts, they have always been understudied or even neglected (cf. Juniper et al., 1989; Adamec, 1997a; Ellison, 2006) as the main focus of studies was always directed to the processes found in terrestrial, rather than aquatic, carnivorous species (cf. Juniper et al., 1989; Adamec, 1997a). Moreover, mainly due to methodological problems, the ecophysiological study of the latter group has lagged much behind that of the former group. As both ecological groups of carnivorous plants are rather dissimilar in their principal morphological and physiological features and also *per se* (submerged *vs.* terrestrial life *sensu* Colman and Pedersen, 2008), it is reasonable and justifiable to distinguish between these groups when making ecologically or physiologically oriented reviews, as did Ellison and Adamec (2011).

For the above reasons, aquatic carnivorous plants, particularly the European species, are reviewed here from an ecophysiological point of view. The ecology of their habitats and characteristics of their growth traits, photosynthesis, mineral nutrition, ecophysiological characteristics of *Utricularia* traps and investment in carnivory in *Utricularia* are here discussed using recent literature.

Ecological characteristics of habitats of aquatic carnivorous plants

Aquatic carnivorous plants usually grow in shallow standing or slowly flowing humic (i.e. dystrophic) waters, often together with vascular aquatic non-carnivorous plants. It can be assumed that the carnivorous plant group also tolerates very high concentrations of humic acids and tannins (very dark waters), while the majority of non-carnivorous plants do not. The total concentration of humic acids and tannins at these sites commonly ranges from 5–20 mg.l^{-1} and may even extend to 60 mg.l^{-1} (Adamec, 2007a, 2008b). Most rootless aquatic carnivorous plants prefer to grow in mildly or strongly dystrophic, oligo-mesotrophic waters. In contrast most submerged, rooted aquatic plants, avoid growing in such waters or at least in strongly dystrophic waters (Adamec, 2012a). The reasons for this avoidance are probably based on the difficulty of submerged plants to root in anoxic, loose organic soil composed of reed, sedge litter or peat. Other unfavourable ecophysiological effects of dark dystrophic water are shading, cooling, excessive concentrations of humic acids and tannins, and low pH. Evidently, all eight European aquatic carnivorous plant species are well adapted to grow even in strongly dystrophic waters (Kamiński, 1987; Navrátilová and Navrátil, 2005; Adamec, 2007a, 2008b, 2009).

Typical dystrophic waters are usually poor in minerals such as N (NH_4^+; NO_3^-), P (the concentration of both is commonly 5–20 µg.l^{-1}), and also K (<0.5 mg.l^{-1}). If the site is not impacted by human activity, the concentrations may even be 5–10 times lower (see Adamec, 1997a; Guisande et al., 2007). The concentrations of SO_4^{2-}, Ca, Mg, and Fe, however, are usually >1 mg.l^{-1} (Guisande et al., 2007) and do not limit plant growth. A partly decomposed, nutrient-poor litter of reeds and sedges usually accumulates in these waters. The slowly decomposing litter gradually releases mineral nutrients, humic acids, tannins, and CO_2. Hence, the waters are usually rather high in free CO_2 concentration ([CO_2]) (0.1–1 mM; Adamec, 1997a,b, 2007a, 2008b, 2009, 2010a). A high [CO_2] >0.15 mM was found to be the principal water chemistry factor supporting vigorous growth and propagation of stenotopic *Aldrovanda vesiculosa* (Adamec, 1999). The same relationship between [CO_2] and growth holds true also in aquatic non-carnivorous plants. In addition, reduced concentrations of dissolved oxygen, within the range 0–12 mg.l^{-1}, were found at many sites supporting aquatic carnivorous plants (Adamec, 1997b, 1999, 2007a, 2010a; Guisande et al., 2000, 2004; Adamec and Kovářová, 2006; Giural and Rougier, 2007). Moreover, the last authors found a marked daily oscillation of [O_2]. The majority of aquatic carnivorous species usually grow in soft to moderately hard (total alkalinity 0.2–2 meq.l^{-1}), acid or neutral waters (pH 5–7.5), but some temperate-zone species may also grow in hard and alkaline waters (pH 8–9.3; see Adamec, 1997a, 2009). Two wide-spread aquatic species, *Utricularia australis* and *U. minor*, were able to grow in a very wide range of pH, the former species within 4.3–9.2, the latter within 3.5–9.3 (cf. Adamec, 1997a, 2008b, 2009; Navrátilová and Navrátil, 2005). Thus, at least in eurytopic species, water pH alone is not important for their field growth.

When considering photosynthetic cost-benefit relationships, Givnish et al. (1984) postulated that, for terrestrial carnivorous plants, carnivory is only beneficial in nutrient-poor, moist, and unshaded habitats. In contrast to terrestrial habitats, many aquatic carnivorous species sustain well also under conditions of very low irradiance (<5% of that in the open; Adamec, 2008b) which complies with irradiation conditions also relevant for non-carnivorous aquatics.

Growth characteristics of aquatic carnivorous plants

Aquatic carnivorous plants markedly differ morphologically and ecophysiologically from terrestrial species: they are always rootless, floating freely below the water surface, or are weakly attached to loose sediments as submerged or partly amphibious types (Taylor, 1989; Guisande et al., 2007). Most species have a linear and modular shoot structure consisting of regularly changing nodes with filamentous leaves and tubular, fragile internodes. In some species, the leaves are arranged in true whorls. Only some species (e.g. *Utricularia volubilis*) are rosette-shaped plants. The majority of linear-shoot species have homogeneous (monomorphic), non-differentiated green shoots bearing traps (e.g. *Aldrovanda vesiculosa*, *Utricularia vulgaris*, *U. australis*). Several species (e.g. *Utricularia intermedia*, *U. ochroleuca*) have dimorphic shoots differentiated into green photosynthetic ones (usually bearing only a few or no traps) and pale carnivorous (trapping) ones with many traps. These species are intermediate in body plan between the aquatic *Utricularia* species with monomorphic shoots and terrestrial species (e.g. *U. uliginosa*, *U. livida*) with aboveground, flat green leaves and belowground, pale carnivorous shoots bearing traps (Taylor, 1989).

Aquatic carnivorous plants exhibit some growth characteristics differentiating them distinctly from all terrestrial carnivorous species (Adamec, 1997a; Ellison and Adamec, 2011). Firstly, adult plants maintain the length of the main shoot approximately constant throughout the season: they show very rapid apical shoot growth but their basal shoot segments age and die at about the same rate (‘conveyer-belt’ shoot growth system). Thus, the new biomass is allocated into branching or flowering only. Under favourable conditions, the apical shoot growth rate of aquatic species with linear shoots was 1.0–1.2 whorls per day in *Aldrovanda* (Adamec, 2000; Adamec and Kovářová, 2006; Adamec et al., 2010a) and even greater in field-grown *U. vulgaris* (1.4–2.8 nodes per day, Friday, 1989), *U. australis* (2.8–4.2 nodes per day, Adamec and Kovářová, 2006; Adamec, 2009) or *U. intermedia* and *U. stygia* (1.9–2.1 nodes per day, Adamec, 2010a). Surprisingly, the apical growth rate of *U. australis* was 2.9–4.2 nodes per day also in an oligotrophic water, though the relative growth rate (RGR) was zero (Adamec, 2009). Thus, very high apical growth rate in aquatic species may not be connected with high RGR. It is also an important strategy in competition with epiphytic algae, which usually densely cover their older shoots (Friday, 1989). In the slowly growing subtropical *U. purpurea*, however, the apical growth rate was only 0.25 nodes per day (Richards, 2001). Unlike all terrestrial carnivorous plants, representing typical S-strategy, high RGRs were reported for a few aquatic species under favourable conditions which rather suggests their R-strategy. The doubling time of biomass (i.e. $\log_e 2/RGR$) in field-grown *Aldrovanda* was only 8.4–21.5 days (Adamec and Kovářová, 2006) or 12.9–23.0 days (Adamec, 1999) and 12.8 days in an outdoor culture (Adamec, 2000), 9.1–33.2 days in field-grown *U. australis* (Adamec and Kovářová, 2006), 6.6–9.2 days in field-grown *U. intermedia* and *U. stygia* (Adamec, 2010a), or 12.4–23.1 days in greenhouse-grown *U. vulgaris*, *U. geminiscapa*, and *U. purpurea* (Pagano and Titus, 2004, 2007). Pagano and Titus (2007) proved about a two- to threefold RGR increase in the three *Utricularia* species as a result of $[CO_2]$ increase. Comparable values between 6.4–34.7 days were recorded by Nielsen and Sand-Jensen (1991) for aboveground biomass in 12 rooted submerged, non-carnivorous species. In contrast, the corresponding values for terrestrial species of the genera *Drosera*, *Genlisea*, and *Sarracenia* are much greater (21–104 days, mean about 35–40 days), suggesting their considerably slower growth (Adamec, 2002, 2008c; Farnsworth and Ellison, 2008; Ellison and Adamec, 2011).

The very rapid growth of rootless aquatic carnivorous plants in nutrient-poor habitats requires ecophysiological adaptations that enable the plants to access the highly limited supplies of mineral nutrients from the water. These adaptations include carnivory, efficient nutrient re-utilization from senescing shoots, and a very efficient nutrient uptake from water (Kamiński, 1987; Kosiba, 1992; Friday and Quarmby, 1994; Adamec, 2000, 2008a,b; Englund and Harms, 2003).

