FLOWER VISITORS HAVE A TASTE FOR SALT, BUT THIS MAY HAVE LITTLE RELEVANCE TO NECTAR EVOLUTION: A COMMENT ON FINKELSTEIN *ET AL.* 2022

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Text—A recent study by Finkelstein et al (2022) has demonstrated that a variety of flower-visiting animals have a taste for salt, such that plants with sodium enriched nectar received more visits and were visited by more animal species compared with control plants. They further suggest that plants could thus attract pollinators through relatively high levels of sodium in their nectar and that this could drive evolution of nectar sodium concentration.
However, as argued below, we reject this latter suggestion, especially because their experimental manipulations departed significantly from natural circumstances and were irrelevant to nectar evolution.

Firstly, the experimental manipulations carried out by Finkelstein et al (2022) involved realistic sugar concentration, but nectar volumes, sugar weight and sodium concentrations were way above natural levels, and hence unrealistic. Finkelstein et al (2022) added artificial nectar to flowers with a concentration of 35% wt/vol, a level that lies within the observed range of reported nectar sugar concentrations for the five plant species included in their study (i.e., 27 to 61% wt/vol; Table 1), and is therefore realistic. However, they added 15 µl of this artificial nectar to each open flower of each plant three times per day, amounting to an additional 45 µl of artificial nectar and 15.75 x 10³ µg of sugar per flower per day, which are levels way in excess of observed standing crop volumes (i.e., 1-7 µl; Table 1), rates of nectar production (i.e., <0.7 µl/h; Table 1), and rate of sugar production (i.e., <2 µg/day), per flower for the five study species. We have been unable to find reports regarding sodium concentrations in nectar from the five plant species, but this concentration was found to be only 0.017% for the congeneric *Penstemon barbeyi* (i.e., 7.5 mmol/L; Hiebert & Calder 1983) and the sodium concentration for 19 plant species that are mostly visited by hummingbirds averaged only 0.008% (i.e., 3.4 mmol/L; Hiebert & Calder 1983). Thus the 1% wt/vol sodium concentration in added nectar (Finkelstein et al. 2022) was also unrealistically high.

Secondly, the observed numerical responses of flower-visitors to experimental nectar addition probably reflect simple efficient foraging rather than visitor attraction. Because nectar addition was carried out just 30min before observations of flower visitors, there would have been insufficient time for these visitors to be 'attracted' to sodium-enriched plants by remembering them and returning to them (Pyke 2016a; Pyke 2016b). As the locations of sodium-enriched and control plants were randomised between days but were the same for three experimental sessions on each day, it is possible that some flower visitors may have foraged during two or more of these sessions on the same day, remembered the locations of the sodium-enriched plants between sessions, preferentially visited them, and thus been attracted to them, but such occurrences are likely to be rare. It is also unlikely that flower visitors can detect sodium levels in floral nectar at a distance, and before visiting and probing a flower, and so this kind of attraction is also unlikely. Instead,

Table 1: Nectar attributes for five plant species included in study by Finkelstein et al (2022). They added 15µL of artificial nectar with concentration 35% wt/vol (i.e., 0.35 mg/µL) to individual flowers three times per day. This amounts to an additional 45 µL of artificial nectar per flower per day, containing 15.75 x 10³ µg sugar. Concentration of added nectar is within observed range for study species, but added nectar volume and nectar sugar greatly exceed observed levels.

Species (Family)	Nectar standing crop per flower (µL)	Nectar volume production per flower	Sugar concentration (% wt/vol)	Sugar production rate	Reference
Monarda didyma (Lamiaceae)	7	2.3 µL/ 36h	27 to 32%		(Whitten 1981)
Penstemon digitalis (Plantaginaceae)		0.7 µL/h	33%		(Burdon et al. 2020)
Geranium sanguineum (Geraniaceae)			46%		(Masierowska 2006)
Geranium sanguineum (Geraniaceae)	0	0			(Philipp & Hansen 2000)
Echinacea purpurea (Asteraceae)		Net 0.07 to 0.19 µL between floral stages depending on stage	33 to 61% depending on floral stage		(Wist & Davis 2006)
Achillea millefolium (Asteraceae)				<2 µg/ flower/ day (i.e., 31 µg/day per capitulum; 17-30 flowers)	(Hicks et al. 2016; Sulborska & Weryszko-Chmielewska 2006)

because nectar attributes for flowers from the same plant tend to be correlated, a flower-visitor that is foraging efficiently, perhaps even optimally, should tend to move from one flower on a plant to another on the same plant if it found the nectar in the first flower to be particularly rewarding (Pyke et al. 2020a). So, if flower-visitors find sodium-enriched nectar to be relatively rewarding, they would tend to visit more flowers on plants with sodium-enriched nectar than on control plants (Pyke et al. 2020a). This would lead, in turn, to greater numbers and higher diversity of flower-visitors observed on sodium-enriched plants compared with control plants, which is exactly what was found (Finkelstein et al. 2022).

Thirdly, for experimental nectar additions to provide information relevant to nectar evolution, they must be carried out at a scale relevant to natural selection, and very different from the scale adopted in Finkelstein et al (2022), and must address various components of biological fitness. To be relevant to nectar evolution, experimental manipulations should imitate what happens to mutant plants producing nectar that differs

slightly from the average attributes (Pyke 2016b). In such experiments, again because nectar attributes of flowers from the same plant, mutant and otherwise, will likely be correlated, a flower-visiting animal that visits a flower on a mutant plant, and finds encountered nectar to be relatively favourable (e.g., slightly higher than average nectar volume, concentration or sodium content), should be more likely to visit another flower on the same plant, than if nectar in the first flower was less favourable (Pyke 2016b). In this way, mutant plants with flowers considered relatively favourable by flower-visitors should have more flowers probed per plant visit than plants with less favourable nectar (Pyke 2016b). In turn, this would result in the plants with favourable nectar receiving and transmitting more pollen, and thus having higher potential reproductive fitness than plants with less favourable nectar (Pyke 2016b). At the same time, there could also be trade-offs within plants between resources used to produce favourable nectar and resources required for other activities (Pyke 2016b; Pyke et al. 2020b). Under these circumstances, average nectar attributes should evolve towards intermediate levels such

that net biological fitness is maximised (Pyke 2016b).

In summary, the experiments carried out by Finkelstein et al (2022), and their results, are irrelevant to possible evolution of sodium levels in floral nectar, through attraction of pollinators, because these experiments involved unnaturally high nectar volumes and sodium concentrations, the results are probably consequences of flower-visitors foraging efficiently at individual plants and not because they are attracted to particular plants, and the experiments did not consider the responses of flower-visitors to nectar attributes differing slightly from the average.

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