## **APPLICATION NOTE**

# **CORN** POLLEN VIABILITY AND DEVELOPMENTAL STAGES

Pollen quality plays a crucial role in successful corn breeding and production. For this purpose, Amphasys developed a process for the rapid determination of corn pollen viability and the assessment of the pollen developmental stage using a technology called impedance flow cytometry (IFC). In addition, IFC allows direct quantification of pollen samples in isotonic media. Use your measurement results to improve your processes!

#### Straightforward 3-Step Workflow



#### AmphaZ32 Impedance Flow Cytometer

- Rapid
- Accurate
- Reproducible
- Label-free
- Portable for on-site analysis

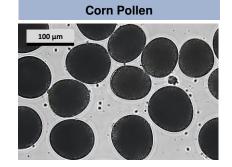


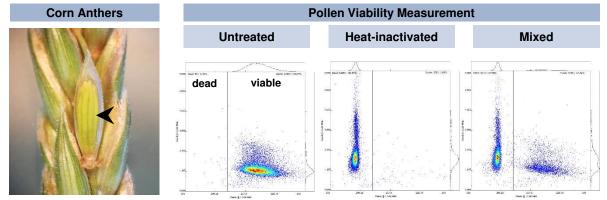
#### Lab-on-a-chip technology

- Small samples required
- Single cell analysis
- Statistically large sample sizes
- Sensitivity and throughput tunable by chip choice

## CORN POLLEN VIABILITY

Pollen viability is a key factor for successful pollination and influenced by many factors, such as temperature, humidity and use of pesticides. Impedance flow cytometry technology allows rapid and accurate corn pollen viability testing with minimal sample preparation. Pollen can be mechanically extracted from anthers by squashing, or shed pollen can be suspended directly in measurement buffer.





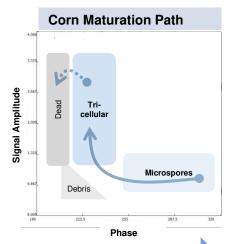


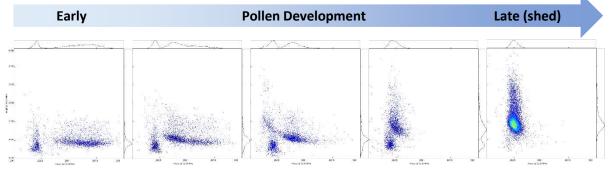


## DEVELOPMENTAL STAGES

The electric characterization of corn pollen allows a visualization of the developmental stage (*Figure series below*). For the first 4 stages, 3 anthers were manually removed from the tassel, squashed in measurement buffer and filtered. For the last stage, shed pollen was collected and directly suspended in measurement buffer. All samples were well mixed and measured using the same settings.

A pollen maturation path could be extracted from the development series. The path shows an initial phase shift to lower phases, followed by an increase in signal amplitude (*figure on the right*). Dead pollen shows an additional phase shift to lower phases and debris is always visible at low signal amplitudes.

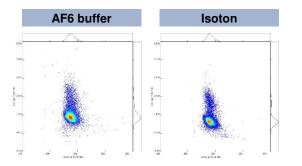




## QUANTIFICATION IN ISOTON

Pollen shed can be quantified by placing pollen collecting devices in the field and subsequently counting the particles that have been trapped. These devices may be filled with an isotonic liquid to preserve the pollen. The Ampha flow cytometers are able to directly quantify pollen that is suspended in isotonic media. Therefore, pollen shed can be monitored easily. Depending on the collection method, concentrating and filtering of the pollen may be necessary.

Corn pollen was suspended in AF6 measurement buffer and Isoton. By adjusting the instrument settings, similar patterns can be obtained. This indicates that pollen in Isoton can be quantified just like with the Amphasys measurement buffers.



#### **Sample Preparation**

- Collection of 3 anthers
- Resuspension and squashing in 1
  ml AF6 measurement buffer
- Filtering using 150 µm filter
- Dilution with 2 ml AF6 buffer, mix well (if necessary even during measurement)

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