Another important growth characteristic, which facilitates propagation, is the production of shoot branches which subsequently develop into separate, new individuals (Adamec, 1999). The branches of *Aldrovanda* always develop into new plants, but in some aquatic *Utricularia* species, initiated branches may not develop beyond the early stages (Adamec, 2009, 2011a). It is generally accepted that the number of branches per shoot or the proportion of the total branch biomass to the total plant biomass are the principal growth parameters to be used as a criterion for plant vigour and propagation rate and therefore also reflect the suitability of a habitat for plant growth (Kamiński, 1987; Adamec, 1999, 2000, 2009, 2011a; Adamec and Kovářová, 2006). In some aquatic carnivorous species studied, branching rate (number of internodes between two branches) was regular under optimum conditions and species specific (see Table 1). The data for *U. australis* indicate that, although branching rate is genetically fixed, it is under ecological regulation. Moreover, in *U. australis* grown under optimum ecological field conditions, regular branching rate of apically initiated branches was disturbed as a result of frequent initiation of branches from axillary buds along the main shoots (Adamec, 2011a). In such cases, the apparent branching rate was commonly as high as 1–3 internodes per branch. If branching rate is divided by the apical shoot growth rate, this parameter, the branching frequency, characterises the real time involved in initiating successive branches on the shoot. Thus, branching frequency is a good criterion for RGR (Adamec and Kovářová, 2006). The authors found a similar branching frequency (4.7–5.5 days per branch) for field-grown *Aldrovanda* and *U. australis*. In both species, however, competitive processes occurred between the production of new whorls and branches: the apical shoot growth rate of branches in nodes per day was only about 70–83 % of that of the main shoot (Adamec, 1999, 2011a). Evidently, knowledge of branching traits is crucial for understanding the growth characteristics in aquatic *Utricularia* species (Adamec, 2011a).

Table 1. Review of branching rates (number of internodes between two branches) in aquatic carnivorous plants from the literature
Means ±SE intervals are shown

Species	**Shoot type**	**Branching rate**	**Reference**
A. vesiculosa	Main shoot	6.2 (range 3–11)	Adamec (1999)
A. vesiculosa	Main shoot	5.3±0.2	Adamec & Kovářová (2006)
U. stygia	Photosynthetic	12.2±0.4	Adamec (2007a)
U. stygia	Carnivorous	6.7±0.2	Adamec (2007a)
U. stygia	Photosynthetic	12.2±0.2	Adamec (2010a)
U. intermedia	Photosynthetic	16.8±0.4	Adamec (2007a)
U. intermedia	Carnivorous	5.9±0.1	Adamec (2007a)
U. intermedia	Photosynthetic	17.7±0.4	Adamec (2010a)
U. australis	Main shoot	22.1±1.2; 10.5±0.4	Adamec (2009)
U. australis	Main shoot	14.5±1.2; 15.1±1.7	Adamec & Kovářová (2006)

Trap ecophysiology of aquatic *Utricularia*

Water flow

Utricularia suction traps are hermetically closed bladders functioning on the basis of negative pressure (Fig. 4; e.g. Sydenham and Findlay, 1975; Juniper et al., 1989; Guisande et al., 2007; Vincent et al., 2011a; Singh et al., 2011). The trap size in aquatic *Utricularia* species is within the range of 1–6 mm, rarely up to 12 mm (Taylor, 1989); these are larger than the traps of terrestrial species in this genus. They are the smallest within all carnivorous plants, but they are considered the most sophisticated traps functionally (Juniper et al., 1989). Contrary to the traps of other species, solutes and suspended particles sucked-in from the ambient water are retained hermetically in the lumen until the trap is senescent. Four types of glands (hairs) occur inside or outside the traps;

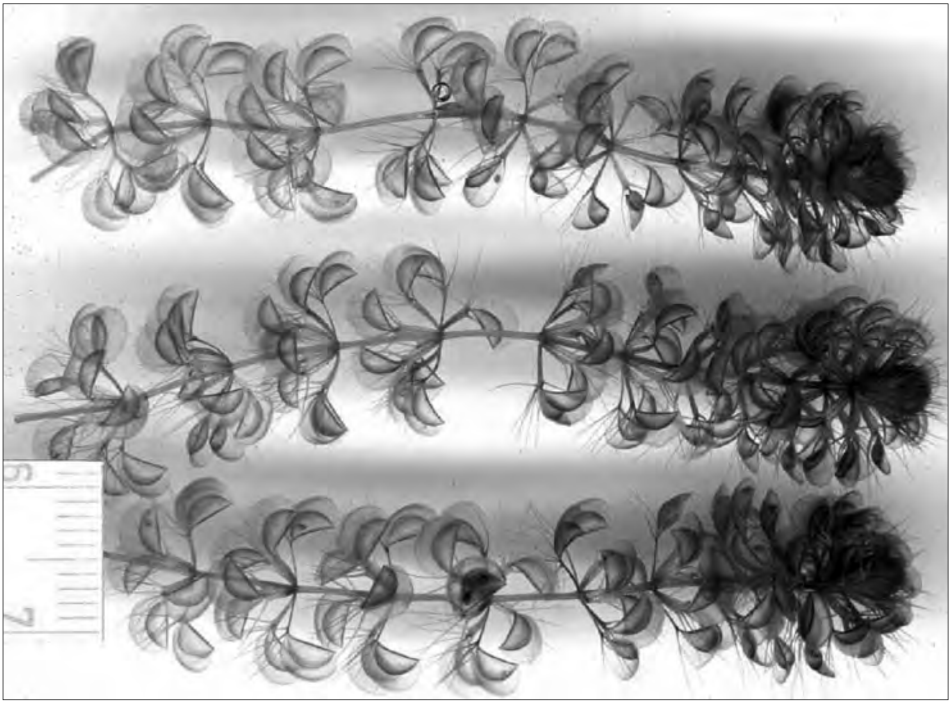

Figure 1. Monomorphic shoots of *Aldrovanda vesiculosa* bearing regular whorls of leaves with snapping traps. Turions (winter buds) are formed at the shoot apices. Scale bars represent 1 mm. Photo: Lubomír Adamec

abundant and large internal quadrifid and bifid glands are principal for trap physiology (see Juniper et al., 1989; Guisande et al., 2007).

Almost all of the knowledge of the biophysical nature of *Utricularia* trap functioning was published in a series of only four studies (Sydenham and Findlay, 1973, 1975; Sasago and Sibaoka, 1985a,b). As it follows from these 'classic' studies a negative pressure of about −16 kPa (i.e. 1/7 of bar) relative to the ambient water is maintained inside a fully reset trap. When a prey species touches sensory hairs situated on the trap door it opens, the small animal is aspirated into the trap and the door closes again. This process of firing is completed within 10–15 ms and is the most rapid plant movement known (Sydenham and Findlay, 1973). Immediately after firing, the negative pressure inside the trap drops to zero, but is soon restored by the rapid removal of c. 40% of the water from the fired trap until the original concave shape is reached. This aspect of trap resetting lasts about 25–30 min and the trap is ready to fire again; the full resetting process lasts much longer. These four studies also found that water is pumped out of the traps by an active process associated with a consumption

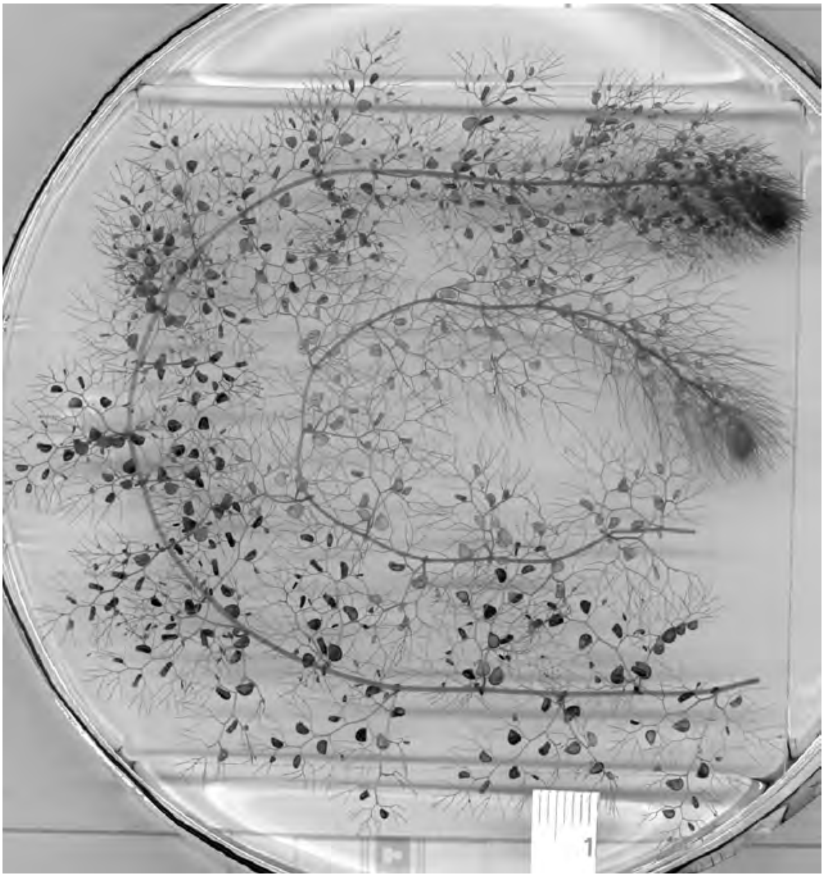

Figure 2. Monomorphic shoots of *Utricularia australis* have photosynthetic function but bear hundreds of suction traps. Scale bars represent 1 mm. Photo: Lubomír Adamec

of metabolic energy, ATP. When inhibitors of aerobic respiration were added to the trap fluid, the water pumping and trap narrowing processes were very markedly blocked: bifid glands attached close to the trap door take part in water pumping and that water is exuded from the pavement epithelium close to the door. Electrophysiological measurements have led to the hypothesis (Sydenham and Findlay, 1975; Sasago and Sibaoka, 1985a) that Cl^- ions are actively taken up from the trap fluid by the bifid glands and, due to their movement, osmotically drag water molecules. Monovalent cations (Na^+, K^+) accompany the Cl^- fluxes, while bivalent cations (Ca^{2+}, Mg^{2+}) rather inhibit them. The second part of the water pathway is still veiled in mystery as it appears that the water is expelled from the cells of the pavement epithelium by the turgor pressure through a very leaky plasmalemma (for solutes <600 Daltons; Sydenham and

