

SUGARS IN FERNS AND LYCOPHYTES GROWING ON ROCKY OUTCROPS FROM SOUTHEASTERN BRAZILIAN COAST

AÇÚCARES EM SAMAMBAIAS E LICÓFITAS CRESCENDO SOBRE AFLORAMENTOS ROCHOSOS DA COSTA SUDESTE BRASILEIRA

Moemy Gomes de MORAES¹; Alexandre Alberto Queiroz de OLIVEIRA²; Marcelo Guerra SANTOS³

1. Universidade Federal de Goiás, Instituto de Ciências Biológicas, Goiânia, GO, Brasil. moemy@ufg.br; 2. Biólogo, Fundação Centro de Ciências e Educação Superior a Distância do Estado do RJ, Rio de Janeiro, RJ, Brasil; 3. Universidade do Estado do Rio de Janeiro, Faculdade de Formação de Professores, São Gonçalo, RJ, Brasil.

ABSTRACT: The extreme conditions in rocky outcrops allow the occurrence of desiccation tolerant species. One strategy of these plants to withstand water shortage is the accumulation of sugars. In this paper, we report sugar levels and profile of three ferns and one lycophyte naturally hydrated growing on rocky outcrops from Southeastern Brazil. *Anemia* species have higher sugar contents than *Doryopteris collina* and *Selaginella sellowii*. The analyzed species have different sugar profiles. The ferns have glucose, fructose and sucrose, and the lycophyte has glucose and trehalose.

KEYWORDS: Ferns. Lycophytes. Resurrection plants. Sucrose. Trehalose.

Rocky outcrops (inselbergs) have strict environmental conditions, among them edaphic dryness. These habitats are centers of diversity for desiccation-tolerant vascular plants, and monocotyledons, ferns and lycophytes are the most notable taxa (POREMBSKI; BARTHLOTT, 2000). Desiccation tolerance in vegetative parts occurs in less than 0.2 % of the vascular flora, allowing them to survive 80-90% of water loss. Desiccation tolerant plants have different strategies to endure water scarcity, among them the presence of sugars, typically high levels of disaccharides (PROCTOR; TUBA, 2002). In this paper, we report sugar levels and profile of naturally hydrated ferns and lycophyte growing on rocky outcrops from southeastern Brazilian coast.

Samples were collected in the rocky outcrops of Morro das Andorinhas (22°58'25" S 43°2'32" W), Serra da Tiririca State Park, Rio de Janeiro state, Brazil. The following species were selected for this study: the ferns *Anemia tomentosa* var. *anthriscifolia* (Schrad.) Mickel, *Anemia villosa* Humb. & Bonpl. ex Willd. (both Anemiaceae), *Doryopteris collina* (Raddi) J.Sm. (Pteridaceae) and the lycophyte *Selaginella sellowii* Hieron. (Selaginellaceae) According to Meirelles et al. (1997) these species are desiccation tolerant. The collection was performed in summer, and there was 12 mm of rainfall in the past 10 days prior to the collection (www.inmet.gov.br).

Four fully expanded turgid leaves of ferns and shoot segments with approximately 10 cm of the lycophyte were harvested from four individuals for each species. Aerial parts were sampled from

9:00 - 12:00 am. Relative water content (RWC) was determined gravimetrically (BARR; WEATHERLEY, 1962). Total sugars (TS) were determined in ethanolic extracts by the anthrone method using glucose as standard (YEMM; WILLIS, 1954). TS profiles were obtained in deionized extracts by High Performance Anion Exchange Chromatography with pulsed amperometric detection (HPAEC-PAD) in a CarboPac PA-1 column (Dionex, Sunnyvale, CA). The elution was a gradient of NaOH according to Santos et al. (2004). The identification of individual sugars was made by comparison with the retention time of authentic standards under the same analytical conditions.

The species were collected naturally hydrated as expressed by the RWC which varied from 80-90% (Table 1). These RWC values are the expected for actively growing plants (GAFF; OLIVER, 2013). Gaff (1987) reported RWC levels lower than 8% for desiccated *A. tomentosa*, *Doryopteris* and *Selaginella* species in South American rocky outcrops. *Anemia caffrorum* (L.) Christenh., a desiccation tolerant fern, is able to survive at 8% RWC in desiccated leaves (FARRANT et al., 2009).

