

BeatBox-iST workflow for efficient, high-throughput protein analysis of different cell types

¹Chloé Moritz, ¹Katrin Hartinger, ¹Zuzana Demianova | ¹PreOmics GmbH, Martinsried, Germany

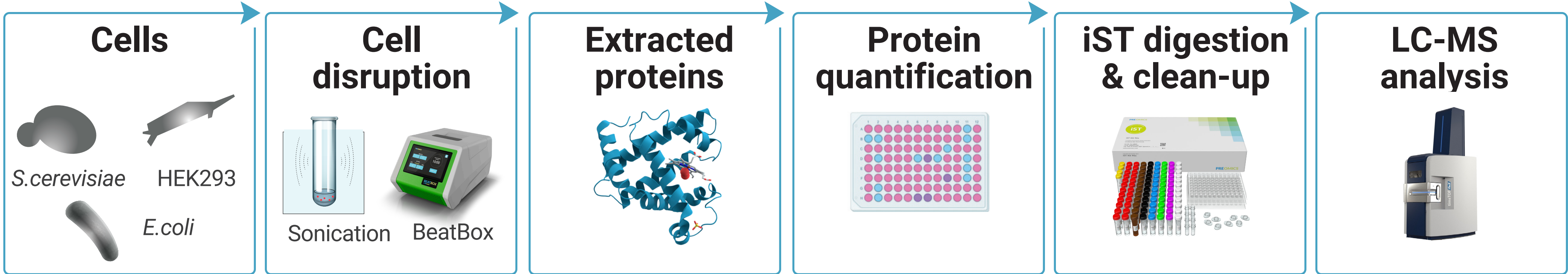
Spotlights

- Up to 96 samples homogenized in 10 min¹
- Small footprint, quiet operation at a steady temperature for the modern lab
- From eukaryotic to prokaryotic cell types
- From thousands to ten million of cells per well
- Seamless integration with iST workflows²

