

Research Article

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Is *Pitcairnia halophila* really a halophyte? Evidence from a germination and growth experiment

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Abstract

Many bromeliads occur in habitats that are potentially affected by salt. *Pitcairnia halophila* is a particularly interesting case in this context due to its growing sites in the spray zone of coastal cliffs in Central America. We conducted an experimental/observational study that explored the physiological and ecological basis of the occurrence of this species. Germination of *P. halophila* was as affected by increasing salinity as that of congeneric species from other natural growing sites, but seedling growth was less impaired in comparison and remained positive even at exposure to half-strength seawater. Interestingly, *in situ* plant nutrient concentrations documented over an entire growing season suggest that this capacity to tolerate salt stress is not tested at the natural growing site – Na⁺ concentrations were consistently low in leaves and shoots and comparable to those of seedlings watered with distilled water in the laboratory. We conclude that *P. halophila* shows the physiological ability to deal with medium levels of salinity, but ecological conditions at the humid tropical growing site allow these plants to avoid salt stress entirely in spite of the close proximity to the sea.

Introduction

The more than 3,000 species in the family Bromeliaceae can be found in diverse habitats (Benzing 2000), most prominently as epiphytes in many tropical and some temperate zone forests (Díaz & Alvarez 2009; Zotz & Schultz 2008), but also terrestrially and rupicolously, for example, in tropical alpine vegetation up to 4000 m a.s.l. (Miller & Silander 1991), on rocky outcrops in campo rupestre (Alves *et al.* 2008) or in shrubby restinga vegetation (Cogliatti-Carvalho *et al.* 2001). Bromeliads can be also quite prominent in tropical coastal vegetation, for example, as epiphytes in mangroves (Díaz-Jiménez 2007; Zotz 2005), growing rupicolously at rocky shores (e.g., *Pitcairnia* or *Dyckia*, Grant 1994; Lüttge *et al.* 1986; Pompelli *et al.* 2006), or in alluvial plains (e.g., *Bromelia humilis*, Medina *et al.* 1989). In the latter habitats, soil-rooted plants can get regularly into direct contact with seawater (Gómez & Winkler 1991), or saline conditions may develop at least seasonally (Medina *et al.* 1989). Even epiphytically growing bromeliads may suffer from salt stress close to the coast when salt spray reaches these plants growing at some height in trees (Griffiths *et al.* 1989; Zotz & Reuter 2009).

However, although there are quite a few bromeliad species that are potentially affected by salt stress in their natural environment, this aspect of their ecology has received little attention (Benzing 2000). *Pitcairnia halophila* L.B.Sm. is a particularly interesting member of the family in the context of salt tolerance, because the species is typically found on cliffs just above the high tide level (Fig. S1, Grant 1994 and G. Zotz, pers. obs.). Given such a growing site, regular exposure to salt spray and even occasional inundation by salt water seem more than likely. There is some information on the physiology of congeneric *Pitcairnia integrifolia*, which is growing in a similar habitat in Trinidad (Lüttge *et al.* 1986). These researchers documented an osmotic pressure of only 0.9 MPa in leaves, which was interpreted as evidence against substantial accumulation of NaCl. In that study, it was also shown that the foliar trichomes were not functional in nutrient and water uptake, which allows the plants to avoid the foliar uptake of salt spray, although this does not explain why such uptake should not occur via roots.

The present study focuses on the effect of salt (mostly sodium chloride) on *Pitcairnia halophila* with a particular emphasis on the early phases of ontogeny that are known to be particularly sensitive to salt stress (Egan & Ungar 1999; Larcher 2003). Both germinating seeds and young plants of *Pitcairnia halophila* and three congenics, which naturally grow in habitats without any input of NaCl, were subjected to different levels of salinity. We show that similar to most other plants found in saline environments (Ungar 1978), germination is quite susceptible to saline conditions via an osmotic effect, but vegetative growth of *Pitcairnia halophila* does not cease until salt concentrations in the soil exceed those equivalent to 50% seawater.

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To provide some ecological context for the results of these laboratory experiments, we also sampled mature *Pitcairnia halophila* individuals over an entire growing season. Remarkably, in spite of their growth in close proximity to the sea, these plants showed consistently low tissue concentrations of sodium.

Materials and methods

Habitat and study species

Pitcairnia halophila L.B.Sm. was originally described by Smith (1964). For a long-time considered endemic to Costa Rica (Grant 1994), it has since been found in neighbouring Panama as well (Cáceres *et al.* 2011). Restricted to a narrow margin along the Pacific coast, typically found on cliffs just above the high tide level, some exposure to salt spray or even occasional inundation by salt water seems a matter of fact and was clearly the reason for the choice of epithet (Grant 1994). For this study, both seed material and entire plants were collected at a locality near David, Panama. The local climate is characterised by 1360 mm of annual precipitation with a pronounced dry season in the first 4 months of the year and average temperatures of 28 °C with little seasonal variation (data are from a station located c. 61 km from our field site, Hidrometeorología ETESA, <https://www.hidromet.com.pa/>, data of 54 years since 1967). After collection of mature capsules, seeds were kept frozen at –20 °C at the University of Oldenburg until the start of the experiments. Plant collections to assess tissue concentrations were also done at the same site. Exact procedures are described below.

Pitcairnia heydlauffii R. Vasquez & P. L. Ibsch has been described relatively recently from Bolivia (Vasquez & Ibsch 2000). It occurs naturally in very wet forest on the foothills of the lower Andes. Seeds came from the Botanical Garden Berlin-Dahlem.

Pitcairnia schultzei Harms occurs in lowland forests in Panama, Colombia and Venezuela (Cáceres *et al.* 2011; WCVP 2020). The seed source was identical to *P. heydlauffii*.

