

1 **Supplementary file 1.**

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3 **Multiple strategies for heat adaptation in rice endosperms**
4 **revealed by on-site cell-specific analysis**

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22 Supplementary file 1.

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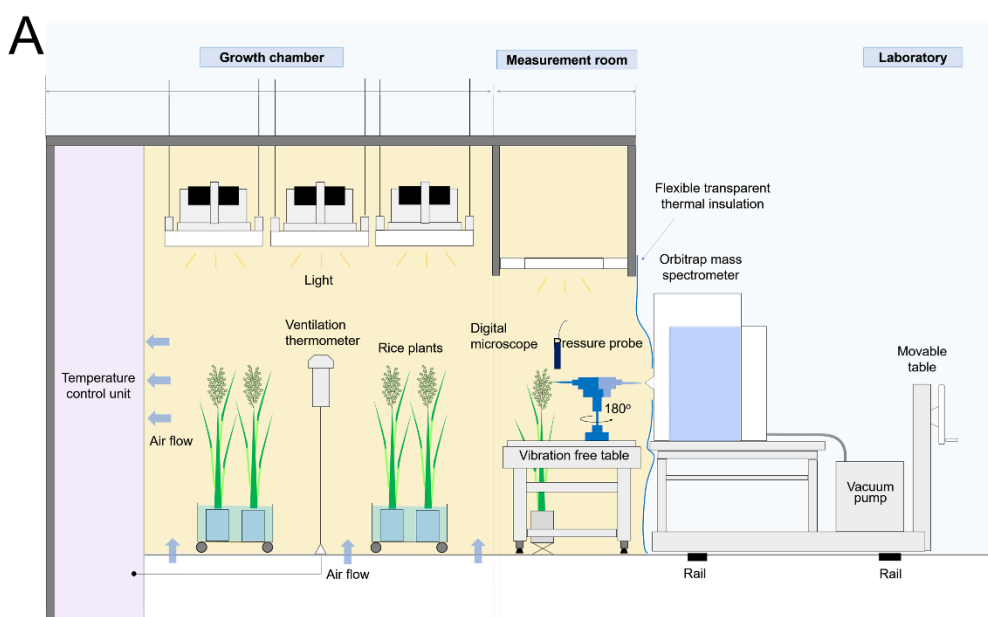
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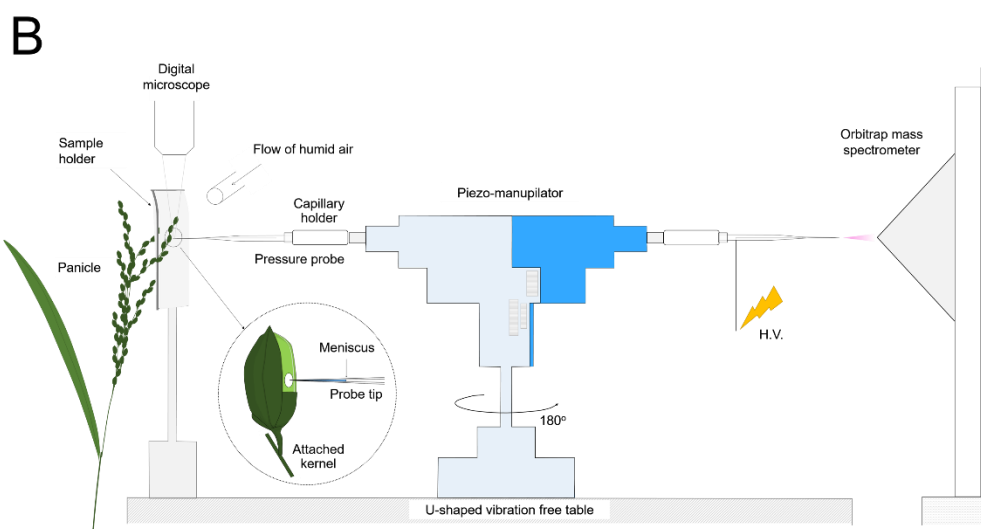
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49 Diagram of the side view of the on-site cell metabolomics system placed in the laboratory (A).
 50 The system is composed of picolitre pressure-probe-electrospray-ionization mass
 51 spectrometry (picoPPESI-MS) and environmental control. The developing plants and the
 52 analytical environment both can be kept at the same environmental conditions using this
 53 system, as shown in light orange, isolated from the laboratory temperature by putting a
 54 flexible transparent thermal insulation (indicated in blue line) at the border. B, expanded
 55 diagram of the picoPPESI-MS system shown in red rectangle in A. This method allows to
 56 perform cell specific metabolome analysis in intact plants being treated at a set temperature.
 57 In the outer endosperms, a part of pericarp tissue was removed (see Materials and Methods).
 58 By utilizing a cell pressure probe, the probe tip was introduced into the target outer
 59 endosperm cells (see Figure. 3A), where the high frequency of chalkiness is expected to
 60 form at maturation, and the cell sap discharged was collected. The Piezo-manipulator was
 61 immediately rotated 180° and then the metabolites in the cell sap were instantly analyzed in
 62 Orbitrap mass spectrometer by applying a high-voltage.