Phyton (Austria) Special issue: "Plant Physiology"	**Vol. 39**	Fasc. 3	(181)-(185)	30. 11. 1999

ETS-Activity as a Measure of Vitality of Different Macrophyte Species

By

Zdenka MAZEJ[1] & Alenka GABERŠČIK[1]

Key words: Vitality of macrophytes, terminal electron transport (ETS) activity, dark respiration.

Summary

MAZEJ Z. & GABERŠČIK A. 1999. ETS-activity as a measure of vitality of different macrophyte species. - Phyton (Horn, Austria) 39 (3): (181) - (185).

The ETS (electron transport system) activity was examined in different macrophyte species. ETS activity shows metabolic potential of the plant while dark respiration rate provides an information on metabolic activity under given condition. In vivo, enzymes may be subjected to the other regulation mechanisms and actual respiration is lower. Certain organisms are capable of responding to the changes in the environment, changing enzyme concentration. *Batrachium trichophyllum,* which colonised shelter littoral and can also survive on the land, had high ETS activity potential whole the season. High values enable this species to survive under changing conditions. The species with wide ecological tolerance *Elodea canadensis, Myriophyllum spicatum, Potamogeton perfoliatus* and *Potamogeton crispus* reached high ETS activity, as well. The vitality of *Potamogeton crispus* was very low when the species was in decline whereas the young plants that appeared in the autumn had approximately 10 times higher ETS activity. *Fontinalis antipyretica* and *Chara* sp. exhibit the lowest ETS activity, because of relatively stable environment above all temperature conditions in deeper water layers. The ratio between actual dark respiration and the ETS activity for different species was calculated. Most species studied use only small part of respiration enzymes (DR/ETS is from 0.08 to 0.35) under unstressed conditions.

Introduction

Terminal electron transport system (ETS) is one of the most important biochemical process in cell metabolism. ETS activity is a measure of metabolic potential of certain tissue, while respiration rate provides only an information on metabolic activity under given condition (PACKARD 1971, G.-TÓTH & al. 1994). In

[1] National Institute of Biology, Večna pot 111, 1000 Ljubljana, Slovenia.

vivo enzymes are subjected to different regulation mechanisms and actual respiration is lower. Thus, an organism is capable to overcome changes or stress without changing enzyme concentration (BERGES & al. 1993).

The ETS activity measurements could be used for estimation of general metabolic activity of plankton, sediment, soil samples, individual organisms (G.-TÓTH 1993) and vitality of roots (URBANC-BERČIČ & GABERŠČIK 1997) or different tissue (GABERŠČIK & MAZEJ 1995, MAZEJ & GABERŠČIK 1996). Respiration/ETS activity (R/ETS) ratio shows the extend of the use of the enzyme potential under certain conditions. Organisms under the stress need more energy, that is why mytohondries increase the production of ATP and the consumption of O_2 increases. In present article ETS activity and dark respiration rate in different macrophyte species from different lakes were monitored in order to estimate their potential to cope with changes in specific habitat.

Material and Methods

Terminal electron transport system (ETS) activity

ETS activity was determined on first fully developed leaf. This determination bases on the reduction of the artificial electrons acceptor iodo-nitro-tetrazolium-chloride and on the spectrophotometrical measurement of the formazan production rate which is in principle, in a direct relation to the oxygen consumption of the investigated tissue. Plant material was homogenised in ice-cold homogenisation buffer in a potter (3-4 minutes; 500 rpm) and then sonicated (20-30 minutes; 40W). It is very important to tear cell membranes and mitochondria membranes during the homogenisation, whereas enzyme activity must not be disturbed. After this treatment the homogenate was centrifuged (4 minutes; 10.000 rpm; 0°C). Within 10 minutes triplicates of homogenate were incubated at 20°C, in substrate solution (NADH, NADPH, Triton-x-100) with iodonitrotetrazolium chloride (INT) for 40 minutes. After stopping the reaction formazan production was determined spectrophotometrically and ETS activity was calculated (KENNER & AHMED 1975, CHRISTENSEN & PACKARD 1979).

Measurements of oxygen consumption

O_2 exchange in dark was measured with Clark-type electrodes (Multichannel Oxymeter, WTW, Germany) on the upper parts of the plants under ambient temperature.

Plant material

Plants were collected in different lakes in different times of the growth season. The following species were included in the experiment: *Myriophyllum spicatum*, *Potamogeton crispus*, *Potamogeton alpinus*, *Batrachium trichophyllum*, *Chara* sp., *Fontinalis antipyretica*, *Elodea canadensis* and *Potamogeton perfoliatus*.

