

NUTRIENT APPLICATION TO ROOTS PROMOTES PRODUCTION OF NON-CARNIVOROUS LEAVES IN *CEPHALOTUS FOLLICULARIS* LABILL. (CEPHALOTACEAE)

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Abstract: Carnivorous plants such as *Sarracenia*, *Drosera*, and *Utricularia* are known to reduce investment in carnivorous traits when nutrients are supplied through their roots and shoots. We hypothesized that *Cephalotus follicularis* would respond similarly by reducing pitcher production when nutrients are available in the growth medium. To test this hypothesis, plants were grown for six months with varying amounts of Osmocote (14-14-14). We measured (1) number of pitchers, (2) number of leaves, and (3) crown size before and after treatment, and compared the differences among treatments. Contrary to our expectations, Osmocote application did not significantly affect pitcher production or crown size. However, higher nutrient treatments significantly increased non-carnivorous leaf production. These results suggest that *C. follicularis*, like other carnivorous plant genera, may shift resource allocation away from carnivory toward nutrient acquisition via roots when nutrients are readily available.

Introduction

Although carnivorous plants capture prey to obtain nutrients, they are also capable of acquiring nutrients through roots and shoots, similar to non-carnivorous plants. The balance between carnivory and nutrient uptake is thought to depend on the costs of producing and maintaining specialized trapping structures relative to the benefits of prey-derived nutrients in nutrient-poor environments (Givnish *et al.* 2018).

Because of this trade-off, carnivorous plants are highly sensitive to nutrient availability and may reduce investment in carnivorous traits when these nutrients are abundant (Fleischmann *et al.* 2018). For example, nutrient addition in terrestrial species has been shown to shift resource allocation away from carnivory: *Sarracenia purpurea* produces more photosynthetic leaves relative to pitchers (Ellison & Gotelli 2002), and *Drosera rotundifolia* reduces mucilage production (Thorén *et al.* 2003).

Similar responses have been documented in *Utricularia*, where nutrient enrichment affects trap number, biomass allocation, and morphology (Adamec 2018; Jobson *et al.* 2000; Kibriya & Jones 2007). Additional studies have linked environmental nutrient conditions to trap traits and prey capture efficiency (Guisande *et al.* 2000, 2004; Adamec 2008). Stable isotope analyses further demonstrate a shift from carnivory to shoot-based nutrient uptake under nutrient-rich conditions (Kurosawa *et al.* 2024).

Cephalotus follicularis, the Albany pitcher plant, is endemic to southwestern Australia and represents a distinct lineage of carnivorous plants. Despite extensive research on other genera, studies examining nutrient-driven shifts in resource allocation in *C. follicularis* remain limited.

To address this gap, we hypothesized that *C. follicularis* would reduce investment in carnivory and shift toward root-based nutrient acquisition when nutrients are supplied to the substrate. To test this, we applied zero (control), one, four, or eight granules of Osmocote (14-14-14) per pot and monitored plant responses over six months. We quantified changes in pitcher number, leaf number, and crown size before and after treatment.

Materials and methods

Plant material and experiment design

The experiment was conducted from 14 May to 14 November 2020. Eight-month-old leaf pullings of *Cephalotus follicularis* were cultivated under LED lighting with a 12-hour photoperiod at 25°C for six months. Plants were grown in 6.35 cm square pots containing a 1:1 mixture of peat moss and sand. The growth medium was heat-sterilized prior to use, and plants were maintained within an insect-free enclosure. Plants were randomly assigned to one of four treatments: 0 (control), 1, 4, or 8 Osmocote granules per pot (n = 8 per treatment). Granules were placed at the corners of each pot to minimize direct contact with plants. Each treatment group was placed on a plastic tray containing 1 cm of reverse osmosis water, which was replenished as needed when the tray dried out.

Measurements

The number of pitchers, number of non-carnivorous leaves, and crown size were measured before and after treatment by three junior high school students (Fig. 1). Crown size was defined as the maximum distance between leaves.



Figure 1. Junior high school students measuring the growth of *Cephalotus follicularis*.

Statistical analysis

Statistical comparisons were performed using Wilcoxon rank-sum tests with Bonferroni correction. All data analyses were performed using R v.4.0.3 and R Studio v.1.3.1093 (R Core Team 2014). Data were plotted using the 'ggpubr' function in the R package 'ggplot2' (Kassambara 2020). Results were reported as mean $\pm$ 1 s.d. and significance level was defined as $\alpha < 0.0167$ after Bonferroni correction.

Results

Pitcher production

No significant differences in pitcher production were observed between the control and any nutrient treatment (Fig. 2a).

Non-carnivorous leaf production

Non-carnivorous leaf production increased with higher nutrient levels. Plants receiving four and eight Osmocote granules produced significantly more leaves compared to the control (Fig. 2b).

Crown growth

Crown size did not differ significantly between the control and treatment groups (Fig. 2c).

Discussion

Our results partially support the hypothesis that *Cephalotus follicularis* alters resource allocation in response to nutrient availability. While nutrient addition did not significantly affect pitcher production or crown growth, it promoted the production of non-carnivorous leaves at higher doses.