Pitcairnia suaveolens Lindl. is endemic to southeast Brazil, where it mostly grows as a lithophyte along streams in Atlantic forest in the state of Rio de Janeiro (Saraiva *et al.* 2015). Seeds were provided by the Blijdorp Zoo, Rotterdam, NL.

Germination experiments

After surface sterilisation following the procedure described by Pickens *et al.* (2003), seeds of all four species were sown in disposable Petri dishes (100 × 15 mm) with filter paper (Machery-Nagel, 651 mm, Ø 83 mm). Each Petri dish was filled with 2 ml of solutions with increasing salt concentrations, from 10 to 50% full seawater concentration. We prepared the solutions from a commercially available concentrate (Tropic Marin®, Dr. Biener Aquarientechnik, Lauterbach, Germany), which is manufactured from chemically pure salts. The stock solution (equivalent to 100% seawater) was dominated by NaCl (10.7 g l⁻¹ Na⁺, 19.3 g l⁻¹ Cl⁻) with all other elements (Ca²⁺, K⁺ and SO₄²⁻) and 70 trace minerals also in natural concentrations. Controls received distilled water. Each Petri dish contained 20 seeds, and each treatment level and the control were replicated 5 times. The Petri dishes, which were sealed with Parafilm® (Bemis, Neenah, Wisconsin, USA), were kept in climate cabinets (Economic Deluxe, Snijders Scientific, Tilburg, the Netherlands) with a light–dark period of 12/12 h (photon flux density c. 60 μmol m⁻² s⁻¹) and a temperature of 25 °C for 28 days. Weighing the dishes before and after that period

indicated minimal water loss, and hence we expect hardly any increase in concentration during the course of the experiment. Germination (= appearance of a whitish radicle contrasting with the brownish testa) was assessed every other day with a dissecting scope (Zeiss 57 50 57, Jena, Germany).

Seawater can impact biological processes via an osmotic and/or an ionic effect. To study the osmotic effect alone, we conducted a second germination experiment with *P. halophila*, using mannitol to establish well-defined water potentials. Mannitol concentrations were varied between 0.04, 0.1, 0.2, 0.4 and 0.6 mol l⁻¹ to generate substrate potentials of 0.1, 0.25, 0.50, 1.0 and 1.5 MPa (Swagel *et al.* 1997). Since the seawater treatments can be expressed as water potentials in a similar way (e.g., 100% seawater has a water potential of about –2.5 MPa, Tomlinson 1986), it is possible to compare the effect of seawater and mannitol on germination, that is, distinguish osmotic and ionic effects.

Growth experiments and nutrient analysis

Seeds were allowed to germinate as described above for the controls of the germination experiment. Once reaching a size of a few mm, juvenile plants were transferred into planting trays (35 × 40 cm) filled with 50/50 (vol %) unfertilised potting soil and sand and were kept in the green house. Ambient conditions were 25–28 °C air temperature, 60–80 % relative humidity and natural light. After reaching a size of c. 2 cm maximum leaf length (LL), plants were transferred to individual pots, which were filled with the same substrate, for at least 1 week before the beginning of experiments. Many ecophysiological responses depend on plant size (Zotz *et al.* 2001). Therefore, in both *Pitcairnia halophila* and *P. suaveolens*, growth experiments were conducted with plants of different size (c. 2 cm and c. 11–15 cm LL). Pot size was 20 cm³ for small and 700 cm³ for large plants, respectively.

Growth experiments also lasted for 28 days. Sample size was 20 per treatment level for small (c. 2 cm LL) plants and 10 per level for large (c. 11–15 cm LL) plants. Plants were watered three times a week with different solutions (10–50 or 60% full seawater concentration and controls (= distilled water)). A diluted standard fertiliser was added to all treatments once a week (Flory 3: 15-10-15, Euflo, Munich, Germany), equivalent to 0.05 mg l⁻¹ NO₃-N, 0.1 mg NH₄-N l⁻¹, 0.1 mg P l⁻¹ (phosphate) and 0.15 mg K l⁻¹. Before the experiment, we had quantified the amount of water needed to saturate the soil in the pots at each watering. Watering each pot with about 10 times this quantity made sure that salt concentrations would not increase over time, and no crusts developed even in the treatment with the highest salt concentrations.

To estimate the initial size of all experimental plants, we took digital photographs from above. Similar photographs were taken from an additional 10 plants per species and experiment, which covered the entire size range of the experimental plants. These 10 plants were then harvested, and fresh weights of shoots and roots were determined. After 48 h at 80 °C, dry mass was determined. The photographs were used to quantify the projected area of each plant with Image J (Rasband 1997-2015). The correlation between projected area and plant dry mass was invariably very tight ($r > 0.95$) and could be used to estimate the initial dry mass of all experimental plants. At the end of the experiment, all surviving plants were harvested, and fresh and dry weights were determined as described above. This plant material was then used to determine the foliar Na⁺ and K⁺ concentrations for each species, plant size class and treatment (4–6 replica). After digestion of the

samples following standard procedures (Allen 2008), tissue Na^+ and K^+ concentrations were determined with an atomic absorption spectrophotometer (AAS, SpectrAA 300, Varian Inc., Darmstadt, Germany) at standard operating conditions.

In situ growth and nutrient concentrations

Over the course of one growing season from April 2019 to January 2020, we collected five times three entire plants at a locality near David, Panama. Plants were immediately transported to the laboratory in the Chiriquí University Science Faculty (Universidad Autónoma de Chiriquí, David, Panamá), where fresh weights of leaves and shoots were determined. Then subsamples were taken, weighed, and dried at 80°C for 48 h. The ratio of dry and fresh weight of the subsamples allowed the calculation of the dry weight of the entire plants. Subsamples were then transported to Germany, where tissue Na^+ and K^+ concentrations were determined with an AAS as described above.