Results and Discussion

Batrachium trichophyllum, which colonises shelter littoral and also survive on the dry land showed the highest ETS activity throughout the whole vegetative season among all species examined. It disappeared for a while, but later on, new plants with very high respiration potential appeared again (Fig. 1). High values of ETS activity enable this species to survive in such changeable habitat. The representatives of this species are not very longliving and new plants are appeared very often (DAHLGREN & CRONBERG 1996). The species with wide ecological tolerance *Elodea canadensis*, *Myriophyllum spicatum* and *Potamogeton perfoliatus* reached high ETS activity, as well. The leaves of *Myriophyllum spicatum*, which overwintered exhibited very low ETS activity early in spring (Fig. 1). Young shoots showed much higher ETS activity. After that there was an obvious decline in summer, followed by an increase in autumn. *Chara* sp. showed similar course, but values were much lower. We determined the highest ETS activity in *Potamogeton crispus*, but only in the certain times of the season (May and September). It is known, that this species spreads very quickly (NICHOLS & SHAW 1986), what was also the case in our lakes. *Potamogeton crispus* usually disappears in summer.

Table 1. Dark respiration under ambient temperature condition (DR), ETS activity and DR/ETS activity ratio in different plant species (the annual mean ± SE).

Species	Dark respiration $mgO_2/gDW/h$	ETS activity $mgO_2/gDW/h$	DR/ETS activity ratio
Chara sp.	0.64 ± 0.09	3.27 ± 0.44	0.25 ± 0.04
Fontinalis antipyretica	0.93 ± 0.77	3.32 ± 1.06	0.26 ± 0.23
Batrachium trichophyllum	1.83 ± 0.26	10.8 ± 2.12	0.23 ± 0.04
Elodea canadensis	1.09 ± 0.12	12.2 ± 0.43	0.10 ± 0.01
Myriophyllum spicatum	1.20 ± 0.22	7.86 ± 1.20	0.24 ± 0.05
Potamogeton alpinus	1.75 ± 0.29	6.08 ± 1.32	0.29 ± 0.03
Potamogeton crispus	1.37 ± 0.19	11.02 ± 1.21	0.16 ± 0.05
Potamogeton perfoliatus	1.10 ± 0.77	10.4 ± 7.4	0.16 ± 0.14

ETS activity of the plants was very low when the species was in decline whereas the activity of young sprouts in autumn was approximately 10 times higher. The ETS activity is becoming lower with the age of the leaves. Higher ETS activity in young leaves is also a consequence of higher ratio of metabolic active/metabolic inactive tissues. Moss *Fontinalis antipyretica* and *Chara* sp. exhibit low ETS activity, because of relatively stable environment above all temperature conditions in deeper water layers.

Mosses have low metabolic rate and even under optimal conditions grow slower than angiospermae (PEŇUELAS & al. 1988). The ratio between dark respiration and the ETS activity for different species was calculated (Table 1). Most species usually use only small part of respiration enzymes. The ratio DR/ETS activity was between 0.08 and 0.35, what could be compared with the data in the literature, as well (PEŇUELAS & al. 1988). PEŇUELAS & al. 1988 obtained a ratio between 0.18 and 0.38.

The ratio was changing throughout the year; it was much higher in the summer period due to higher temperature. At that time actual respiration increased due to stress conditions what revealed also from actual/potential respiration ratio.