In this hydrated state, the TS levels in leaf blades of *Anemia* species were higher than 20% (Table 1). This content may be the result of an operating photosynthesis since the leaves were fully expanded and hydrated. The exceeding photoassimilates are exported to meet the demand in other parts, like the synthesis and storage of starch grains found in the petiole of *A. villosa* and *A.*

tomentosa (RIBEIRO et al., 2007). *D. collina* and *S. sellowii* have around 8% of TS of the dry weight. Ghasempour et al. (1998) reported 0.69 to 4.79 % (in hydrated) and 0.64% to 5.2% (in dehydrated) for desiccation tolerant angiosperms. *Selaginella*

tamariscina (P. Beauv.) Spring had higher levels of soluble carbohydrate (~ 15%) either in hydrated or desiccated leaves (LIU et al., 2008).

Table 1. Relative water content (RWC) and total sugars (TS) of ferns and lycophyte growing in rocky outcrops from Southeastern Brazil. Data expressed as mean (\pm SE), n=4. Different letters in columns indicate significant differences ($P < 0.00924$; $N = 4$; Kruskal-Wallis - Student-Newman-Keuls *post-hoc* test).

Species	RWC (%)	TS ($\text{mg.g}^{-1}\text{DW}$)
<i>A. tomentosa</i>	80.0 \pm 4.6 a	200.92 \pm 30.19 a
<i>A. villosa</i>	87.8 \pm 2.5 a	235.42 \pm 19.02 a
<i>D. collina</i>	88.3 \pm 2.3 a	81.31 \pm 19.85 b
<i>S. sellowii</i>	90.0 \pm 6.3 a	87.16 \pm 15.62 b

Sucrose, glucose and fructose were detected in *A. tomentosa*, *A. villosa* and *D. collina* (Figure 1). Sucrose accumulates in dried leaves of some desiccation tolerant species, as the fern *Anemia caffrorum* (FARRANT et al., 2009). This disaccharide is recognized as the main protectant in desiccation tolerant species (GAFF; OLIVER, 2013).

Trehalose is present in *S. sellowii* (Figure 1). This disaccharide has an extensive biotechnological potential and is typical of desiccation tolerant organisms, ranging from

bacteria to lycophytes (CROWE et al., 1998). However, this sugar is not necessarily required for desiccation tolerance in *Selaginella* since it occurs in desiccation sensitive *Selaginella moellendorffii* Hieron. (YOBI et al., 2012).

Our results were the first report on the sugars of South American desiccation tolerant species. *Anemia* species have higher sugar contents than *D. collina* and *S. sellowii*. The sugar profile is distinct in both groups: ferns have glucose, fructose and sucrose while the lycophyte has glucose and trehalose.

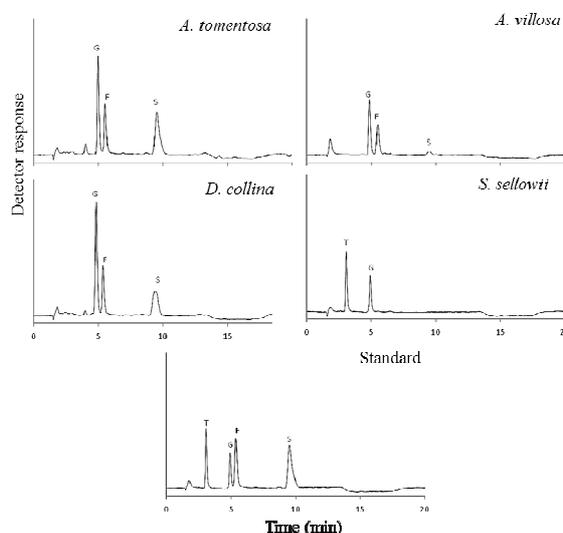


Figure 1. Total soluble carbohydrate profile of aerial parts of ferns and lycophyte from rocky outcrops in Southeastern Brazil, obtained by HPAEC-PAD. G - glucose, F- fructose, S- sucrose, T- trehalose.

ACKNOWLEDGEMENTS

The authors thank to FAPERJ (Rio de Janeiro, Brazil) for the financial support.