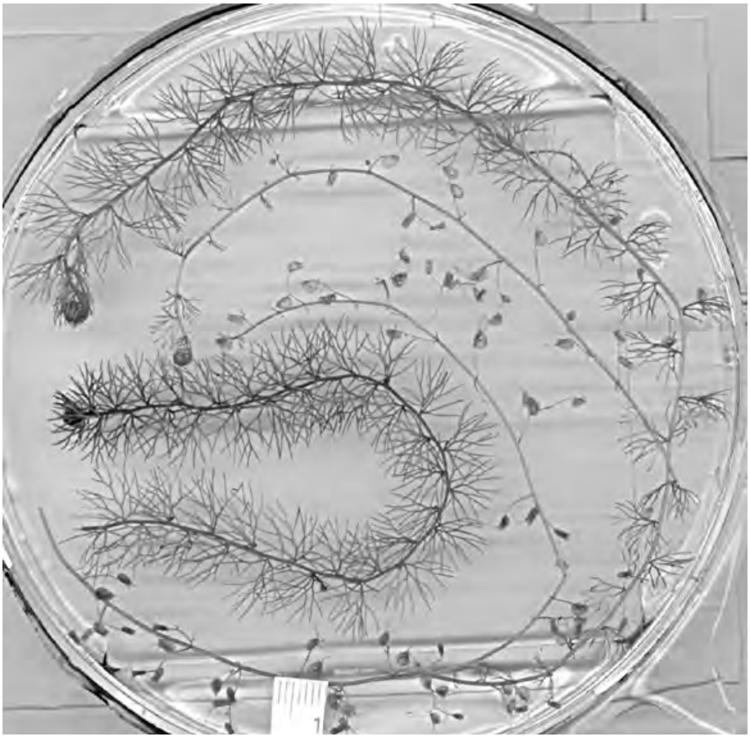

Figure 3. Dimorphic shoots of *Utricularia ochroleuca* s. str. are differentiated in green photosynthetic shoots with a few traps and pale carnivorous shoots bearing dozens of traps. Scale bars represent 1 mm. Photo: Lubomír Adamec

Findlay, 1975). Traps can also pump out water in moist air or when immersed in a liquid paraffin oil (Sasago and Sibaoka, 1985a; Adamec, 2012c).

In 2011, several papers were published which crucially changed and specified our understanding of the biophysical processes associated with *Utricularia* trap firing. The unique use of a high-speed camera has revealed that the essence of the reversible trap door opening and closing is the rapid curvature inversion called buckling and unbuckling (Joyeux et al., 2011; Singh et al., 2011; Vincent et al., 2011a,b; Vincent and Marmottant, 2011). As a result of the negative pressure inside the trap, the curved trap door is metastable and any tiny mechanical perturbation (mediated by prey through the trigger hairs) triggers the curvature inversion (buckling) leading to trap opening and inflation. When the negative pressure is gone the trap door spontaneously returns to its initial curvature, closing the door again. Thus, the negative pressure inside the traps is an essential component part of the opening and closing mechanism. Moreover, the complete process of trap firing was found to last only 2–5 ms in several species.

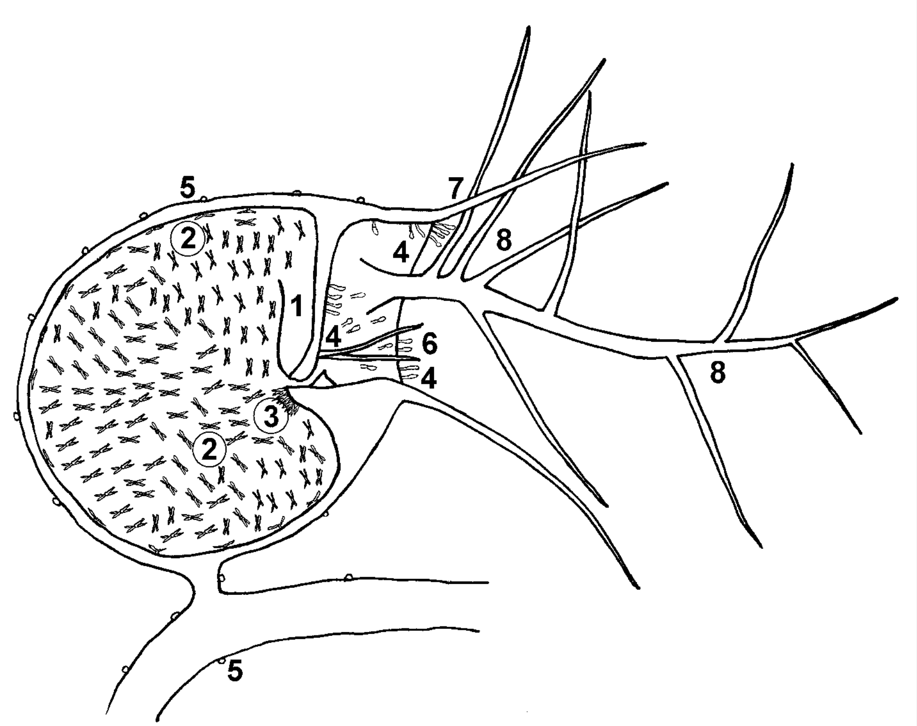

Figure 4. Schematic longitudinal section through a trap of *Utricularia* with glands and other structures (modified by J. Vrba, from Juniper et al., 1989). 1 – trap door, 2 – quadrifid glands, 3 – bifid glands, 4 – stalked mucilage glands, 5 – spherical sessile glands, 6 – trigger hairs, 7 – rostrum, 8 – antennae

New important challenges for *Utricularia* trap ecophysiology have very recently arisen from the discovery of spontaneous firing in *Utricularia* traps. Using both a high-speed camera for intact shoots (Vincent et al., 2011a,b) and a linear position sensor for excised traps (Adamec, 2011b,c), a more or less regular trap firing without any mechanical stimulation was confirmed in several aquatic *Utricularia* species and in two trap age categories. Spontaneous firings occurred 0.3–2.4 times during the 1-day resting period and the mean time between two spontaneous firings varied between 5–16 h. Quantitatively (trap thickness increase due to firing, resetting rate), spontaneous firings resembled mechanically stimulated ones (Adamec, 2011b). In two *Utricularia* species, Vincent et al. (2011b) subdivided the traps after the time patterns of spontaneous firings into metronomic, random and bursting. It is possible that a spontaneous firing occurs as a ‘safety valve’ protecting the integrity of the trap door. It was also found that the traps pumped water out after firing for at least 5–10 h until a steady-state was attained (Adamec, 2011b). Moreover, as opposed to Sydenham and Findlay (1973) who postulated a concept of an internal negative pressure sensor regulating water pumping in the trap, the most recent data (Adamec, 2011c) suggest a permanent pumping out of water from the traps. These data are based mainly on the fact that no lag-period in water pumping from the trap was detected within the first 2 s after trap firing, which occurred after a 3-h resetting period. This can mean that either the water permanently recirculates through some leaks under the trap door or the mechanism of water pumping becomes thermodynamically inefficient at high negative pressure though it runs permanently.

Taxonomic aspects and basic functional characteristics of trap firing (magnitude of firing and resetting rate as 'trap efficiency') were compared in 13 aquatic *Utricularia* species of the mean trap length between 2.0 to 5.3 mm (Adamec, 2011c). On the absolute scale, the 13 species differed considerably in their firing and resetting rates. Significant interspecific differences were also found in the magnitude of firing (in total 3.7–4.2 times) and resetting rates (10–24 times) per unit trap thickness or length. Overall, traps of *U. australis*, *U. stellaris* and *U. inflata* showed the greatest firing and resetting rates. The relative magnitude of firing per unit trap thickness or length showed a highly significant negative correlation with both trap thickness and length and the same also held for the relative resetting rates. Smaller and narrower traps are thus relatively more effective at trap firing and resetting than larger traps. Neither firing nor resetting characteristics were significantly different between unfed and prey-fed traps of *U. reflexa* and this was also true for the occurrence of spontaneous firings.