Statistical analysis

All analyses were conducted with R version 4.0.2 (R Development Core Team 2019).

Germination

Germination success after 28 days (= % of germinated seeds) was used as response variable. The effect of seawater and mannitol on germination was assessed by calculating the proportional reduction in germination success at varying salt or mannitol concentrations as compared to controls in distilled water. This reduction was expressed as percentage. Since the different salt and mannitol concentrations are equivalent to defined water potentials, it was possible to directly compare their effects on germination. Without clear expectation on the specific relationship between water potentials and response variable, we simply chose the regressions that gave the best fit for each data set, that is, linear regressions, a polynomial and an exponential regression.

Growth

For each plant, the relative growth rate (RGR) in $\% \text{ day}^{-1}$ was estimated following Hunt (1982) as:

$$\text{RGR} = (\ln DW_e - \ln DW_b) / \Delta t,$$

that is, the difference between plant dry weight at the end and the beginning of the experiment divided by Δt , that is, the number of days. The relationship of RGR and different salt concentrations in the irrigation solutions was assessed with linear regressions.

Foliar nutrient concentrations

The relationship of foliar nutrient concentrations and the salt concentrations in the watering solutions was clearly linear in smaller and larger *P. halophila* and was therefore assessed with linear regression models. Due to a divergent, non-linear response of *P. suaveolens* to varying salt concentrations in the medium, the data of that species were analysed with two one-way ANOVAs in the case of Na^+ , while there was clearly no change in K^+ concentrations with varying seawater concentrations.

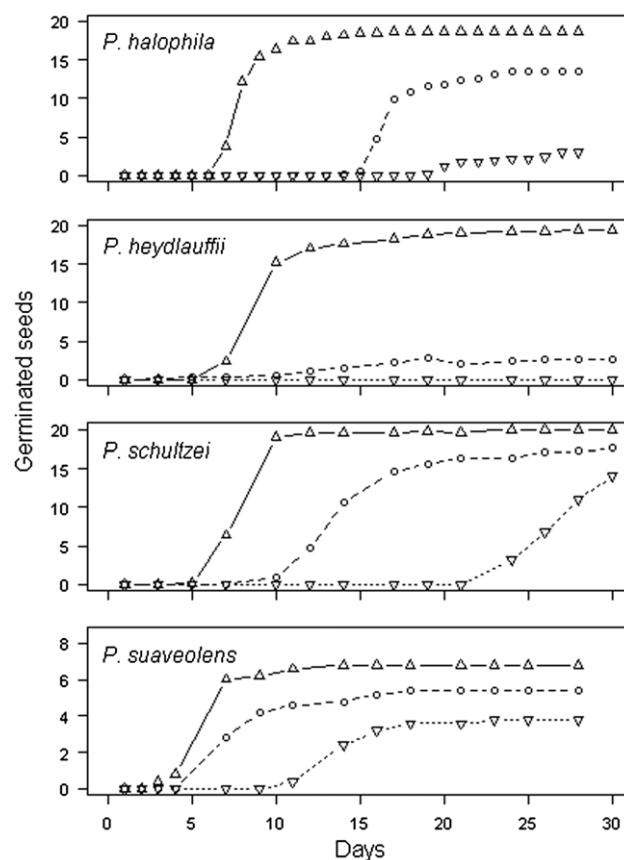


Figure 1. Cumulative germination of seeds of *Pitcairnia halophila*, *P. heydlauffii*, *P. schultzei* and *P. suaveolens*. Data are means of 5 Petri dishes with 20 seeds each. Different symbols indicate the used salt concentrations: distilled water (Δ), 30% seawater (\circ) and 50% seawater (∇). Note that only selected treatment levels are shown for reason of clarity. The y-axes differ because absolute germination was low in *Pitcairnia suaveolens* even in the controls.

Results

Germination

P. halophila germination of the controls was complete after c. 2 weeks (Figure 1). Maximum germination in the three congeners was reached after a similar period, although in one species, *P. suaveolens*, only about a third of all seeds germinated. Both speed of germination and final germination success was a function of salt concentration in the medium, the negative effect varying with species (Figures 1 and 2). Most affected by salt was *P. heydlauffii*, which showed a substantial reduction in germination even at rather high water potentials. At 50% seawater (equivalent to -1.25 MPa), when germination completely ceased in this species, the average reduction reached c. 50% in *P. suaveolens* and *P. schultzei* and -84% in *P. halophila* (Figure 2). Expressed in terms of the resulting water potentials, salt or mannitol had very similar effects on germination in *P. halophila*.

Growth and survival

Under control conditions, small *P. suaveolens* seedlings grew more than twice as fast as *P. halophila* seedlings, while RGRs of larger individuals were comparable in the two species (Figure 3). Increasing salt concentrations in the watering solutions lead to a linear decrease in RGR, irrespective of species and plant size. In spite of this