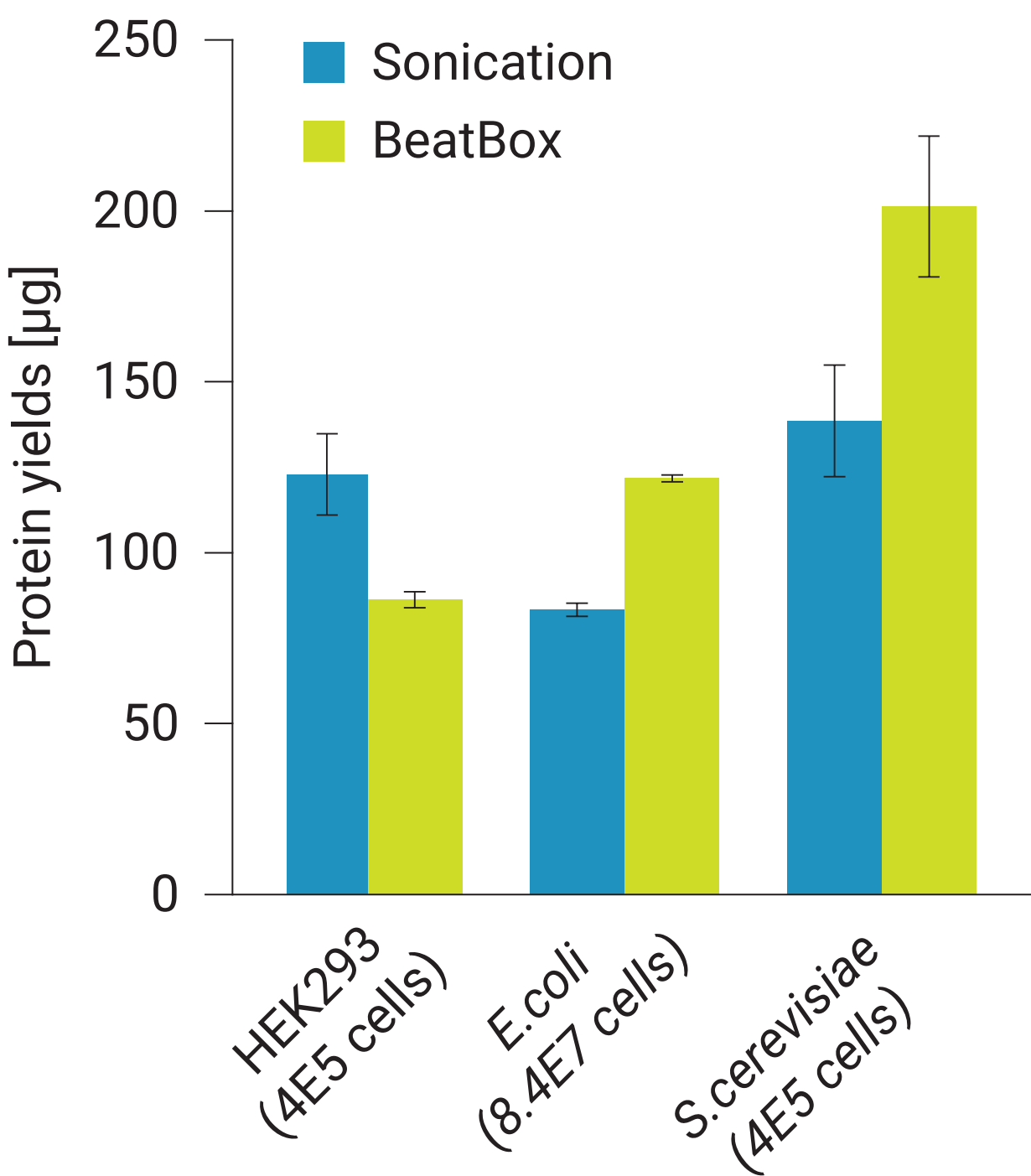
Materials & Methods

- **Input:** *S.cerevisiae* (4.4E6 cells), *E.coli* (8.4E7 cells) and HEK293 (4E5 cells)
- **Sonication:** Boiling step (95 °C, 10 min) followed by 10 sonication cycles, 30 sec on, 30 sec off in iST LYSE buffer
- **BeatBox:** No boiling step except for yeast (95 °C, 10 min), followed by 10 min homogenization with standard power setting in iST LYSE buffer
- **Sample digestion/peptide clean-up:** iST workflow
- **Protein assay:** Micro BCA™ Protein Assay Kit (ThermoFisher Scientific)
- **MS and data analysis:** EASY-nLC™ 1200 - TimsTOF Pro, MaxQuant (v 2.0.1.0), STRING (v11.5)