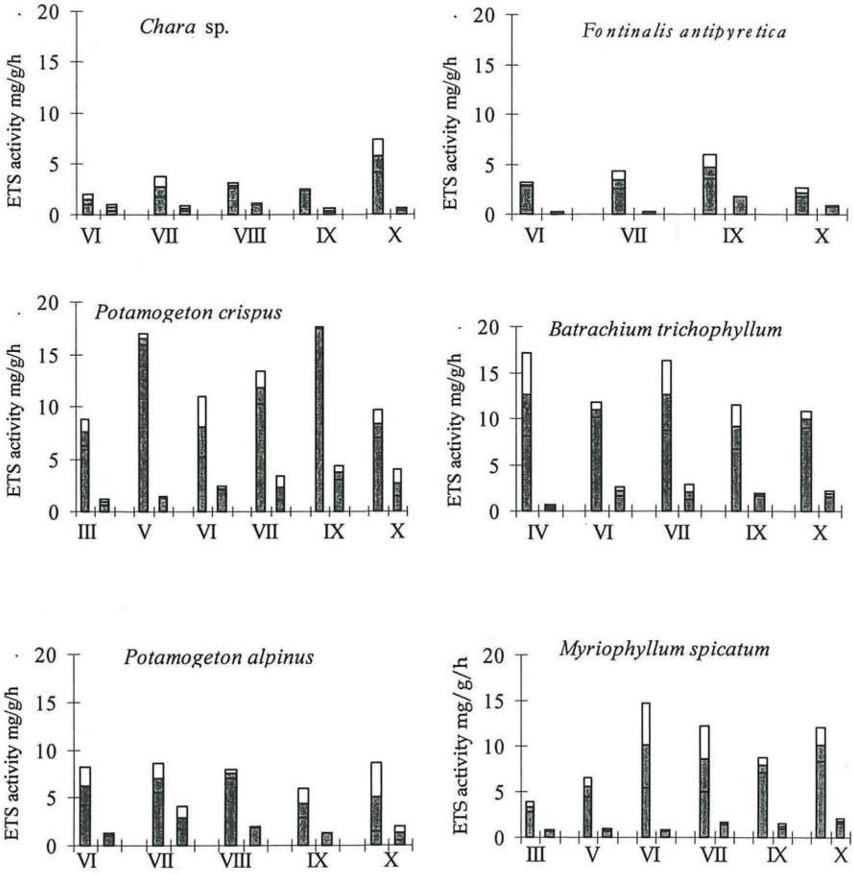

Fig. 1. ETS activity in the leaves and dark respiration of the upper parts of the shoots of different macrophytes *Potamogeton crispus, Batrachium trichophyllum*, *Potamogeton alpinus, Myriophyllum spicatum*, *Chara* sp. and *Fontinalis antipyretica* throughout the vegetative season (from March (III) to October (X)) 1997 (average ± SE, n= 9-15).

References

BERGES J.A., ROFF J.C. & BALLANTYNE J. S. 1993. Enzymatic indices of respiration and ammonia excretion: relation to body sizes and food levels. - Plankton Res. 15: 239-254.

CHRISTENSEN J.P. & PACKARD T.T. 1979. Respiratory electron transport activities in phytoplancton and bacteria: comparison of methods. - Limnol. Oceanogr. 24: 576-583.

DAHLGREN G. & CRONBERG L. 1996. Species differentiation and relationship in *Ranunculus* subgenus *Batrachium* (*Ranunculaceae*) elucidated by isozyme electrophoresis. - Symb. Bot. Ups. 31(3): 91-104.

G.-TÓTH L. 1993. Measurement of the terminal electron system (ETS)-activity of plancton and sediment. - Proceedings of the ILEC/UNEP International Training Course: 149-152.

— , LANGÓ Z., PADISÁK J. & VARGA E. 1994. Terminal electron transport system (ETS)-activity in the sediment of Lake Balaton, Hungary. - Hydrobiologia 281: 129-139.

GABERŠČIK A. & MAZEJ Z. 1995. Photosynthetic performance and photoinhibition in two species of *Potamogeton* from lake Bohinj (Slovenia). - Acta bot. Gallica 142 (6): 667-672.

KENNER A.A. & AHMED S.I. 1975. Measurements of electron transport activities in marine phytoplankton. - Mar. Biol. 33: 119-127.

MAZEJ Z. & GABERŠČIK A. 1996. The vitality of some macrophyte species in the watercourse Ižica at different time of the year. - Proceedings of the International Group Meeting IAD-SIL. September 1 - 4, 1996 Bohinj, Slovenia: 47-51.

NICHOLS S. A. & SHAW B. H. 1986. Ecological life histories of the three aquatic nuisance plants, *Myriophyllum spicatum*, *Potamogeton crispus*, and *Elodea canadensis*. - Hydrobiologia 131: 3-21.

PACKARD T.T. 1971. The measurement of respiratory electron-transport activity in marine phytoplankton. - Journal of Marine Research 29 (3): 235-234.

PEÑUELAS J., MURILLO J. & AZCÓN-BIETO J. 1988. Actual and potential dark respiration rates and different electron transport pathways in freshwater aquatic plants. - Aquatic Botany 30: 353-362.

URBANC-BERČIČ O. & GABERŠČIK A. 1997. Reed stands in constructed wetlands: "Edge effect" and photochemical efficiency of PS II in common reed. - Wat. Sci. Tech. 5: 143-147.