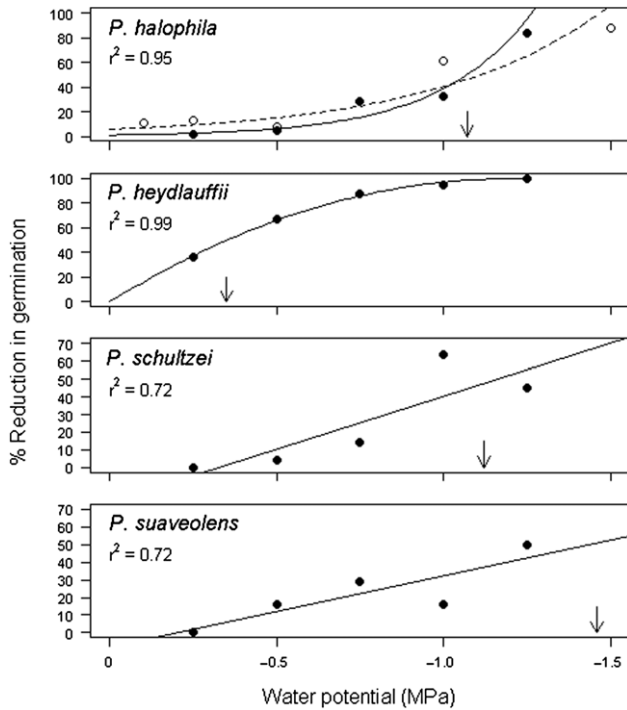


Figure 2. The impact of diluted seawater and mannitol solutions of comparable water potential on germination in *Pitcairnia halophila*, *P. heydlauffii*, *P. schultzei* and *P. suaveolens* expressed as the reduction of germinated seeds after 28 days compared to controls with distilled water. Data are means of 5 runs (seawater) and 3 runs (mannitol solutions) with 20 seeds each. The solid lines represent regression lines (seawater: power functions or linear regressions, $p < 0.05$) and the dashed line is a significant regression (mannitol). The explained variation (r^2) of the relationships of seawater concentration and germination is indicated in each graph. Note that the y-axes vary. Arrows indicate the water potential leading to a 50% reduction in germination (see also Table 1). Note that seawater has a water potential of c. -2.5 MPa.

qualitative resemblance, the two species differed considerably in their overall response to increasing salt concentrations. For one, the linear reduction in RGR was steepest in small *P. suaveolens* plants (-0.17 [% d^{-1}] [% seawater] $^{-1}$) and smallest in larger *P. halophila* plants (-0.05 [% d^{-1}] [% seawater] $^{-1}$). Moreover, even the *P. halophila* individuals subjected to the highest concentrations looked entirely healthy after 28 days of treatment, whereas all *P. suaveolens* plants at concentrations $\geq 30\%$ seawater developed brownish leaves. In the majority of cases, the number of necrotic leaves of plants receiving the highest salt concentrations (50 and 60%) exceeded that of healthy looking ones; a considerable number of individuals died, that is, the central part of the rosettes was rotten.

Foliar nutrient concentrations

Under control conditions, average tissue Na^+ concentrations [Na^+] ranged from 0.32 to 0.38 $mM g_{DW}^{-1}$ (Figure 4a), while K^+ concentrations [K^+] ranged from 0.9 to 1.2 $mM g_{DW}^{-1}$ (Figure 4b), resulting in quite similar Na/K ratios of 0.26–0.44 in both species. Irrigation with different concentrations of seawater invariably led to an increase in [Na^+], although the two species showed different trends: there was a tight linear relationship between % seawater and tissue [Na^+] in *P. halophila*, while in *P. suaveolens* increasing concentrations in the irrigation solution of $\leq 30\%$ led to a relatively steep increase in tissue [Na^+] without any further change at higher concentrations of seawater. The response of [K^+] was inconsistent in the four experiments with a slight decrease, an increase and

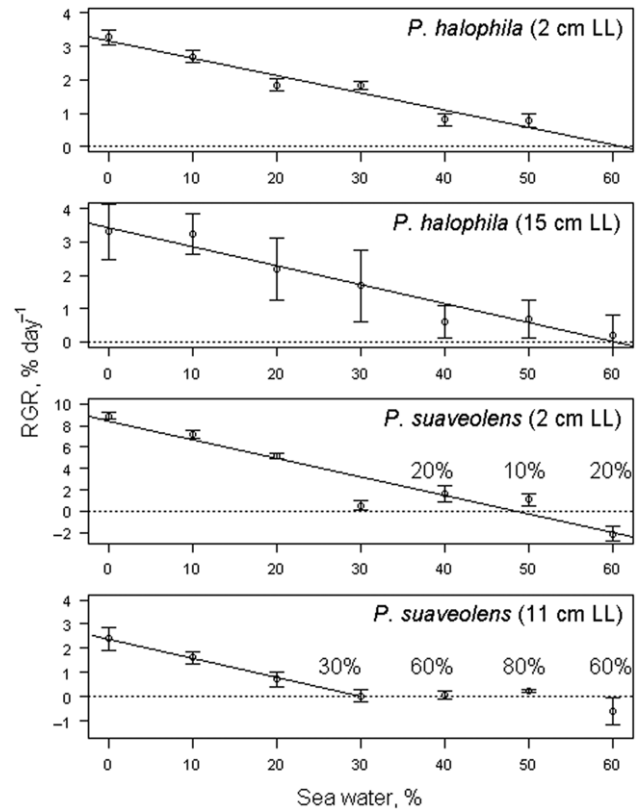


Figure 3. The relationship of relative growth rate and saltwater of varying concentrations in two *Pitcairnia* species. Data are means \pm SD ($n = 10$ for larger and 20 for smaller individuals). Solid lines are significant linear regressions ($p < 0.001$) with the following slopes: -0.05 , -0.05 , -0.17 and -0.08 . Because of the high mortality at high salt concentrations, only values for $\leq 30\%$ seawater were included in the regression analysis for larger *P. suaveolens*. Mortality rates are given as percentages.

twice no change (Figure 4b). In relative terms, the resulting changes in the Na/K ratio were strongest in 2-cm small *P. halophila* seedlings (Na/K ratio at 50% seawater = 2.03; + 500% compared to the control), in absolute terms the highest Na/K ratio was observed in *P. suaveolens* seedlings with $Na/K > 3.1$.