Prey digestion

A pH value of 5.0 ± 0.1 occurred in trap fluid in four aquatic *Utricularia* species independently of digesting of prey (Sirová et al., 2003). This suggests that trap fluid pH is well regulated. Though several types of hydrolytic enzymes were described from *Utricularia* traps using biochemical and cytochemical methods (protease, esterase, acid phosphatase; see Juniper et al., 1989), only the activity of phosphatases at pH 4.7 was determined as significant by an *in situ* analysis of empty trap fluid in four aquatic *Utricularia* species (Sirová et al., 2003). Trap activities of α- and β-glucosidases, β-hexosaminidases, and aminopeptidases at the pH of 4.7 were usually lower by one or two orders of magnitude and were usually higher in the culture water at the same pH. Thus, a large proportion of the trap enzymatic activity, with the exception of phosphatases, entered the traps from the ambient water after firing. Generally, the activity of all enzymes was independent of prey digestion and was not inducible by prey or loading of N and P salts into the traps (Sirová et al., 2003; Adamec et al., 2010b). On the basis of a very efficient total N uptake from prey in *U. vulgaris* traps (Friday and Quarmby, 1994), it is evident that proteinaceous N as the main N source from prey must also be effectively digested and absorbed. The absence of aminopeptidases (proteases) in traps could be compensated by the autolysis of dead prey tissues. Yet the discrepancy between the findings of very low or zero protease activity in the trap fluid with or without prey, and in the presence of large secretory vesicles (Golgi apparatus) rich in proteases in quadrifid glands (Heslop-Harrison, 1975; Vintéjoux and Shoar-Ghafari, 2005) still cannot be explained. As proteases commonly have a very diverse substrate specificity it is possible to suspect the used microfluorimetric method that it involves only a minor part of all present proteases in the fluid.

Similarly, the same discrepancy exists between the invariably high phosphatase activity found in the trap fluid and a very low activity of enzyme labelled fluorescence (ELF) of phosphatase usually detected on the surface of quadrifid glands (Sirová et al., 2003; Adamec et al., 2011). At least a part of this discrepancy might be explained by methodical limitation of the ELF method (cf. Płachno et al., 2006). Moreover, it is not clear which proportion of any enzyme activity in the filtered trap fluid is produced by trap glands alone or various trap commensals. Consistently high trap fluid activities of phosphatases in all species imply that P uptake from prey or detritus might be more important than that of N for the plant.

The role of commensals in traps

Commensal microorganisms (mainly bacteria, algae (*Euglena*), ciliates, rotifers; e.g. Richards, 2001; Peroutka et al., 2008; Alkhalaf et al., 2009, 2011; Płachno et al., 2012) occur and propagate in the traps of many aquatic *Utricularia* species. The question of their role in trap functioning and possible benefit for the plants is often discussed. Presumably, some of these commensals participate, to various extent, in prey digestion by producing their own enzymes (Richards, 2001; Sirová et al., 2003), which has been demonstrated for phosphatases in commensal bacteria and unicellular algae (Płachno et al., 2006; Sirová et al., 2009). A slightly reduced growth of the wetland species *U. uliginosa* after the feeding on *Euglena* culture as prey (Jobson et al., 2000) suggests that the real relationship may even be slightly parasitic. Nevertheless, for some aquatic *Utricularia* species with low trapping efficiency (e.g. *U. purpurea*) in barren waters, commensal communities in traps seem to be more beneficial for the plants than the trapping of prey alone (Richards, 2001; Sirová et al., 2009.). As shown recently, commensals presumably participate in providing the traps without prey with N and P (Sirová et al., 2009). In traps without prey, which had sucked in some detritus or phytoplankton from the ambient water during incidental firings, a miniature microbial food web may run. Its main components are bacteria, Dinophyta, ciliates and rotifers. Similar interactions were found in the digestion fluid in *Sarracenia* pitchers (Gray et al., 2006). Moreover, in filtered fluids from empty traps of two field-grown *Utricularia* species, high concentrations of organic carbon (60–310 mg.l^{-1}), organic N (7–25 mg.l^{-1}), and soluble P (0.2–0.6 mg.l^{-1}) were detected (Sirová et al., 2009). Nevertheless, the total content of both C, N, and P in the trap fluid, including mainly the particulate form (i.e. the commensal organisms and detritus), was several times greater (in mg.l^{-1}): C, 632–1570; N, 21–81; P, 0.9–4.2. The concentrations usually increased with trap age and correlated with commensal biomass. On the basis of phospholipid fatty acid analysis of the trap commensal biomass, the occurrence of a complex microbial food web in the trap fluid was revealed, with bacteria forming >75% of the viable microbial biomass.

In a two-day experiment on *U. australis* and *U. vulgaris* using ^{13}C, a great proportion of newly fixed CO_2 was allocated from shoot bases not only to shoot apices, but also to mature shoot segments (Sirová et al., 2010). Total carbon allocation in plant tissues rapidly decreased with increasing age of the shoot segments but the ratio of C exuded into the trap fluid to that in plant tissues increased markedly with age – twice as much newly fixed C was allocated into the trap fluid than the plant tissue in the oldest analysed segments. Overall, 20–25% of the newly fixed C was allocated into the trap fluid. Sirová et al. (2011) have recently proven that C exudates fuel respiration of the prevailingly heterotrophic microbial commensal community within the empty traps of three aquatic *Utricularia* species. Up to 30% of the total dissolved organic C analysed in the trap fluid were easily metabolised compounds (mainly glucose, fructose, sucrose and lactate) and the proportion of exuded compounds, as well as their microbial utilisation, decreased with increasing mineral nutrient supply (N, P) and trap age. The total concentration of 46 analysed organic compounds in the trap fluid ranged between c. 30–150 mg.l^{-1} (i.e. 1.2–8.7 mM of organic C); comparable values between 9–78 mg.l^{-1} were found in another study in three *Utricularia* species (Borovec et al., 2012). Moreover, shaded plants exhibited lower concentrations of organics in their trap fluid. This extensive C supply to the trap fluid is undoubtedly an important additional maintenance (energetic) cost of traps. It is thus possible to conclude that the dominant part of C in the commensal organisms is provided by the plant (Sirová et al., 2009, 2010, 2011). Yet, on the basis of record high photosynthetic rate of photosynthetic *Utricularia* shoots (Adamec, 2006, 2013) the plants can afford such 'gardening' despite their very rapid growth. Beneficially, the plant could gain growth-limiting N and P from phytoplankton and detritus decomposed in the fluid of traps without any animal prey. It can be concluded that the concentration of organic compounds in the trap fluid is partly species specific and depends on cultivation conditions (irradiance) and water chemistry. High concentration of organic acids in the fluid could determine the low pH values of about 5 (Sirová et al., 2003, 2009; Adamec et al., 2010b) indicating also a high buffering capacity. The N_2 fixation mediated mainly by cyanobacteria occurred on the outer trap surface in *U. inflexa* (Wagner and Mshigeni, 1986). Owing to the specific chemical conditions, it could also run inside the traps and provide the traps with N. Sirová et al. (2014) have recently estimated $^{15}N_2$ fixation rate in traps and shoots of aquatic *Utricularia* and *Aldrovanda* but have confirmed a low ecological importance of the fixation for the seasonal plant N gain. Older traps and shoots were more active implying the key role of periphyton on the fixation. Presumably, the reason for limited N_2 fixation is the high concentration of NH_4-N (2.0–4.3 mg.l^{-1}) found in the trap fluid of two *Utricularia* species.

Traps presumably support this microbial food web energetically by supplying organic matter, and as a trade-off for this, they could obtain growth limiting N and P from decomposed detritus or phytoplankton. Thus, aquatic *Utricularia* species, which grow in very oligotrophic habitats with low prey availability, are 'bacterivorous' or 'detritivorous' rather than carnivorous.

Obviously, the most recent findings on trap operation – spontaneous firing and water recirculation – can better explain how growth-limiting N and P enter the traps from the ambient medium and become a substrate for the microbial food web. Evidently, spontaneous firing of each trap could lead to a substantial gain of N and P for the traps. During its three-week life-span, each trap can aspirate around 15 times its own volume in surrounding water (40% trap volume × 2 daily spontaneous firings × 20-day life-span). If the trap commensals help to decompose this 'non-animal' prey, it could theoretically imply a significant N and P gain for the plant, especially for those plants in barren waters which trap few animals. The literature assumed that this N and P gain by empty traps could be ecologically important but no experimental quantification has been given so far (Richards, 2001; Peroutka et al., 2008; Alkhalaf et al., 2009, 2011; Sirová et al., 2009, 2010, 2011). When, however, one considers the published data on the mean total N and P content (both dissolved and particulate, see above) within a middle-aged trap of *U. purpurea* and *U. foliosa* (Sirová et al., 2009) and the mean total N and P content at natural oligo- and mesotrophic sites of *U. australis* in the Czech Republic (Adamec, 2008b), a simple budget model of N and P gain can be made. When water recirculation through the traps is also accounted for then, surprisingly, only a very slow accumulation of total N and P can occur within empty traps (see Adamec, 2011c for all details). [Model data: trap volume 5 µl, water recirculation rate 1 µl.h^{-1} for 20 h a day, only NH_4-N and PO_4-P influx from the ambient water, mean natural concentrations of NH_4-N and

PO_4-P at the sites.] Simply, it would take between 40–70 days for an empty trap to accumulate the measured N or between 15–23 days for them to accumulate the P if there were only spontaneous firing and water recirculation mechanisms occurring. By the application of the model data for the relationship between trap volume and biomass and for shoot N and P content in *U. australis*, one can further estimate that the total N and P amount inside the traps represents only about 3.5% of total plant N and 1.2% of P (Adamec, 2011c).

Several conclusions can be drawn from these model data.

A) The calculated N and P input rates from the ambient water are so low that each trap without prey can only reach these nutrient levels after a long time, which is comparable with or even longer than the trap life-span. Thus, such a low N and P gain cannot be ecologically important for the plant mineral nutrition at all.

B) Since even young *Utricularia* traps (c. 1 week old) contained a relatively high total N and P content comparable with that in the older traps (Sirová et al., 2009), the traps could take up neither N nor P from the trap fluid for the plant itself as the total N and P content in the trap fluid would have been much lower.

