





## Investigating pollinators diet: a comparison of sampling techniques for pollen and nectar

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## Abstract:

Pollinator insects are declining worldwide and recent research is highlighting how their diet is a fundamental piece in the definition of their health status. Pollinators rely mainly on floral resources (i.e., pollen and nectar) for their sustainment and the study of their nutritional ecology thus represents a key step in the definition of conservation strategies. Despite its key role, nutrition has always been overlooked by the research community and a reason for this can be the hindrance represented by the sampling of pollen and nectar that are usually present in small amounts making it hard to collect enough quantity for the chemical analyses. To cope with this issue researchers have developed a large series of sampling methodologies that aim to simplify the collection process, but this has led to a lack of uniformity in the techniques employed in different studies thus making it difficult to compare the results produced. Here we compared some of the more commonly used sampling techniques for pollen and nectar by looking at the differences that they introduce in the nutritional profile of floral rewards. Along with that we also proposed a novel methodology for pollen sampling based on the use of a portable vacuum. Pollen was sampled from three different species (i.e Alstroemeria aurea, Hippeastrum vittatum, Tropaeolum majus) while nectar was collected from three other species (Agapanthus praecox, Russelia equisetiformis, Salvia greggii) cultivated at the institute CREA of San Remo, Italy. We evaluated the sugar and secondary metabolites composition in the nectar while for pollen we also looked at its protein and lipid composition using UV-vis assays and HRMS analyses. Our results highlighted how different techniques can introduce biases in the nutritional profiling of floral rewards with alteration either in the quantities detected or in the overall composition or both, furthermore, the data obtained supported the use of a novel technique that we propose both for its high recovery capacity and accuracy that was found not significantly different from the control techniques represented by the use of the mesh. This study stresses the necessity of adopting uniform sampling procedures in different studies to compare the different outcomes given the growing importance of the field of pollinators' nutritional ecology for their conservation.