Seasonal changes in above-ground biomass and tissue nutrient concentrations *in situ*

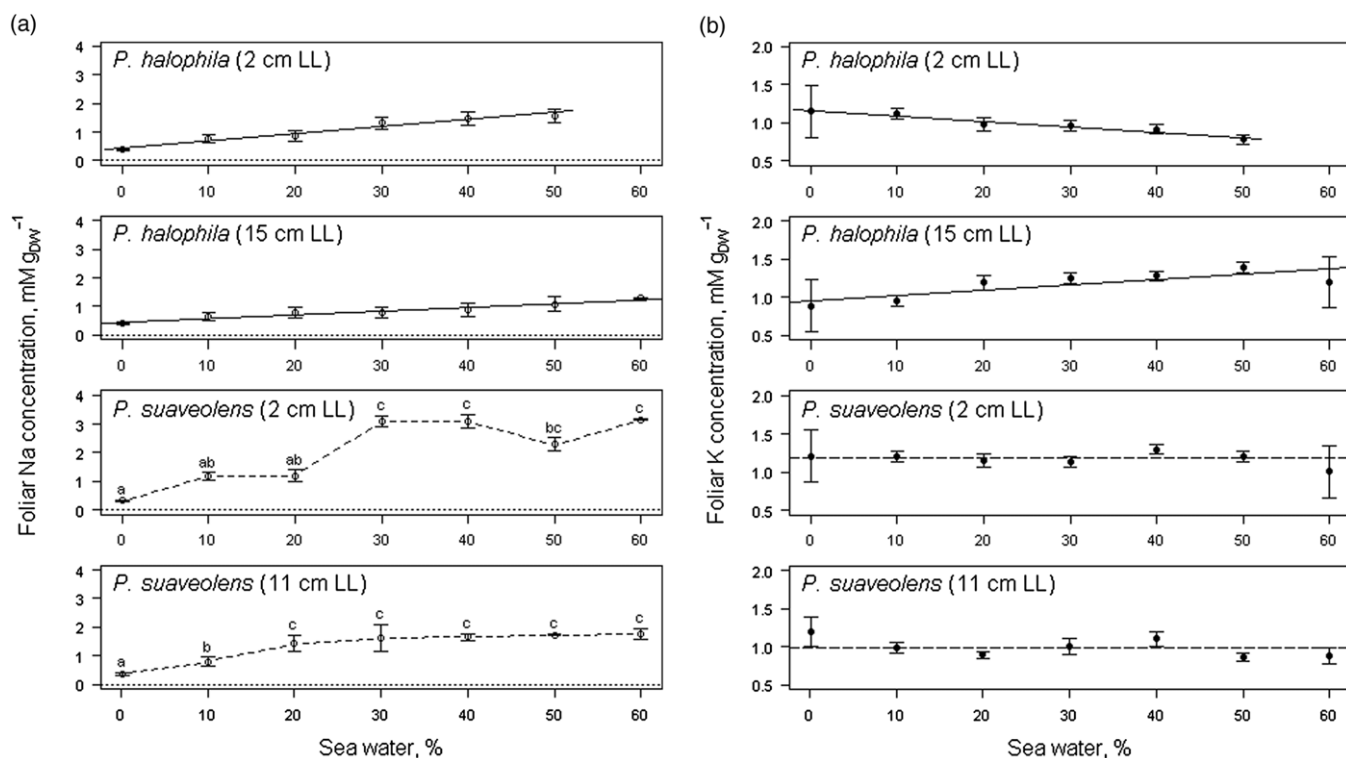
The natural growing site in Panama is characterised by a strong seasonality in precipitation and some variation in temperature (Figure 5). Plants bolted with the beginning of the rainy season and increased in biomass until the end of that season, while at the last collecting date in the early dry season in January 2020 seeds had already dispersed, and leaves and stalks had already started to desiccate. Tissue concentrations of sodium and potassium varied very little during the 9-month study period with average tissue [Na^+] of c. 0.15 $mM g_{DW}^{-1}$ in leaves and c. 0.30 $mM g_{DW}^{-1}$ in stems. The concentrations of K^+ averaged 0.5 $mM g_{DW}^{-1}$ in leaves and 0.3 $mM g_{DW}^{-1}$ in stems. The Na/K ratio showed no clear seasonal variation neither in stems nor leaves but was consistently higher in the former (1.0 ± 0.6 vs. 0.4 ± 0.2 , means \pm SD).

Discussion

There is no universally accepted definition of 'halophyte' (Flowers & Colmer 2008), and any attempt to categorise plants in a

Table 1. Effect of the water potential (Ψ) of test solutions on the germination success of bromeliad seeds. Given is the Ψ that led to a 50% reduction in germination compared to controls (distilled water). Values were obtained from the figures or tables in the cited publications.

Species	Ψ 50% (MPa)	Source
<i>Alcantarea imperialis</i>	-1.0	Pereira et al. (2009)
<i>Dyckia encholirioides</i>	-0.4–-1.4	Pompelli et al. (2006) ¹
<i>Neoglaziovia variegata</i>	-0.5	Silveira et al. (2011)
<i>Pitcairnia flammaea</i>	-1.1	Pereira et al. (2009)
<i>Pitcairnia halophila</i>	-1.1	This study
<i>Pitcairnia heydlauffii</i>	-0.4	This study
<i>Pitcairnia schultzei</i>	-1.1	This study
<i>Pitcairnia suaveolens</i>	-1.4	This study
<i>Tillandsia recurvata</i>	-0.5	Montes-Recinas et al. (2012)
<i>Vriesea gladioliflora</i>	-1.0	Correa and Zotz (2014)
<i>Vriesea heterostachys</i>	-0.8	Pereira et al. (2009)
<i>Vriesea penduliflora</i>	-0.8	Pereira et al. (2009)
<i>Vriesea sanguinolenta</i>	-0.8–-0.9	Zotz and Reuter (2009) ²
<i>Vriesea viridiflora</i>	-0.9	Correa and Zotz (2014)
<i>Vriesea vittata</i>	-0.5	Correa and Zotz (2014)
<i>Werauhia lutheri</i>	-0.7	Correa and Zotz (2014)

