



Introduction

This Quick Guide is a starting point for new experiments:

- New species (e.g. measuring tomato pollen for the first time)
- New type of sample material (e.g. measuring stored samples instead of fresh samples)
- New type of analysis (e.g. quantifying the number of pollen grains instead of viability testing)

Getting Started

- 1 Project Goal**
 - Define the project goal. Key aspects are:
 - Which species?
 - Type of sample material (fresh pure pollen, fresh pollen from anthers, dehydrated stored pollen, pollen on stigma after manual pollination, pollen from pollen traps...)?
 - Type of analysis (viability, counting, ploidy)?
 - Statistical sample size and number of repetitions?
TIP: The number of samples and biological replicates depends on the variability of the material. It is worthwhile quantifying this variability to define the sampling and replicate strategy.

Note: Pollen viability on a single plant can vary significantly depending on where on the plant the sample is taken, the developmental stage of the flower, the time of the day and environmental influences. The following factors affecting pollen viability should be considered when collecting pollen:

 - Time of the day
 - Variability within a plant: Compare pollen from flowers of a similar position on the plant and equal developmental stage.
 - Environmental and other abiotic factors: Application of pesticides, humidity, temperature and light intensity
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- 2 Experimental Setup**
 - Check the **Pollen Analysis Instructions** on www.amphasys.com/downloads and search for the species you are interested in. Names are mentioned in Latin and English.
 - For each species, the corresponding measurement buffer (AF buffer), filter and chip type is mentioned. Make sure that you have those materials available. We strongly recommend using the setup mentioned in the list.
TIP: If you know that your pollen is considerably larger or smaller than the reference values stated in the Pollen Analysis Instructions, get in touch with Amphasys support to check whether the setup must be adapted.
 - In case the species is not in the list, find out more about the approximate cell size and the plant family. With this information get in touch with Amphasys support to request the recommended experimental setup.
TIP: You can determine the cell size by microscopy using a size reference, or by literature research.
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- 3 Sampling Strategy**
 - The sampling strategy depends on the goal of the experiment. Don't hesitate to contact Amphasys support for ideas.
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- 4 Sample Preparation**
 - Have a look at the [Sample Preparation Quick Guide](#) to identify a suitable methodology.
 - In addition, Amphasys support can provide more detailed and species-specific tips and tricks.
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- 5 Measurement Template**
 - Amphasys provides validated measurement templates for most applications on the Amphasys website. Don't hesitate to download a template or contact us to get a customized one.
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- 6 Test Run**
 - Prepare samples and perform a series of measurements to validate the selected sampling, sample preparation and instrumental settings.
 - The Amphasys [Quick Guides](#) will help you setting up and performing those experiments.
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- 7 Validation**
 - Don't hesitate to contact us for assistance and guidance throughout this setup process or for reviewing the final setup.
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