Supplementary Materials

Manuscript title

Distinct reproductive strategy of two endemic Amazonian quillworts

Authors

Cecilio F. Caldeira[1]*; Arthur V. S. Lopes[2]; Keyvilla C. Aguiar[1]; Aline L. Ferreira[2]; João V.S. R. Araujo[2]; Vinnícius M. S. Gomes[2]; Daniel B. Zandonadi[2]; Cinthia B. Abranches[1]; Silvio J. Ramos[1]; Markus Gastauer[1]; Naiara V. Campos[2]; Lísia M. S. Gestinari[2]; Luis A. Prado[2]; Fernando Marino Gomes dos Santos[3]; Rodrigo L. Martins[2]; Francisco A. Esteves[2]; Guilherme Oliveira[1]; Mirella P Santos[2]*

Author affiliations

[1]Instituto Tecnológico Vale – Belém 35400-000, Pará, Brazil

[2]Instituto de Biodiversidade e Sustentabilidade NUPEM/UFRJ – Macaé 27900-000, Rio de Janeiro, Brazil

[3] Environmental Studies Office, Vale, Belo Horizonte, Minas Gerais, Brazil

***Corresponding author**

cecilio.caldeira@itv.org, Instituto Tecnológico Vale. Boaventura da Silva, 955. Belém/PA, Brazil

Table S1. Experimental setup applied to examine the sexual reproduction of two *Isoëtes* L. species endemic to Serra dos Carajás/Brazil.

#	Species	Number of plants	Number of reps	Number of megaspores	Total of megaspores
1	*Isoëtes serracarajensis*	2	13 (5-4-4)*	23 - 40	448
2	*Isoëtes cangae*	6	34 (17-12-5)	15 - 50	1280
3	*Isoëtes cangae*	2	12 (4-4-4)	40 - 50	540
4	*Isoëtes cangae*	3	21 (6-6-2)	50	700
5	*Isoëtes cangae*	2	12 (4-4-4)	50 - 100	1000

*Number of repetitions per treatment are shown between parentheses for self-fertilization, cross-fertilization and apomixis.

Supplementary Materials

Table S2. Description of ISSR polymorphic primers validated for the genus *Isoetes*.

ID	SEQUENCES	At (°C)	TNL
LB(14)	5’- GAGAGAGAGAGAGAGAT- 3’	45,5 °C	10
LB(10)	5’- ACACACACACACACACG- 3’	53 °C	7
LB(6)	5’- ACACACACACACACACCC- 3’	52 °C	9
LB(5)	5’- AGAGAGAGAGAGAGAGYT- 3’	52 °C	8
LB(52)	5’-ATCATCATCATCATCATCC-3’	48 °C	8
LB (18)	5-AGAAGAAGAAGAAGAAGAAGAAGAYC-3’	50 °C	10

ID: Identification of the primers Sequences (5’-3’); At (°C): anneling temperature; TNL: total number of loci

Supplementary Materials

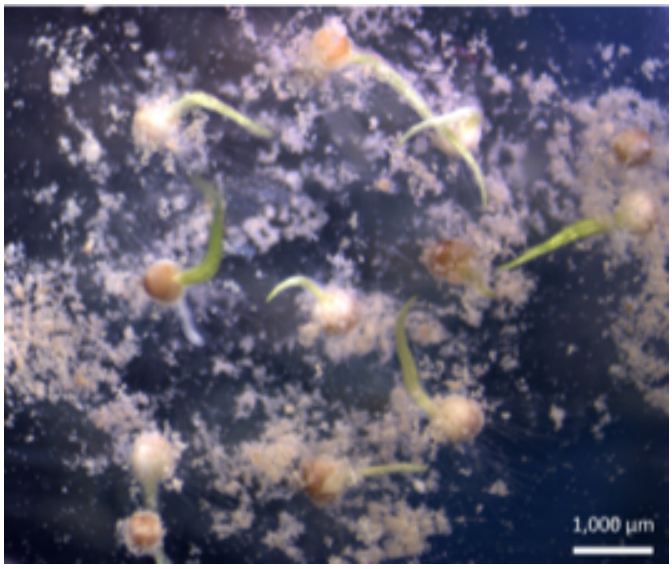

Figure S1. A subset of *Isoëtes cangae* germinated megaspores from apomixis treatment (isolated for 15 days after rhizoid emission and archegonia appearance) that developed into sporophytes a few days after microspore addition.