RESUMO: Nas condições ambientais extremas dos afloramentos rochosos ocorrem espécies tolerantes à dessecação. Uma das estratégias dessas plantas para resistir à restrição hídrica é o acúmulo de açúcares. No presente

estudo, avaliamos a concentração e o perfil de açúcares de três espécies de samambaias e uma licófitas, naturalmente hidratadas crescendo sobre afloramentos rochosos no sudeste do Brasil. As espécies de *Anemia* possuem maior concentração de açúcares em relação a *Doryopteris collina* e *Selaginella sellowii*. As espécies analisadas mostraram perfis distintos de açúcares. As samambaias apresentam glicose, frutose e sacarose, enquanto a licófitas glicose e trealose.

PALAVRAS-CHAVE: Licófitas, Plantas Revivescentes, Sacarose, Samambaias. Trealose.

REFERENCES

- BARR, H.; WEATHERLEY, P. A re-examination of the relative turgidity technique for estimating water deficit in leaves. **Aust. J. Biol. Sci.**, East Melbourne, v. 15, n. 3, p. 413-428, 1962.
- CROWE, J. H.; CARPENTER, J. F.; CROWE, L. M. The role of vitrification in anhydrobiosis. **Annu. Rev. Physiol.**, Palo Alto, v. 60, n. 1, p. 73-103, 1998.
- FARRANT, J. M.; LEHNER, A.; COOPER, K.; WISWEDEL, S. Desiccation tolerance in the vegetative tissues of the fern *Mohria caffrorum* is seasonally regulated. **Plant J.**, Oxford, v. 57, n. 1, p. 65-79, 2009.
- GAFF, D. Desiccation tolerant plants in South America. **Oecologia**, Berlin, v. 74, n. 1, p. 133-136, 1987.
- GAFF, D. F.; OLIVER, M. The evolution of desiccation tolerance in angiosperm plants: a rare yet common phenomenon. **Funct. Plant Biol.**, Collingwood, v. 40, n. 4, p. 315-328, 2013.
- GHASEMPOUR, H.; GAFF, D. F.; WILLIAMS, R. P. W.; GIANELLO, R. D. Contents of sugars in leaves of drying desiccation tolerant flowering plants, particularly grasses. **Plant Growth Regul.**, Dordrecht, v. 24, n. 3, p. 185-191, 1998.
- LIU, M. S.; CHIEN, C. T.; LIN, T. P. Constitutive components and induced gene expression are involved in the desiccation tolerance of *Selaginella tamariscina*. **Plant Cell Physiol.**, Kyoto, v. 49, n. 4, p. 653-663, 2008.
- MEIRELLES, S. T.; MATTOS, E. A.; SILVA, A. C. Potential desiccation tolerant vascular plants from southeastern Brazil. **Pol. J. Environ. Stu.**, Olsztyn, v. 6, n. 4, p. 17-21, 1997.
- POREMBSKI, S.; BARTHLOTT, W. Granitic and gneissic outcrops (inselbergs) as centers of diversity for desiccation-tolerant vascular plants. **Plant Ecol.**, Dordrecht, v. 151, p. 19-28, 2000.
- PROCTOR, M. C.; TUBA, Z. Poikilohydry and homoihydry: antithesis or spectrum of possibilities? **New Phytol.**, Lancaster, v. 156, n. 3, p. 327-349, 2002.
- RIBEIRO, M. L. R.; SANTOS, M. G.; MORAES, M. G. Leaf anatomy of two *Anemia* Sw. species (Schizaeaceae-Pteridophyte) from a rocky outcrop in Niterói, Rio de Janeiro, Brazil. **Rev. Bras. Bot.**, São Paulo, v. 30, n. 4, p. 695-702, 2007.
- SANTOS, H. P.; PURGATTO, E.; MERCIER, H.; BUCKERIDGE, M. S. The control of storage xyloglucan mobilization in cotyledons of *Hymenaea courbaril*. **Plant Physiol.**, Rockville, v. 135, n. 1, p. 287-299, 2004.
- YEMM, E.; WILLIS, A. The estimation of carbohydrates in plant extracts by anthrone. **Biochem. J.**, London, v. 57, n. 3, p. 508-514, 1954.
- YOBI, A.; WONE, B. W.; XU, W.; ALEXANDER, D. C.; GUO, L.; RYALS, J. A.; OLIVER, M. J.; CUSHMAN, J. C. Comparative metabolic profiling between desiccation-sensitive and desiccation-tolerant species of *Selaginella* reveals insights into the resurrection trait. **Plant J.**, Oxford, v. 72, n. 6, p. 983-999, 2012.