C) To account for all of these findings, it is evident that the traps exude also N and P to enhance the microbial community. It is possible that inoculation of young traps by microorganisms stimulates the traps to exude both organic C and also N and P. Therefore, it seems that *Utricularia* traps without animal prey have no nutritional benefit from the trap commensal community and that the trap microorganisms behave rather as parasites than commensals. The extent of this N and P exudation to the trap fluid can only be a few % of the total plant N and P amount.

D) It can be inferred that the N and P uptake affinity of *Utricularia* shoots growing in very oligotrophic, barren waters with very low prey availability is very high (the limits for uptake should be below 1 μM for NH_4^+ and 0.1 μM for phosphate), while that for N- and P-containing substances in the traps without prey can be very low or zero (cf. Sirová et al., 2009). This conclusion has recently been supported by finding of high NH_4^+ concentration in the trap fluid of two *Utricularia* species (Sirová et al., 2014).

E) If the trap microorganisms are beneficial for the plant, the ecological benefit can occur only in traps with captured prey to facilitate prey digestion. The trap microorganisms in traps represent additional ecological costs for trap maintenance.

F) If capturing animal prey is crucially important for the nutritional benefit of carnivory in *Utricularia*, the strategy of prey capturing could ‘drive’ the evolution of traps in aquatic *Utricularia* species, while the interaction with the trap commensal organisms could be more marginal.

Oxygen regime and trap respiration

Surprisingly, zero O_2 concentration was consistently detected in the fluid of excised and intact traps (without prey) of six aquatic *Utricularia* species bathed in an oxygenated medium, regardless of trap age and irradiance (Adamec, 2007b). Thus, there is normally anoxia inside the traps which can incidentally be interrupted due to trap firing (trapping of prey or spontaneous firing) for short periods of time. The potential aerobic respiration of the inner glands and trap walls is so high that all O_2 is exhausted to zero within 10–40 min. The traps can pump out water and reset the negative pressure within 30 min, which requires high amounts of energy derived from aerobic respiration, and is prevented by respiration inhibitors (Sydenham and Findlay, 1975). Yet it is not clear how the traps (glands) provide sufficient ATP energy for their demanding functions under anoxia, though a mitochondrial mutation of cytochrome *c* oxidase found in *Utricularia* should provide greater energetic power for the traps (Laakkonen et al., 2006). Evidently, the inner trap glands possess an extremely high O_2 affinity (well below 0.5–1 µM; see Adamec, 2012b), using the permanent O_2 influx from the trap wall intercellulars. Finally, transcriptomic global gene expression analysis in *U. gibba* has recently confirmed that traps significantly over-express genes involved in respiration (Ibarra-Laclette et al., 2011). The extremely low O_2 concentration in the trap fluid is thus a result of a functional compromise: it must be very low to reliably kill the captured prey (requiring c. >15–30 µM) but higher than a threshold for an effective aerobic respiration (>c. 0.4 µM). In traps with captured prey, anoxia causes prey to die of suffocation, while all trap commensals are adapted to facultative anoxia (Adamec, 2007b).

Photosynthesis of aquatic carnivorous plants

While the maximum net photosynthetic rate (P_{max}) per unit dry weight (DW) or area of leaves of slowly growing terrestrial carnivorous species is, on average, 2–5 times lower than that in different functional groups of terrestrial non-carnivorous species (Ellison, 2006; Ellison and Adamec, 2011), P_{max} in aquatic carnivorous species with linear shoots (eight species, 40–160 mmol O_2.kg^{-1} fresh weight.h^{-1}) is comparable with the highest values found in aquatic non-carnivorous species (30–110 mmol.kg^{-1} fresh weight.h^{-1}; Adamec, 1997b, 2006, 2013; Ellison and Adamec, 2011). Surprisingly, very high P_{max} was also found in old shoots of *U. australis* and *U. vulgaris* without functional traps (Adamec, 2013). Thus, very high P_{max} is typical for aquatic carnivorous species with rapid growth but is also a prerequisite for this rapid growth as the rapid, permanent decay of senescent shoot segments causes a great loss of structural and non-structural carbohydrates (Adamec, 2000). In amphibious carnivorous species with dimorphic shoots, P_{max} of photosynthetic shoots could increase considerably if the shoots become emergent (Colmer and Pedersen, 2008).

Traps of aquatic species as physiologically very active organs have high respiration rates (RD) and represent great photosynthetic costs, as hypothesized by Givnish et al. (1984). In six aquatic *Utricularia* species, trap RD per unit fresh weight (5.1–8.6 $\mathrm{mmol.kg^{-1}.h^{-1}}$) was 1.7–3.0 times greater than that in leaves on carnivorous or photosynthetic shoots and nearly-maximum P_{max} in photosynthetic leaves exceeded that in the traps (5.2–14.7 $\mathrm{mmol.kg^{-1}.h^{-1}}$) 7–10 times (Adamec, 2006). Thus, very high RD:P_{max} ratio in traps of these species (50–140%), unlike that in leaves (3.6–8.2%), means that there are high maintenance and photosynthetic costs of traps: in *U. stygia* and *U. intermedia* with dimorphic shoots, the trap RD could amount to 34–44% of the total plant respiration, while 63% in *U. australis* with monomorphic shoots (Adamec, 2006, 2007a, 2008b). However, in *U. macrorhiza*, mean trap RD was only about 10% higher than that in leaves and trap P_{max} in lake water was as much as 41–67% of that in the leaves (Knight, 1992); similarly 67% in *Aldrovanda* (Adamec, 1997b).

Aquatic carnivorous plants usually grow in waters with high $[CO_2] > 0.1$ mM. This fact is very important as all aquatic species tested so far use only CO_2 for photosynthesis (see Adamec, 1997a,b; Adamec and Kovářová, 2006; Pagano and Titus, 2007). The recent finding of slight HCO_3^- use in *U. australis* induced by growing at high pH of about 9.2 (Adamec, 2009) deserves further study. Generally, in several aquatic carnivorous species growing in the field or culture, CO_2 compensation points (CO_2 CPs) fell within the range 1.5–13.2 µM (Adamec, 1997a,b, 2009; Adamec and Kovářová, 2006; Pagano and Titus, 2007); similar values of 1.5–10 µM are reported in aquatic non-carnivorous plants (Maberly and Spence, 1983). In 17 culture-grown species or accessions of aquatic carnivorous plants of both genera, CO_2 CPs (mean 5.3 µM, range 1.9–13.6 µM) were similar to those found in these species growing in vitro (mean 5.2 µM, range 2.5–8.8 µM; Adamec and Pásek, 2009). A highly significant relationship was found between the CO_2 CP values and $[CO_2]$ in the culture water. Similarly, CO_2 CPs in *U. australis* growing at 17 sites of different trophic levels ranged within 0.7–6.1 µM (mean 2.6 µM) but correlated significantly with neither of the internal or water chemistry factors nor with the capture of prey (Adamec, 2009).

The influence of prey capture on P_{max} and RD was investigated in *Aldrovanda* and *U. australis* growing in an outdoor culture (Adamec, 2008a). Both species fed on zooplankton grew significantly faster than unfed ones. Feeding increased P_{max} by 59% in *Aldrovanda* but decreased it by 25% in *U. australis*. CO_2 CP was unchanged due to feeding in *Aldrovanda* but increased from 5.2 to 9.2 µM in *U. australis*. The RD values stayed unchanged in both species. Thus, the hypothesis by Givnish et al. (1984) on stimulation of photosynthesis by catching prey has not been supported, although more data are needed. Carnivory should partly compensate for photosynthetic CO_2 uptake, but the uptake of organic carbon from prey in aquatic carnivorous plants has never been quantified. Yet, organic carbon uptake from prey in aquatic species may be ecologically important under CO_2 shortage (see Adamec, 1997a): field-grown *Aldrovanda* was also able to grow at pH > 9.0 when catching numerous prey (Adamec, 1999), and greenhouse-grown *U. vulgaris* fed on prey grew better and branched more only at higher pH values of 7.6–9.1 (Kosiba, 1992).

Mineral nutrition in aquatic carnivorous plants

Although rootless aquatic carnivorous plants grow in mineral-poor habitats they have similar macroelement composition to rooted aquatic non-carnivorous species (cf. Dykyjová, 1979; Adamec, 1997a, 2008a,b; Ellison and Adamec, 2011). Their tissue nutrient content (%DW) in young shoots is usually between 1.0–4.0 for N; 0.12–0.50 P; 1.5–5.0 K; 0.15–3.0 Ca; 0.2–0.7 for Mg. *U. australis* growing in very oligotrophic waters with low prey availability kept a relatively high shoot nutrient content (Adamec, 2008b). Regardless of a marked polarity of tissue N, P, and Ca content along shoots and the great differences between leaves and traps (Adamec, 2000, 2008b, 2010b), mean shoot content of these five macroelements in aquatic carnivorous plants is about 1.5–3 times greater than that in terrestrial carnivorous plant leaves (cf. Ellison, 2006; Ellison and Adamec, 2011) and could reflect much faster growth in aquatic species. A marked polarity of tissue N, P, Ca content, indicating a very efficient N and P reutilization from senescing shoot segments, was found in rapidly growing aquatic species (Adamec, 1997a, 2000, 2008b, 2014). The Ca polarity was opposite, whereas K and Mg contents were constant. Culture-grown *Aldrovanda* reutilized 88% N and 67% P (Adamec, 2000) and field-grown *U. australis* on average only 48% N and 72% P (Adamec, 2008b). However, the slowly growing *U. purpurea* re-utilized only 37% N and 71% P (see Adamec, 1997a). Moreover, a very effective reutilization of N and P can be assumed in autumnal shoots forming turions (Adamec, 2000). Thus, aquatic species permanently lose only a relatively small part of their N and P in their senescent shoots (like terrestrial species; Adamec, 1997a) but all K, Ca, and Mg. Zero K reutilization in senescent shoots of aquatic species greatly contrasts with very efficient K reutilization in terrestrial ones (41–99%; Adamec, 2002, 2014).