¹Range of four germination experiments at different temperatures.²Range of three different populations.**Figure 4.** The relationship of foliar Na (a) and K (b) concentrations and salt water of varying concentrations in the rooting medium of two species of *Pitcairnia*. Data are means \pm SD ($n = 10$ for *P. halophila* or fewer (2–9) in those *P. suaveolens* treatments with mortality, see Figure 3). Solid lines represent significant linear regressions ($p < 0.001$), different small letters indicate significant differences between treatments (ANOVA, *post hoc* Tukey's test) and dashed horizontal lines are grand means when groups did not differ.

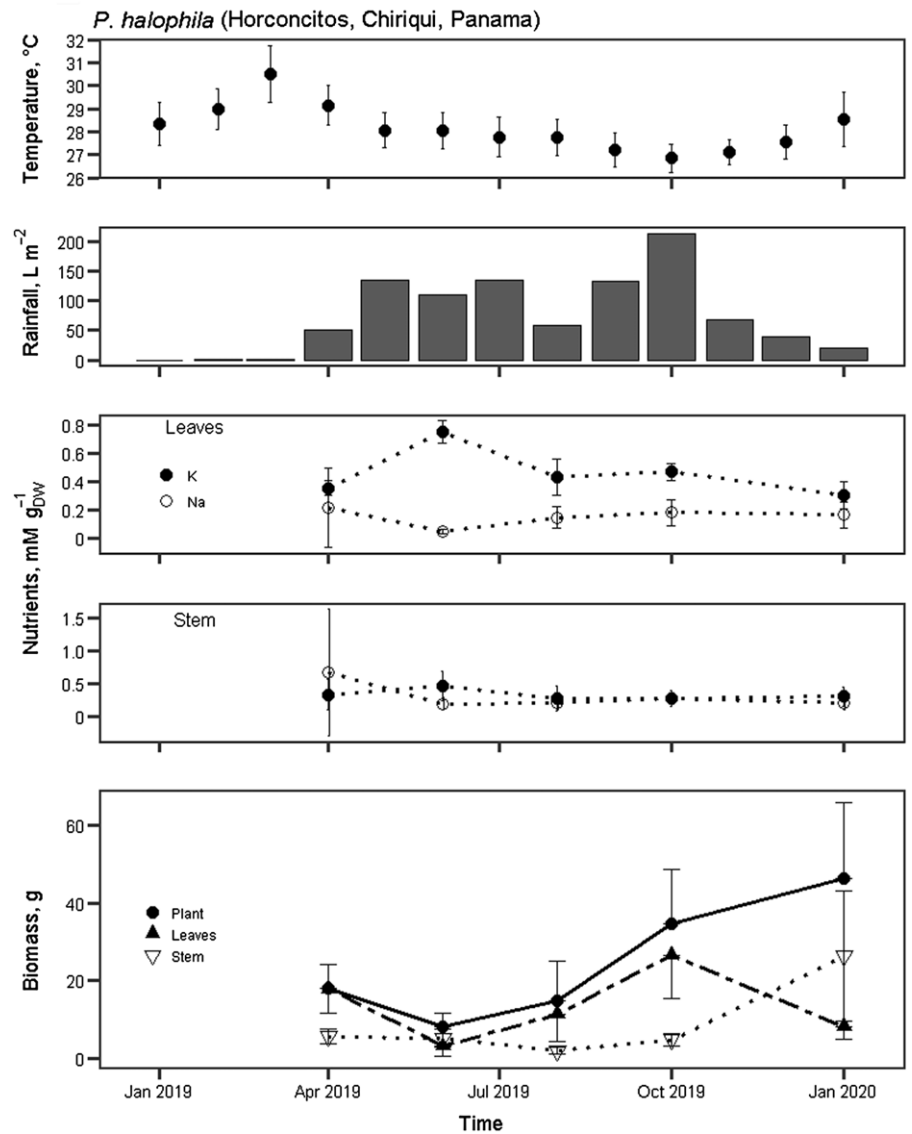


Figure 5. Seasonal changes in air temperature (°C, monthly averages \pm SD), in rainfall (monthly totals in mm), in leaf and stem concentrations of Na and K (means \pm SD, $n=3$), and in leaf, stem and total plant biomass (means \pm 1SD, $n=3$) at a natural growing site in Panama. The year 2019 was relatively dry with 945 mm, which amounts to c. 70% of the long-term average.

dichotomous way will result in legitimate criticism. Nevertheless, it is often inevitable to conceptualise potentially continuous biological variation as categories, compare, for example, Bennett *et al.* (2013) for halophytic versus glycophytic Poaceae, Smith and Winter (1996) for CAM versus C_3 plants or Zotz *et al.* (2021) for epiphytes versus terrestrially rooted plants. Thus, acknowledging this caveat and embracing the definition proposed by Flowers and Colmer (2008): ‘plants with the ability to complete the life cycle in a salt concentration of at least 200 mM NaCl (= 1/3 seawater) under conditions similar to those that might be encountered in the natural environment’, the results of this study provide some arguments that *P. halophila* may be categorised as halophyte, although we note that we have only studied early-life stages.

