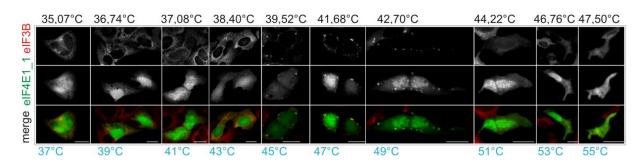
## Additional figure 1. Precise optimization and accurate calibration of temperature measurement

## were essential for efficient and reproducible SGs formation.



U2OS cells were grown on a glass coverslip and transfected with an expression vector coding for the GFP-eIF4E1\_1 fusion protein. Nineteen hours post-transfection, the cultivation dish was placed on a preheated thermoblock, and a submersible temperature probe was placed into the medium in direct contact with the coverslip. The medium was subsequently exchanged with another one pre-warmed to the required temperature. The dish containing the coverslip was closed, covered with a plastic box and incubated on the thermoblock for 30 min. Temperatures of the pre-warmed medium and the thermoblock were set experimentally to reach the required temperature on the coverslip. The temperature was read at the beginning and at the end of the treatment. The calculated mean temperature values are displayed above each panel. The corresponding temperature values that were set on the thermoblock are shown below each panel in blue. Following heat shock, cells were fixed and assessed for eIF3B-stained SGs. The temperature gradient clearly shows that only a narrow range of high-fevered temperatures was suitable for the efficient induction of SG formation. Scale bar, 20 µm.