Considerable differences in tissue nutrient content occur between shoots and traps in aquatic carnivorous plants of both genera. The content of N, Ca, and Mg is usually greater in photosynthetic shoots than in the traps, while the opposite is true for P and K demonstrating a considerable 'mineral' cost of carnivory (Adamec, 2008b, 2010b). In *U. australis* with monomorphic shoots and mean proportion of trap DW 38%, traps in adult shoots contained about 30% total N, 53% P, and 51% K within the shoots (Adamec, 2008b).

Aquatic carnivorous species grow in oligo-mesotrophic waters with [NH_4^+] usually dominating over [NO_3^-] to a considerable extent. *Aldrovanda* and some aquatic *Utricularia* species preferentially took up NH_4^+ to NO_3^- from diluted NH_4NO_3 solutions (Adamec, 1997a, 2000; Fertig, 2001). Phosphate uptake by *Aldrovanda* apical shoot parts was two times faster than that by basal parts, but the finding that *Aldrovanda* takes up K^+ only by the basal parts (Adamec, 2000) could be explained as an interaction with NH_4^+ uptake (Adamec, unpubl.). Besides, aquatic species can take up a substantial amount of mineral nutrients also from prey. Yet, only Friday and Quarmby (1994) have quantified an efficiency of N uptake from prey in aquatic species. In *U. vulgaris* fed on mosquito larvae, they estimated the efficiency of N uptake to be at least 83% of the total prey N. About 52% total plant N was obtained from the prey. Probably, the efficiency of N uptake from prey in *Utricularia* traps may be even higher than that in terrestrial species (cf. Adamec, 2002). P was also taken up rapidly from the prey but P reutilization from old shoot segments was much better than that of N.

There are several data showing marked effects of prey utilization on the growth of aquatic species, both in a culture and in the field (for the review see Adamec, 1997a, 2000, 2008a; Englund and Harms, 2003; Adamec et al., 2010a). Feeding led to longer shoots, greater DW, faster apical shoot growth, greater RGR, and, especially, increased branching as main means of propagation. It may be summarised that catch of prey has usually a greater effect in *Aldrovanda* than in aquatic *Utricularia* species. Thus, carnivory in aquatic species is at least as important as in terrestrial ones. However, it is still unclear how this growth effect of carnivory is induced since tissue N and P contents in apical or young shoot segments in prey-fed plants were lower compared to unfed plants (Adamec, 2000, 2008a, 2011d). A hypothesis that N and P absorbed from prey supports preferentially essential growth processes associated with cell division in shoot apices (Adamec, 2008a) was supported only in *Aldrovanda*, but not in *U. australis* and *U. bremii* (Adamec, 2011d).

Another important ecological parameter associated with mineral nutrition is the proportion of seasonal (daily) N and P gain obtained from prey. In *U. macrorhiza*, the proportion of seasonal N gain from carnivory was estimated to be about 75% (Knight, 1988). In robust *U. foliosa* growing at a nutrient-poor site with extremely low prey availability in Florida, the mean proportion was only about 0.9% N and 3.5% P (Bern, 1997). At sites with greater prey availability, these values could be one order of magnitude higher. Using a model containing literature-based data (Adamec, 1997a, 2000, 2008b; Adamec and Kovářová, 2006), it is possible to calculate which proportion of the daily N and P gain can be obtained due to capturing one small *Cyclops* (DW 25 µg) daily in *Aldrovanda* and *U. australis*. In rapidly growing plants (plant biomass doubled in 15 d) this covers about 15% daily N and 4% P gain in smaller *Aldrovanda*, while only about 0.62% N and 0.56% P gain in larger *U. australis*. However, if plant growth is zero and the plants maintain constant biomass, which is the case under unfavourable growth conditions, then it covers up to 100% daily N and about 16% P gain in *Aldrovanda* and about 1.8% N and 2.6% P gain in *U. australis*. As in terrestrial species, the estimated values show that the ecological importance of N and P uptake from prey depends primarily on the quantity of captured prey. Capture of prey in aquatic species is thus one of the decisive factors for their rapid growth and especially for their propagation.

Regulation of investment in carnivory in *Utricularia*

In aquatic *Utricularia* species, the proportion of traps to the total plant biomass as the structural cost (investment) in carnivory is 10–65%, but this proportion is regulated flexibly by the plants to minimize the costs of carnivory according to habitat factors: particularly water chemistry, prey capture, and irradiance (Knight and Frost, 1991; Friday, 1992; Bern, 1997; Guisande et al., 2000, 2004; Richards, 2001; Englund and Harms, 2003; Manjarrés-Hernández et al., 2006; Porembski et al., 2006; Kibriya and Jones, 2007; Adamec, 2007a, 2008b, 2009). Moreover, this regulation may be different in various species. Yet, in most cases, increased mineral nutrient availability either in the ambient water or prey led to decreased investment in carnivory in the terms of trap number per leaf or proportion of trap biomass. The number of traps per leaf in *U. foliosa* decreased in waters with increasing [NO_3^-] but simultaneous decreased capture of prey (Guisande et al., 2004). Thus, it is not possible to separate both factors. However, the number of traps per leaf in the same species correlated statistically significantly (inversely proportionally) with shoot P and especially N content (Bern, 1997). Out of all nutrient factors investigated in field-grown *U. australis*, only tissue N content in young shoot segments significantly (negatively) correlated with trap proportion (Adamec, 2008b). These results consistently support the ‘nutrient’ hypothesis that all external nutrient factors, which decrease tissue N content in young shoots (poor prey capture, low [NH_4^+], high [CO_2], etc.), increase trap production in young shoots and *vice versa*, as a negative feedback regulation. This finding is consistent with the suggestion of Guisande et al. (2004) that ambient N sources are a limiting factor regulating investment in carnivory. However, Kibriya and Jones (2007), studying *U. vulgaris*, confirmed a central regulatory role for P, while both N and P were found to play the regulating role in *U. foliosa* (Bern, 1997). This negative feedback also helps to stabilize the tissue contents of other mineral nutrients. As prey capture also supports plant growth, growth rate itself is obviously a component of this endogenous regulatory system. Moreover, some data show that the 'nutrient' regulation of trap proportion in aquatic *Utricularia* is subject to 'photosynthetic' regulation (Bern, 1997; Englund and Harms, 2003; Adamec, 2008b, 2015). At low photosynthetic rate (low irradiance, [CO_2]), trap proportion is relatively low or even zero indicating that the CO_2 regulation is superior to the 'nutrient' one. In three aquatic *Utricularia* species, CO_2 addition to the cultivation water markedly increased the investment in carnivory in all species, while the effect of prey addition was much less (Adamec, 2015). The investment in all species correlated significantly and negatively with shoot N and P contents but highly significantly and positively with the mean trap DW.

Concluding remarks: further research

As shown in the review, aquatic carnivorous plants are ecophysiologically quite dissimilar to their terrestrial counterparts, the main ecophysiological strategy of which as S-strategists is slow growth (Ellison and Adamec, 2011), while the former plants – especially European species – grow very rapidly and exhibit rather R strategy of growth and thorough K strategy of utilization of sources, except for CO_2 availability. The principal growth traits in rootless aquatic species with linear shoots – very rapid apical growth, rapid basal shoot decay, and high RGR – are associated with very steep physiological polarity along the shoots and require a combination of several ecophysiological processes. The newest data on the biophysics of *Utricularia* traps (trap door buckling, spontaneous firings, water recirculation) together with those on the interactions of trap microbial community with traps (presence of complete food web, exudation of organics into the traps) have markedly changed the classic view of trap function.

To obtain further insight into the ecophysiology of aquatic carnivorous plants, the following directions of research could be considered and the questions raised could be answered (see also Ellison and Adamec, 2011 and Adamec, 2012b).

- The effect of growth enhancement due to carnivory: which physiological effects are primary and which are secondary? What is the role of tissue N and P content in this growth enhancement? Is it possible to assume that the positive growth effect is caused by stimulation of cell divisions in shoot apices?

- Steep growth polarity in rootless aquatic carnivorous plants and their very rapid apical shoot growth associated with steep physiological polarity imply the involvement of physiological processes that are insufficiently understood (e.g. phytohormone production and distribution).
- What is the affinity of shoots for mineral nutrient uptake from water? Is shoot nutrient uptake from the ambient water stimulated by prey capture (in analogy with terrestrial species)? What is the efficiency of mineral nutrient uptake from prey?
- As opposed to terrestrial species, aquatic carnivorous species do not re-utilise K^+ from senescent shoots and lose all K^+ in them. Yet animal prey is a poor source of K^+ and the ambient waters can be poor in K^+. What are the K^+ uptake characteristics of shoots of aquatic carnivorous plants (localisation of uptake, affinity, uptake rates)?
- The importance of phytoplankton and detritus as a potential nutrient source (N, P, K, Mg) for *Utricularia* in barren waters could be elucidated on the basis of estimation of the matter in the trap fluid and modelling.
- In *Utricularia*, what is the role of the commensal community in the nutrient interactions within the plant having traps with or without prey?

Acknowledgements

This study was partly funded by the Czech Scientific Foundation (Research Project No. P504/11/0783) and the Long-term research development project (RVO 67985939). Thanks are due to Prof. Georg Janauer for critically reading the manuscript and valuable comments.