The last statement is seemingly at odds with the results of the germination experiment because germination of *P. halophila* was strongly affected by relatively low levels of salinity, and half-strength seawater almost completely inhibited germination (Figures 1 and 2). However, a majority of halophytes germinate better in the absence of salt, and a compilation of NaCl concentrations inhibiting germination in a range of beach, cliff and foredune species by Baskin and Baskin (2001) lists quantitatively

comparable inhibitory effects to those observed in *P. halophila*. Our comparison of germination success in salt water versus manitol solutions (Figure 2) indicates that this inhibition in the tested range of concentrations is not related to ionic toxicity but simply reflects an osmotic effect (Lambers *et al.* 2008). The severity of the observed inhibition can also be compared with the available data for 14 species of bromeliads (Table 1). The water potential leading to 50% reduction in germination success ($\Psi_{-50\%}$) in *P. halophila* is comparable to $\Psi_{-50\%}$ of three congeners (*P. schultzei* and *P. suaveolens*, this study, and *P. flammaea*, Pereira *et al.* 2009) and $\Psi_{-50\%}$ of some species in other genera (*Vriesea* and *Alcantarea*). In most tested bromeliads, germination is more severely affected by low water potentials.

While *P. halophila* was not particularly tolerant to lower water potentials during germination, plant growth was much less affected by low Ψ in this species than in the studied congener (Figure 3). At the cut-off defined by Flowers and Colmer (2008), the RGRs still reached about 30% of the maximum value. Moreover and in contrast to *P. suaveolens* plantlets, which showed visual signs of damage and quite high foliar Na⁺ concentrations (Figure 4), *P. halophila* looked invariably healthy during the entire course

of the experiment and was capable of maintaining relatively low Na^+ concentrations in their leaves, particular in larger plants. Notably, growth was low in *P. halophila* even under control conditions (Figure 3). Such a low growth potential could make this species a poor competitor, which is possibly one reason why it is restricted to rock outcrops and is not found in adjacent vegetation.

The seeds of *P. halophila* are released with the beginning of the dry season (pers. obs.). Under natural conditions, germination will probably not occur until the onset of the rainy season, which should assure relatively low salinity in soil during germination and initial establishment. Thus, the fact that germination of this species was rather susceptible to lower water potentials may not be very relevant ecologically.

The field observations (Figure 5) also suggest that the demonstrated ability of *P. halophila* to grow under somewhat saline conditions may actually not be tested *in situ* even at a growing site in immediate vicinity to the sea. The tissue concentrations of sodium in both leaves and stems were consistently low and comparable to those of the controls in the greenhouse experiment. Given that plants rest during the dry season, the frequent and substantial precipitation during the rainy season when plants actively grow seems to dilute any input from salt spray to a degree that the plants do apparently not have to deal with elevated soil salinity *in situ*.

In conclusion, we present the results of an experimental/observational study that explored the physiological and ecological basis of the occurrence of *P. halophila* in the spray zone of cliffs in Central America. Our study gives a mixed picture: while the species is quite susceptible to moderate salinity during germination, growth of plantlets was still possible when watering solutions reached half-strength seawater, which is typically taken as a characteristic of halophytes. However, *in situ* observations suggest that the conditions at the natural growing sites with abundant precipitation during the growing season are not saline, and thus, ecologically, *P. halophila* does not face the challenges of a halophyte.

Supplementary material. To view supplementary material for this article, please visit <https://doi.org/10.1017/S0266467422000116>

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Competing interests. The author(s) declare none.