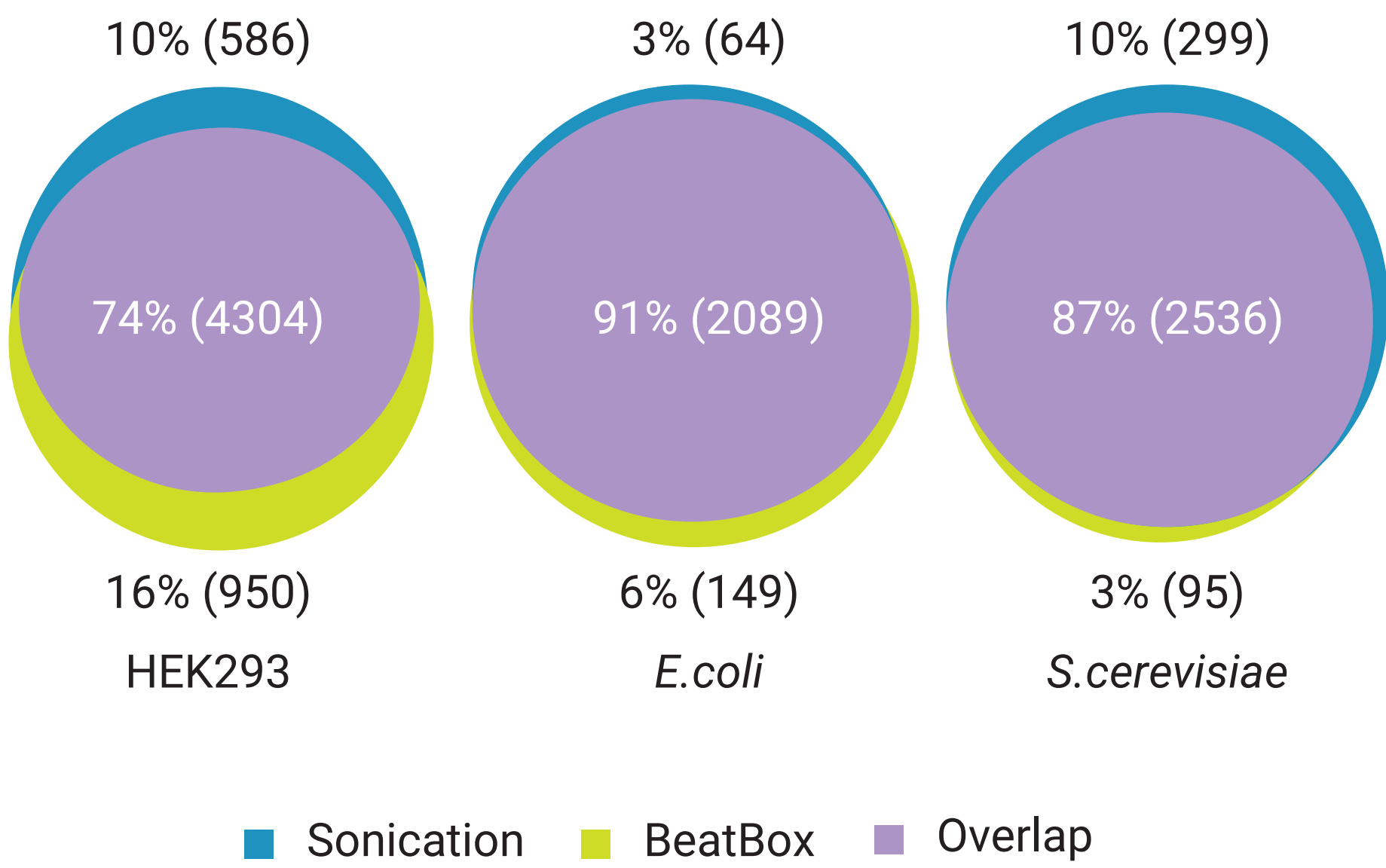
Workflow



Results & Discussion



Protein yields



Protein identification rate

Shared GO	BeatBox specific GO	Sonication specific GO
Intracellular	Organelle membrane	Catalytic complex
Intracellular organelle	Microtubule cytoskeleton	Autophagosome
Organelle	Microtubule organizing center	
Intracellular membrane-bounded organelle	Centrosome	
Membrane-bounded organelle	Spindle	
Cytoplasm	TRAPP complex	
Nucleus	Endosome	
Nucleoplasm	Ubiquitin ligase complex	
Cytosol	Endomembrane system	
Nuclear lumen	Bounding membrane of organelle	
Protein-containing complex	Organelle envelope	
Cellular anatomical entity	Vesicle tethering complex	
Transferase complex	Lysosomal membrane	
Intracellular organelle lumen		

Specific Gene Ontology Cellular Component

- BeatBox **significantly improves protein yields** for *E.coli* and *S.cerevisiae*
- For HEK cells, BeatBox provided better protein IDs despite lower protein yields
- **Similar protein groups** IDs were obtained for each species
- HEK293 cell line shows a higher difference between extracted proteins by BeatBox and the sonication technique
- **CVs below 3%** for protein groups and peptides IDs
- HEK293 cell line lysed by BeatBox show solid improvements in the enrichment of **membrane-related protein groups**

Conclusions

- BeatBox provides high-throughput, efficient, and reproducible lysis across various cell types
- Similar and in-depth proteomic data were obtained using both homogenization techniques
- Equal protein IDs and superb reproducibility were achieved with 4-time less cells in the sample
- Enrichment of cellular component protein groups was measured in BeatBox lysed cells.

References

¹Technical note: BeatBox: Tissue homogenization simplified (www.preomics.com/resources)
²Application note: High-throughput homogenization technique for deeper analysis of multiple mouse tissue proteomes (www.preomics.com/resources)