Our results are consistent with observations in other carnivorous plant genera, where increased nutrient availability reduces investment in carnivorous structures and promotes allocation to roots and shoots (Ellison & Gotelli 2002; Thorén *et al.* 2003; Givnish *et al.* 2018; Kurosawa *et al.* 2024). However, *C. follicularis* did not exhibit a statistically significant reduction in pitcher number. This may suggest that pitcher production is less plastic in this species, or that longer experimental durations or higher nutrient inputs (in dosage or frequency) are required to detect such effects. The observed increase in leaf production indicates a shift in resource allocation toward photosynthetic structures when nutrients are readily available in the substrate. This finding supports the broader hypothesis that carnivorous plants optimize the costs and benefits of carnivory in response to environmental nutrient availability (Givnish *et al.* 2018). Although plants were maintained under insect-free conditions, the possibility of nutrient acquisition from soil-borne invertebrates cannot be excluded.

Future studies should explore longer-term responses, changes in biomass, physiological measurements (e.g., nutrient uptake rates; leaf N contents), and potential trade-offs between leaf and pitcher function to better understand resource allocation strategies in *C. follicularis*.

Additionally, this study demonstrates the feasibility of involving junior high school students in meaningful scientific research. Their participation in data collection highlights the potential for educational outreach to contribute to ecological studies.

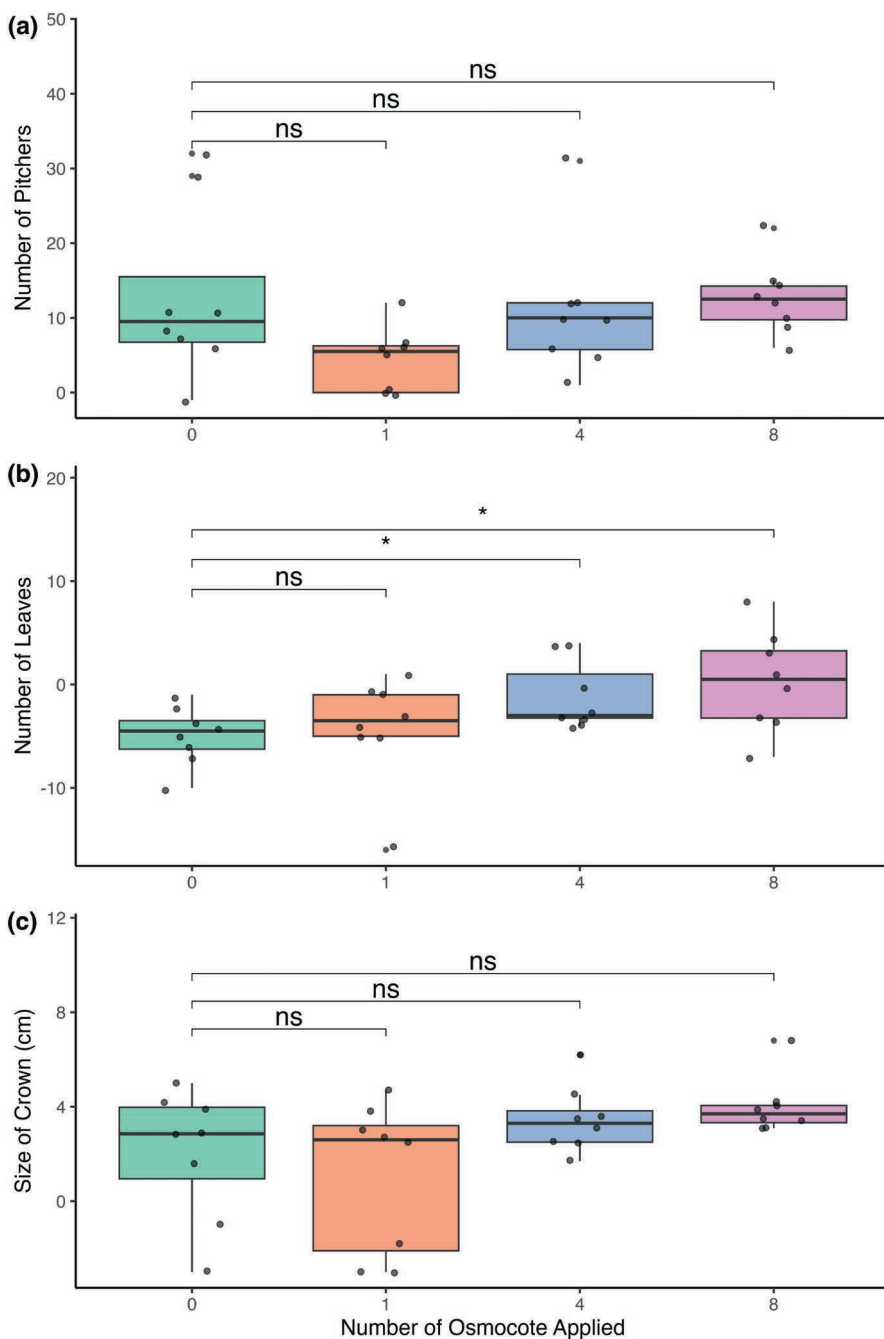


Figure 2. Boxplots showing (a) the number of pitchers, (b) the number of non-carnivorous leaves, and (c) the crown sizes across treatment groups. Points represent individual observations. Statistical comparisons were performed using Wilcoxon rank-sum tests with Bonferroni correction. *, $p < 0.0167$; ns, not significant.

Note: The raw data are in an online supplement available via <https://doi.org/10.55360/cpn553.ek593> redirect and directly on the CPN website.

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