Supplementary Materials

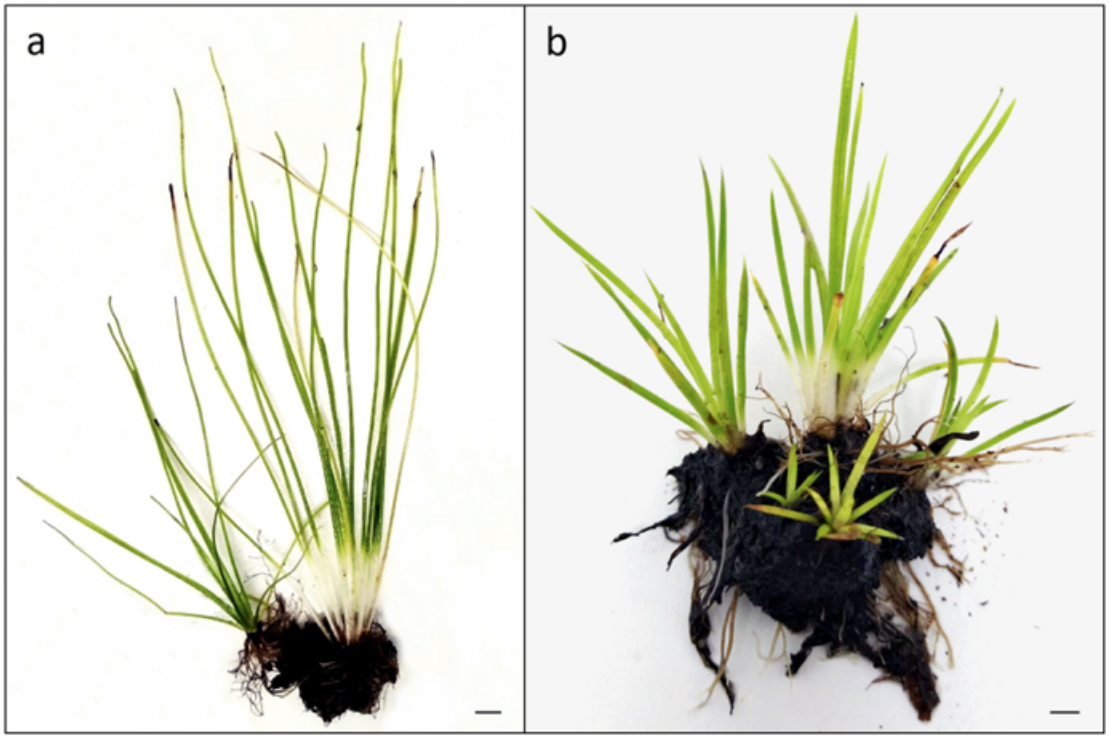

Figure S2. Mother plants and their respective sporophytes of *Isoëtes cangae* (a) and *Isoëtes serracarajensis* (b) reproduced in greenhouse conditions.

Supplementary Materials

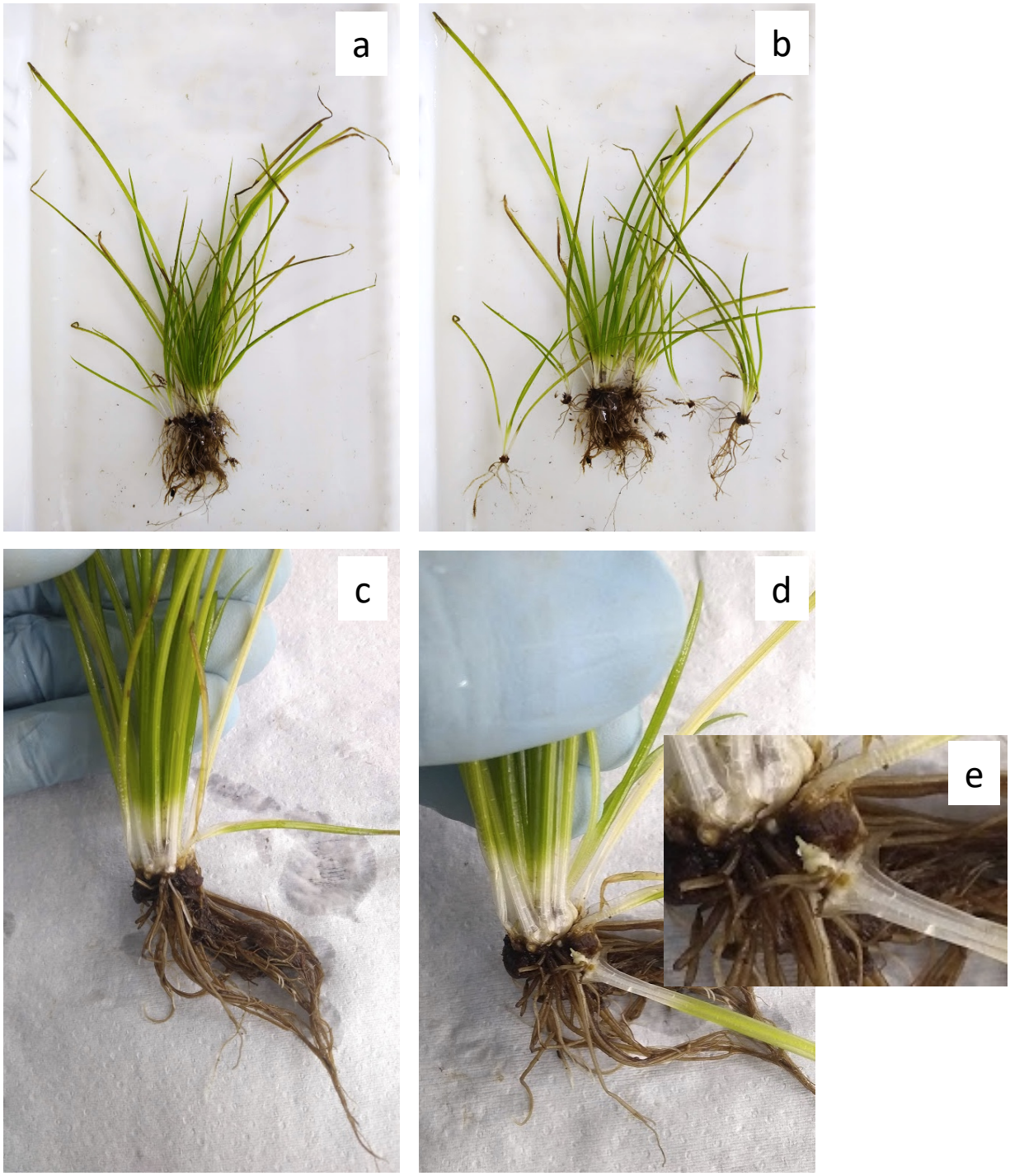

Figure S3. The vegetative reproduction of *Isoëtes serracarajensis*. (a) and (b) New sporophytes (tillers) attached to the mother plant that can easily be detached. (c) and (d) Another adult plant and its respective tillers. (e) Details of root emission from the new corm fragment at the leaf base.