References

- Allen SE (ed.) (2008) *Chemical analysis of ecological materials*. Oxford: Blackwell Scientific, 565 p.
- Alves RJV, Kolbek J and Becker J (2008) Vascular epiphyte vegetation in rocky savannas of southeastern Brazil. *Nordic Journal of Botany* **26**, 101–117.
- Baskin CC and Baskin JM (2001) *Seeds: Ecology, biogeography, and evolution of dormancy and germination*. San Diego: Academic Press, 666 p.
- Bennett TH, Flowers TJ and Bromham L (2013) Repeated evolution of salt-tolerance in grasses. *Biology Letters* **9**, 20130029.
- Benzing DH (2000) *Bromeliaceae - Profile of an adaptive radiation*. Cambridge: Cambridge University Press, 690 p.
- Cáceres GDA, Schulte K, Schmidt M and Zizka G (2011) A synopsis of the *Bromeliaceae* of Panama, including new records for the country. *Willdenowia* **41**, 357–369.
- Cogliatti-Carvalho L, Nunes-Freitas AF, Rocha CFDd and Van Sluys M (2001) Variation in structure and composition of Bromeliaceae at five zones of “restinga” in Jurubatiba National Park, Macaé, RJ. *Revista Brasileira de Botânica* **24**, 1–9.
- Correa S and Zotz G (2014) The influence of collecting date, temperature and moisture regimes on the germination of epiphytic bromeliads. *Seed Science Research* **24**, 353–363.
- Díaz MDS and Alvarez CC (2009) Plant regeneration through direct shoot formation from leaf cultures and from protocorm-like bodies derived from callus of *Encyclia mariae* (Orchidaceae), a threatened Mexican orchid. *In vitro Cellular & Developmental Biology-Plant* **45**, 162–170.
- Díaz-Jiménez P (2007) Flora epífita de los manglares de Tabasco, México. *Kuxulkab - Universidad Juárez Autónoma de Tabasco* **8**, 11–17.
- Egan TP and Ungar IA (1999) The effects of temperature and seasonal change on the germination of two salt marsh species, *Atriplex prostrata* and *Salicornia europaea*, along a salinity gradient. *International Journal of Plant Sciences* **160**, 861–867.
- Flowers TJ and Colmer TD (2008) Salinity tolerance in halophytes. *New Phytologist* **179**, 945–963.
- Gómez MA and Winkler S (1991) Bromelias en manglares del Pacífico de Guatemala. *Revista de Biología Tropical* **39**, 207–214.
- Grant JR (1994) Notes on the coastal Costa Rican endemic *Pitcairnia halophila*. *Journal of the Bromeliad Society* **44**, 170–172.
- Griffiths H, Smith JAC, Lüttge U, Popp M, Cram WJ, Diaz MA, Lee HSL, Medina E, Schäfer C and Stimmel KH (1989) Ecophysiology of xerophytic and halophytic vegetation of a coastal alluvial plain in northern Venezuela. IV. *Tillandsia flexuosa* Sw. and *Schomburgkia humboldtiana* Reichb., epiphytic CAM plants. *New Phytologist* **111**, 273–282.
- Hunt R (1982) *Plant growth curves - the functional approach to plant growth analysis*. London: Edward Arnold, 248 p.
- Lambers H, Chapin III FS and Pons TL (2008) *Plant physiological ecology* (2nd Ed edition). New York: Springer, 540 p.
- Larcher W (2003) *Physiological plant ecology*. New York: Springer, 513 p.
- Lüttge U, Klauke B, Griffiths H, Smith JAC and Stimmel KH (1986) Comparative ecophysiology of CAM and C3 bromeliads. V. Gas exchange and leaf structure of the C3 bromeliad *Pitcairnia integrifolia*. *Plant, Cell and Environment* **9**, 411–419.
- Medina E, Cram WJ, Lee HS, Lüttge U, Popp M, Smith JAC and Diaz MA (1989) Ecophysiology of xerophytic and halophytic vegetation of a coastal alluvial plain in northern Venezuela. I. Site description and plant communities. *New Phytologist* **111**, 233–243.
- Miller GA and Silander JA (1991) Control of the distribution of giant rosette species of *Puya* (Bromeliaceae) in the Ecuadorian Paramos. *Biotropica* **23**, 124–133.
- Montes-Recinas S, Márquez-Guzmán J and Orozco-Segovia A (2012) Temperature and water requirements for germination and effects of discontinuous hydration on germinated seed survival in *Tillandsia recurvata* L. *Plant Ecology* **213**, 1069–1079.
- Pereira AR, Andrade ACS, Pereira TS, Forzza RC and Rodrigues AS (2009) Comportamento germinativo de espécies epífitas e rupícolas de Bromeliaceae do Parque Estadual do Ibitipoca, Minas Gerais, Brasil. *Revista Brasileira de Botânica* **32**, 827–838.
- Pickens KA, Affolter JM, Wetzstein HY and Wolf JHD (2003) Enhanced seed germination and seedling growth of *Tillandsia eizii* in vitro. *Hortscience* **38**, 101–104.
- Pompelli MF, Fernandes D and Guerra MP (2006) Germination of *Dyckia encholirioides* (Gaudichaud) Mez var. *encholirioides* under saline conditions. *Seed Science and Technology* **34**, 759–763.
- R Development Core Team (2019) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-900007-900050, URL <http://www.R-project.org>
- Rasband WS (1997–2015) *ImageJ*. Bethesda, Maryland, USA: U. S. National Institutes of Health.

- Saraiva DP, Mantovani A and Forzza RC** (2015) Insights into the evolution of *Pitcairnia* (Pitcairnioideae-Bromeliaceae), based on morphological evidence. *Systematic Botany* **40**, 726–736.
- Silveira DG, Pelacani CR, Cardoso Antunes CG, Rosa SS, Duarte Souza FV and Ferreira de Santana JR** (2011) Germinative response of *Neoglaziovia variegata* (Arruda) Mez seeds. *Ciencia E Agrotecnologia* **35**, 948–955.
- Smith JAC and Winter K** (1996) *Taxonomic distribution of crassulacean acid metabolism*. In Winter, K and Smith, JAC (eds.), *Crassulacean acid metabolism. Biochemistry, ecophysiology and evolution*. Berlin: Springer, pp. 427–436.
- Smith LB** (1964) Notes on Bromeliaceae, XXI. *Pitcairnia*. *Phytologia* **10**, 1–55.
- Swagel EN, Bernhard A Van H and Ellmore GS** (1997) Substrate water potential constraints on germination of the strangler fig *Ficus aurea* (Moraceae). *American Journal of Botany* **84**, 716–722.
- Tomlinson PB** (1986) *The botany of mangroves*. Cambridge: Cambridge University Press, 419 p.
- Ungar IA** (1978) Halophyte seed germination. *Botanical Review* **44**, 233–264.
- Vasquez R and Ibisch PL** (2000) Drei neue Bromelienarten aus Bolivien: *Billbergia mohammadii*, *Pitcairnia heydlauffii* und *Puya cochabambensis*. *Die Bromelie* **1**, 11–19.
- WCVP** (2020) World Checklist of Vascular Plants, version 2.0. Facilitated by the Royal Botanic Gardens, Kew. Published on the Internet; <http://wcvp.science.kew.org/>. Retrieved 17 July 2020.
- Zotz G** (2005) Differences in vital demographic rates in three populations of the epiphytic bromeliad, *Werauhia sanguinolenta*. *Acta Oecologica* **28**, 306–312.
- Zotz G and Reuter N** (2009) The effect of exposure to sea water on germination and vegetative growth of an epiphytic bromeliad. *Journal of Tropical Ecology* **25**, 311–319.
- Zotz G and Schultz S** (2008) The vascular epiphytes of a lowland forest in Panama - species composition and spatial structure. *Plant Ecology* **195**, 131–141.
- Zotz G, Hietz P and Schmidt G** (2001) Small plants, large plants - the importance of plant size for the physiological ecology of vascular epiphytes. *Journal of Experimental Botany* **52**, 2051–2056.
- Zotz G, Weigelt P, Kessler M, Kreft H and Taylor A** (2021) EpiList 1.0 - A global checklist of vascular epiphytes. *Ecology* **102**, e03326.