References

Adamec, L., 1995. Ecological requirements and the European distribution of the aquatic carnivorous plant *Aldrovanda vesiculosa* L. *Folia Geobot. Phytotax.* 30, 53–61.

Adamec, L., 1997a. Mineral nutrition of carnivorous plants: A review. *Bot. Rev.* 63, 273–299.

Adamec, L., 1997b. Photosynthetic characteristics of the aquatic carnivorous plant *Aldrovanda vesiculosa*. *Aquat. Bot.* 59, 297–306.

Adamec, L., 1999. Seasonal growth dynamics and overwintering of the aquatic carnivorous plant *Aldrovanda vesiculosa* at experimental field sites. *Folia Geobot.* 34, 287–297.

Adamec, L., 2000. Rootless aquatic plant *Aldrovanda vesiculosa*: physiological polarity, mineral nutrition, and importance of carnivory. *Biol. Plant.* 43, 113–119.

Adamec, L., 2002. Leaf absorption of mineral nutrients in carnivorous plants stimulates root nutrient uptake. *New Phytol.* 155, 89–100.

Adamec, L., 2006. Respiration and photosynthesis of bladders and leaves of aquatic *Utricularia* species. *Plant Biol.* 8, 765–769.

Adamec, L., 2007a. Investment in carnivory in *Utricularia stygia* and *U. intermedia* with dimorphic shoots. *Preslia* 79, 127–139.

Adamec, L., 2007b. Oxygen concentrations inside the traps of the carnivorous plants *Utricularia* and *Genlisea* (Lentibulariaceae). *Ann. Bot*. 100, 849–856.

Adamec, L., 2008a. The influence of prey capture on photosynthetic rate in two aquatic carnivorous plant species. *Aquat. Bot.* 89, 66–70.

Adamec, L., 2008b. Mineral nutrient relations in the aquatic carnivorous plant *Utricularia australis* and its investment in carnivory. *Fundam. Appl. Limnol.* 171, 175–183.

Adamec, L., 2008c. Soil fertilization enhances growth of the carnivorous plant *Genlisea violacea. Biologia* 63, 201–203.

Adamec, L., 2009. Photosynthetic CO_2 affinity of the aquatic carnivorous plant *Utricularia australis* (Lentibulariaceae) and its investment in carnivory. *Ecol. Res*. 24, 327–333.

Adamec, L., 2010a. Field growth analysis of *Utricularia stygia* and *U. intermedia* – two aquatic carnivorous plants with dimorphic shoots. *Phyton* 49, 241–251.

Adamec, L., 2010b. Mineral cost of carnivory in aquatic carnivorous plants. *Flora* 205, 618–621.

Adamec, L., 2011a. Shoot branching of the aquatic carnivorous plant *Utricularia australis* as the key process of plant growth. *Phyton* 51, 133–148.

Adamec, L., 2011b. The comparison of mechanically stimulated and spontaneous firings in traps of aquatic carnivorous *Utricularia* species. *Aquat. Bot.* 94, 44–49.

Adamec, L., 2011c. Functional characteristics of traps of aquatic carnivorous *Utricularia* species. *Aquat. Bot*. 95, 226–233.

Adamec, L., 2011d. By which mechanism does prey capture enhance plant growth in aquatic carnivorous plants: Stimulation of shoot apex? *Fundam. Appl. Limnol.* 178, 171–176.

Adamec, L., 2012a. Why do aquatic carnivorous plants prefer growing in dystrophic waters? *Acta Biol. Sloven*. 55, 3–8.

Adamec, L., 2012b. News in ecophysiological research on aquatic *Utricularia* traps. *Carniv. Plant Newslett*. 41, 92–104.

Adamec, L., 2012c. Firing and resetting characteristics of carnivorous *Utricularia reflexa* traps: Physiological or only physical regulation of trap triggering? *Phyton* 52, 281–290.

Adamec, L., 2013. A comparison of photosynthetic and respiration rates in six aquatic carnivorous *Utricularia* species differing in morphology. *Aquat. Bot.* 111, 89–94.

Adamec, L., 2014. Different reutilization of mineral nutrients in senescent leaves of aquatic and terrestrial carnivorous *Utricularia* species. *Aquat. Bot*. 119, 1–6.

Adamec, L., 2015. Regulation of the investment in carnivory in three aquatic *Utricularia* species: CO_2 or prey availability? *Phyton* 55, 131–148.

Adamec, L., Kovářová, M., 2006. Field growth characteristics of two aquatic carnivorous plants, *Aldrovanda vesiculosa* and *Utricularia australis*. *Folia Geobot*. 41, 395–406.

Adamec, L., Pásek, K., 2009. Photosynthetic CO_2 affinity of aquatic carnivorous plants growing under nearly-natural conditions and in vitro. *Carniv. Plant Newslett*. 38, 107–113.

Adamec, L., Sirová, D., Vrba, J., 2010a. Contrasting growth effects of prey capture in two carnivorous plant species. *Fundam. Appl. Limnol.* 176, 153–160.

Adamec, L., Sirová, D., Vrba, J., Rejmánková, E., 2010b. Enzyme production in the traps of aquatic *Utricularia* species. *Biologia* 65, 273–278.

Adamec, L., Vrba, J., Sirová, D., 2011. Fluorescence tagging of phosphatase and chitinase activity on different structures of *Utricularia* traps. *Carniv. Plant Newslett.* 40, 68–73.

Alkhalaf, I.A., Hübener, T., Porembski, S., 2009. Prey spectra of aquatic *Utricularia* species (Lentibulariaceae) in northeastern Germany: The role of planktonic algae. *Flora* 204, 700–708.

Alkhalaf, I.A., Hübener, T., Porembski, S., 2011. Microalgae trapped by carnivorous bladderworts (*Utricularia*, Lentibulariaceae), analysis, attributes and structure of the microalgae trapped. *Plant Div. Evol*. 129, 125–138.

Bern, A.L., 1997. Studies on nitrogen and phosphorus uptake by the carnivorous bladderwort *Utricularia foliosa* L. in South Florida wetlands. MSc. thesis, Florida Int. Univ., Miami.

Borovec, J., Sirová, D., Adamec, L., 2012. Light as a factor affecting the concentration of simple organics in the traps of aquatic carnivorous *Utricularia* species. *Fundam. Appl. Limnol*. 181, 159–166.

Casper, S.J., Krausch, H.D., 1981. *Süsswasserflora von Mitteleuropa. Band 24, Pteridophyta und Anthophyta*. VEB Gustav Fischer Verlag, Jena.

Colmer, T.D., Pedersen, O., 2008. Underwater photosynthesis and respiration in leaves of submerged wetland plants: gas films improve CO_2 and O_2 exchange. *New Phytol.* 177, 918–926.

Dykyjová, D., 1979. Selective uptake of mineral ions and their concentration factors in aquatic higher plants. *Folia Geobot. Phytotax*. 14, 267–325.

Ellison, A.M., Adamec, L., 2011. Ecophysiological traits of terrestrial and aquatic carnivorous plants: are the costs and benefits the same? *Oikos* 120, 1721–1731.

Englund, G., Harms, S., 2003. Effects of light and microcrustacean prey on growth and investment in carnivory in *Utricularia vulgaris*. *Freshwater Biol.* 48, 786–794.

Farnsworth, E.J., Ellison, A.M., 2008. Prey availability directly affects physiology, growth, nutrient allocation and scaling relationships among leaf traits in ten carnivorous plant species. *J. Ecol.* 96, 213–221.

Fertig, B., 2001. Importance of prey derived and absorbed nitrogen to new growth: preferential uptake of ammonia or nitrate for three species of *Utricularia.* MSc-thesis, Brandeis Univ., Waltham, MA, USA.

Friday, L.E., 1989. Rapid turnover of traps in *Utricularia vulgaris* L. *Oecologia* 80, 272–277.

Friday, L.E., 1992. Measuring investment in carnivory, seasonal and individual variation in trap number and biomass in *Utricularia vulgaris* L. *New Phytol.* 121, 439–445.

Friday, L.E., Quarmby, C., 1994. Uptake and translocation of prey-derived ^{15}N and ^{32}P in *Utricularia vulgaris* L. *New Phytol.* 126, 273–281.

Givnish, T.J., Burkhardt, E.L., Happel, R.E., Weintraub, J.D., 1984. Carnivory in the bromeliad *Brocchinia reducta*, with a cost/benefit model for the general restriction of carnivorous plants to sunny, moist, nutrient-poor habitats. *Am. Nat.* 124, 479–497.

Gray, S.M., Miller, T.E., Mouquet, N., Daufresne, T., 2006. Nutrient limitation in detritus-based microcosms in *Sarracenia purpurea*. *Hydrobiologia* 573, 173–181.

Guiral, D., Rougier, C., 2007. Trap size and prey selection of two coexisting bladderwort (*Utricularia*) species in a pristine tropical pond (French Guiana) at different trophic levels. *Int. J. Limnol.* 43, 147–159.

Guisande, C., Andrade, C., Granado-Lorencio, C., Duque, S.R., Núñez-Avellaneda, M., 2000. Effects of zooplankton and conductivity on tropical *Utricularia foliosa* investment in carnivory. *Aquat. Ecol.* 34, 137–142.

Guisande, C., Aranguren, N., Andrade-Sossa, C., Prat, N., Granado-Lorencio, C., Barrios, M.L., Bolivar, A., Núñez-Avellaneda, M., Duque, S.R., 2004. Relative balance of the cost and benefit associated with carnivory in the tropical *Utricularia foliosa*. *Aquat. Bot.* 80, 271–282.

