

Extended Data Fig. 3 PABP forms condensates in response to heat stress, associated with Fig. 3.

a-b. As illustrated in the epidermal cells of *N. benthamiana* leaves, PABP-GFP is co-localized with the nuclear marker H2B-mCherry. (A) Merged fluorescence images and (B) fluorescence intensity profiles (red solid lines) confirm nuclear localization.

Scale bar = 20 μm.

c-d. Co-expression of PABP-GFP and plasma membrane marker OslT16A-mcherry shows no localization of PABP to the plasma membrane. (c) Merged fluorescence images and (d) fluorescence intensity profiles (red solid lines) confirm the lack

- of membrane localization, suggesting PABP is primarily localized in the cytoplasm and nucleus. Scale bar = $20 \mu m$.
- e. PABP-GFP forms condensates in both cytoplasm and nucleus under HS conditions at 30°C and 35°C for 30 minutes in the epidermal cells of N. benthamiana leaves. Scale bar = 20 μ m.
- f. Investigation of the condensate frequencies of PABP in nuclei and cytoplasm HS conditions at 30°C and 35°C. Statistical analysis was performed using one-way ANOVA (* p < 0.05, ** p < 0.01). The resulting data are presented as the mean \pm SD (n = 5 biological replicates). For each biological replicate, at least 50 cells were analyzed.
- g. Pretreatment with cycloheximide (CHX) suppresses the formation of PABP condensates in the epidermal of N. benthamiana under heat stress, while DMSO (control) treatment does not affect condensate formation. Furthermore, neither CHX nor DMSO induces PABP condensate formation at room temperature. Scale bar = $20 \mu m$.
- h. Investigation of the condensate frequencies of PABP in nuclei and cytoplasm under CHX treatment. Statistical analysis was performed using one-way ANOVA (* p < 0.05, ** p < 0.01). The resulting data are presented as the mean \pm SD (n = 5 biological replicates). For each biological replicate, at least 50 cells were analyzed.
- i. Time-lapse imaging of PABP-GFP under HS conditions at 42° C (with images captured every 2 seconds) reveals the rapid formation of PABP condensates in the cytoplasm of epidermal cells in *N. benthamiana*. Scale bar = $20 \, \mu m$.