Guisande, C., Granado-Lorencio, C., Andrade-Sossa, C., Duque, S.R., 2007. Bladderworts. *Funct. Plant Sci. Biotechnol.* 1, 58–68.

Heslop-Harrison, Y., 1975. Enzyme release in carnivorous plants. In: Dingle, J.T., Dean, R.T. (Eds.), *Lysozymes in Biology and Pathology. Vol. 4.* 525–578. North Holland Publishing, Amsterdam.

Ibarra-Laclette, E., Albert, V.A., Perez-Torres, C.A., Zamudio-Hernandez, F., Ortega-Estrada, M.J. De, Herrera-Estrella, A., Herrera-Estrella, L., 2011. Transcriptomics and molecular evolutionary rate analysis of the bladderwort (*Utricularia*), a carnivorous plant with a minimal genome. *BMC Plant Biol.* 11, 101.

Jobson, R.W., Morris, E.W., Burgin, S., 2000. Carnivory and nitrogen supply affect the growth of the bladderwort *Utricularia uliginosa*. *Aust. J. Bot.* 48, 549–560.

Joeyux, M., Vincent, O., Marmottant, P., 2011. Mechanical model of the ultrafast underwater trap of *Utricularia*. *Phys. Lett.* E83, 021911.

Juniper, B.E., Robins, R.J., Joel, D.M., 1989. *The Carnivorous Plants.* Academic Press Ltd., London.

Kamiński, R., 1987. Studies on the ecology of *Aldrovanda vesiculosa* L. I. Ecological differentiation of *A. vesiculosa* population under the influence of chemical factors in the habitat. *Ekol. Pol.* 35, 559–590.

Kibriya, S., Jones, J.I., 2007. Nutrient availability and the carnivorous habit in *Utricularia vulgaris*. *Freshwater Biol.* 52, 500–509.

Knight, S.E., 1988. The ecophysiological significance of carnivory in *Utricularia vulgaris*. Ph.D. thesis, Univ. Wisconsin, Madison, USA.

Knight, S.E., 1992. Costs of carnivory in the common bladderwort, *Utricularia macrorhiza*. *Oecologia* 89, 348–355.

Knight, S.E., Frost, T.M., 1991. Bladder control in *Utricularia macrorhiza*, lake-specific variation in plant investment in carnivory. *Ecology* 72, 728–734.

Kosiba, P., 1992. Studies on the ecology of *Utricularia vulgaris* L. I. Ecological differentiation of *Utricularia vulgaris* L. population affected by chemical factors of the habitat. *Ekol. Pol.* 40, 147–192.

Laakkonen, L., Jobson, R.W., Albert, V.A., 2006. A new model for the evolution of carnivory in the bladderwort plant (*Utricularia*), adaptive changes in cytochrome c oxidase (COX) provide respiratory power. *Plant Biol.* 8, 758–764.

Maberly, S.C., Spence, D.H.N., 1983. Photosynthetic inorganic carbon use by freshwater plants. *J. Ecol.* 71, 705–724.

Manjarrés-Hernández, A., Guisande, C., Torres, N.N., Valoyes-Valois, V., González-Bermúdez, A., Díaz-Olarte, J., Sanabria-Aranda, L., Duque, S.R., 2006. Temporal and spatial change of the investment in carnivory of the tropical *Utricularia foliosa*. *Aquat. Bot.* 85, 212–218.

Navrátilová, J., Navrátil, J., 2005. Stanovištní nároky některých ohrožených a vzácných rostlin rašelinišť Třeboňska. [Environmental factors of some endangered and rare plants in Třeboň's mires.] In Czech. *Zprávy Čes. Bot. Spol. (Prague)* 40, 279–299.

Nielsen, S.L., Sand-Jensen, K., 1991. Variation in growth rates of submerged rooted macrophytes. *Aquat. Bot.* 39, 109–120.

Pagano, A.M., Titus, J.E., 2004. Submersed macrophyte growth at low pH, contrasting responses of three species to dissolved inorganic carbon enrichment and sediment type. *Aquat. Bot.* 79, 65–74.

Pagano, A.M., Titus, J.E., 2007. Submersed macrophyte growth at low pH, carbon source influences response to dissolved inorganic carbon enrichment. *Freshwater Biol.* 52, 2412–2420.

Peroutka, M., Adlassnig, W., Volgger, M., Lendl, T., Url, W.G., Lichtscheidl, I.K., 2008. *Utricularia*, a vegetarian carnivorous plant? Algae as prey of bladderwort in oligotrophic bogs. *Plant Ecol.* 199, 153–162.

Płachno, B.J., Adamec, L., 2007. Differentiation of *Utricularia ochroleuca* and *U. stygia* populations in Třeboň basin, on the basis of quadrifid glands. *Carniv. Plant Newslett. (Fullerton)* 36, 87–95.

Płachno, B.J., Adamec, L., Lichtscheidl, I.K., Peroutka, M., Adlassnig, W., Vrba, J., 2006. Fluorescence labelling of phosphatase activity in digestive glands of carnivorous plants. *Plant Biol.* 8, 813–820.

Płachno, B.J., Łukaszek, M., Wołowski, K., Adamec, L., Stolarczyk, P., 2012. Aging of *Utricularia* traps and variability of microorganisms associated with that microhabitat. *Aquat. Bot.* 97, 44–48.

Porembski, S., Theisen, I., Barthlott, W., 2006. Biomass allocation patterns in terrestrial, epiphytic and aquatic species of *Utricularia* (Lentibulariaceae). *Flora* 201, 477–482.

Rice, B.A., 2011. Reversing the roles of predator and prey: A review of carnivory in the botanical world. In: Seckbach, J., Dubinski, Z. (Eds.), *All Flesh is Grass. Plant-Animal Interrelationships. Cellular Origin, Life in Extreme Habitats and Astrobiology*, Vol. 16. Springer Science + Business Media B.V., Dordrecht, Heidelberg, London, New York, 491–518 pp.

Richards, J.H., 2001. Bladder function in *Utricularia purpurea* (Lentibulariaceae), is carnivory important? *Am. J. Bot.* 88, 170–176.

Sasago, A., Sibaoka, T., 1985a. Water extrusion in the trap bladders of *Utricularia vulgaris* I. A possible pathway of water across the bladder wall. *Bot. Mag.* 98, 55–66.

Sasago, A., Sibaoka, T., 1985b. Water extrusion in the trap bladders of *Utricularia vulgaris* II. A possible mechanism of water outflow. *Bot. Mag.* 98, 113–124.

Singh, A.K., Prabhakar, S.P., Sane, S.P., 2011. The biomechanics of fast prey capture in aquatic bladderworts. *Biol. Lett.* 7, 547–550.

Sirová, D., Adamec, L., Vrba, J., 2003. Enzymatic activities in traps of four aquatic species of the carnivorous genus *Utricularia*. *New Phytol.* 159, 669–675.

Sirová, D., Borovec, J., Černá, B., Rejmánková, E., Adamec, L., Vrba, J., 2009. Microbial community development in the traps of aquatic *Utricularia* species. *Aquat. Bot.* 90, 129–136.

Sirová, D., Borovec, J., Picek, T., Adamec, L., Nedbalová, L., Vrba, J., 2011. Ecological implications of organic carbon dynamics in the traps of aquatic carnivorous *Utricularia* plants. *Funct. Plant Biol.* 38, 583–593.

Sirová, D., Borovec, J., Šantrůčková, H., Šantrůček, J., Vrba, J., Adamec, L., 2010. *Utricularia* carnivory revisited: plants supply photosynthetic carbon to traps. *J. Exp. Bot.* 61, 99–103.

Sirová, D., Šantrůček, J., Adamec, L., Bárta, J., Borovec, J., Pech, J., Owens, S.M., Šantrůčková, H., Schäufele, R., Štorchová, H., Vrba, J., 2014. Dinitrogen fixation associated with shoots of aquatic carnivorous plants: is it ecologically important? *Ann. Bot.* 114, 125–133.

Sydenham, P.H., Findlay, G.P., 1973. The rapid movement of the bladder of *Utricularia* sp. *Aust. J. Biol. Sci.* 26, 1115–1126.

Sydenham, P.H., Findlay, G.P., 1975. Transport of solutes and water by resetting bladders of *Utricularia*. *Aust. J. Plant Physiol.* 2, 335–351.

Taylor, P., 1989. *The Genus Utricularia: A Taxonomic Monograph.* Kew Bulletin, Additional Series XIV. HMSO, London.

Thor, G., 1988. The genus *Utricularia* in the Nordic countries, with special emphasis on *U. stygia* and *U. ochroleuca*. *Nord. J. Bot.* 8, 213–225.

Vincent, O., Weisskopf, C., Poppinga, S., Masselter, T., Speck, T., Joyeux, M., Quilliet, C., Marmottant, P., 2011a. Ultra-fast underwater suction traps. *Proc. R. Soc. B* 278, 2909–2914.

Vincent, O., Roditchev, I., Marmottant, P., 2011b. Spontaneous firings of carnivorous aquatic *Utricularia* traps: Temporal patterns and mechanical oscillations. *PLoS ONE* 6, e20205.

Vincent, O., Marmottant, P., 2011. Carnivorous *Utricularia*: The buckling scenario. *Plant Signal. Behav.* 6, 1752–1754.

Vintéjoux, C., Shoar-Ghafari, A., 2005. Glandes digestives de l'Utriculaire, ultrastructures et fonctions. *Acta Bot. Gall.* 152, 131–145.

Wagner, G.M., Mshigeni, K.E., 1986. The *Utricularia*-Cyanophyta association and its nitrogen-fixing capacity. *Hydrobiologia* 141, 255–261.