PREOMICS

iST Technology

Sample Preparation You Can Trust

Setting the Standard for Protein Analysis

Sample Collection

BEATBOX™

INPUT

Protein Extraction
(Tissue homogenization & cell lysis)

PROTEOMICS SAMPLE PREPARATION

PROTEIN

PEPTIDE

Protein Solubilization & Denaturation
(Lysis, denaturation, reduction & alkylation)

Protein Digestion
(Lys-C & trypsin)

Peptide Purification
(Peptide clean-up)

iST WORKFLOW FULLY AUTOMATABLE ON THE PREON™

3-step iST workflow for proteomics sample preparation

LYSE	DIGEST	PURIFY
10 MIN AT 95°C	1 HOUR AT 37°C	1 HOUR AT RT

DATA

ANALYSIS

LC-MS & Data Analysis

Universal Solution
- From biological fluids, cells and tissues to plants
- Versatile format for protein input from 1 μg to 100 μg
- Diverse workflows: label-free & chemical labeling

Reliable Results
- Automatable all-in-one solution
- Standardized workflows for reproducibility
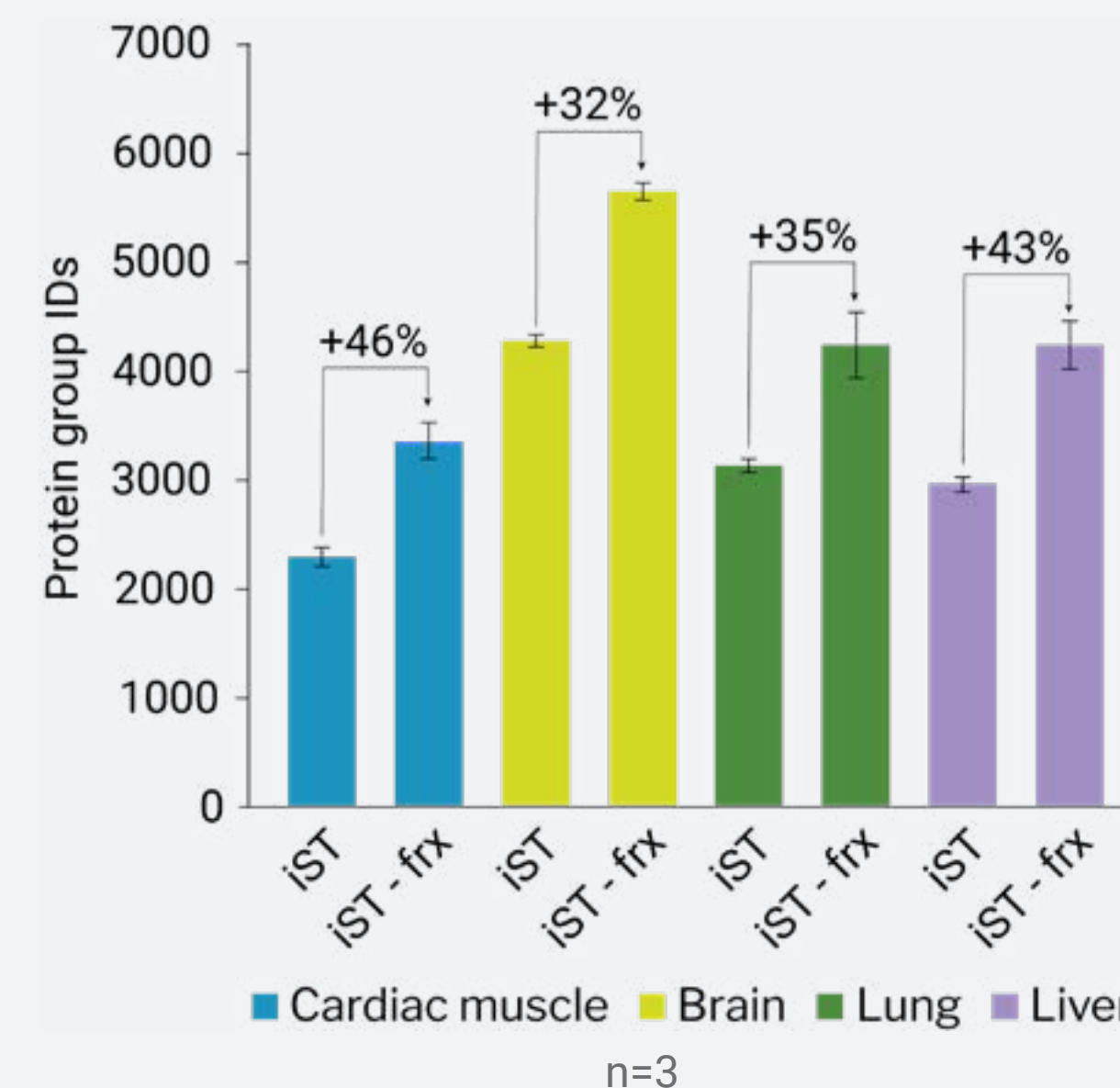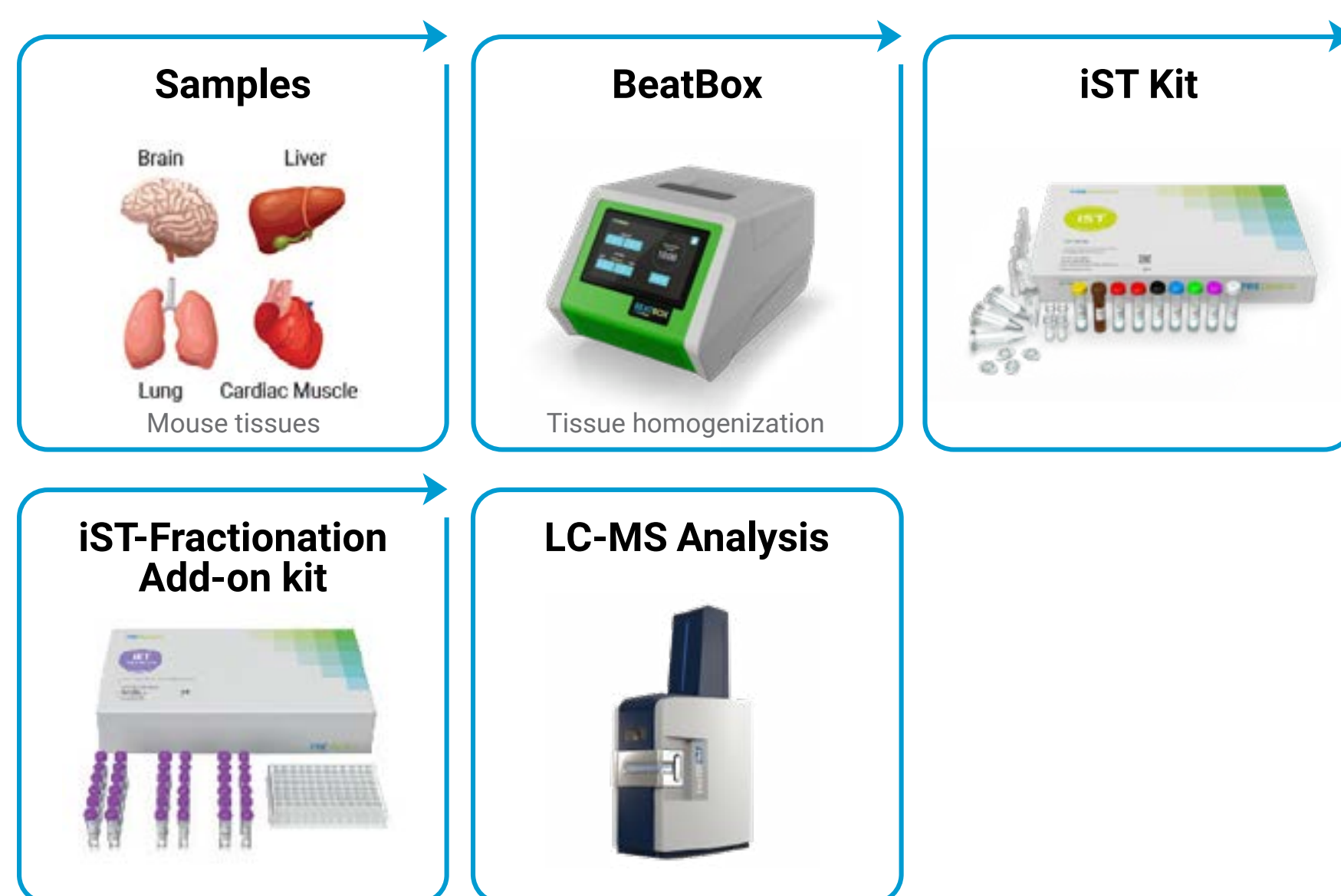- Validated by scientific community

Focus on Biology
- Minimal hands-on time
- Concentrate on data analysis
- Purified peptides enhance your LC-MS performance

iST Technology Applications

PREOMICS

Reproducible in-depth coverage of a variety of tissue proteomes

Samples

Brain   Liver

Lung   Cardiac Muscle

Mouse tissues

BeatBox

Tissue homogenization

iST Kit

iST-Fractionation Add-on kit

LC-MS Analysis

Superb coverage of various mouse tissue proteomes was achieved by combining BeatBox tissue homogenization with the iST-fractionation (iST-frx) workflow.

Outstanding reproducibility (CV ~9%) of tissue homogenization is achieved using the BeatBox – iST workflow.

3-step peptide fractionation increases the proteome depth by ~40% compared to unfractionated samples.
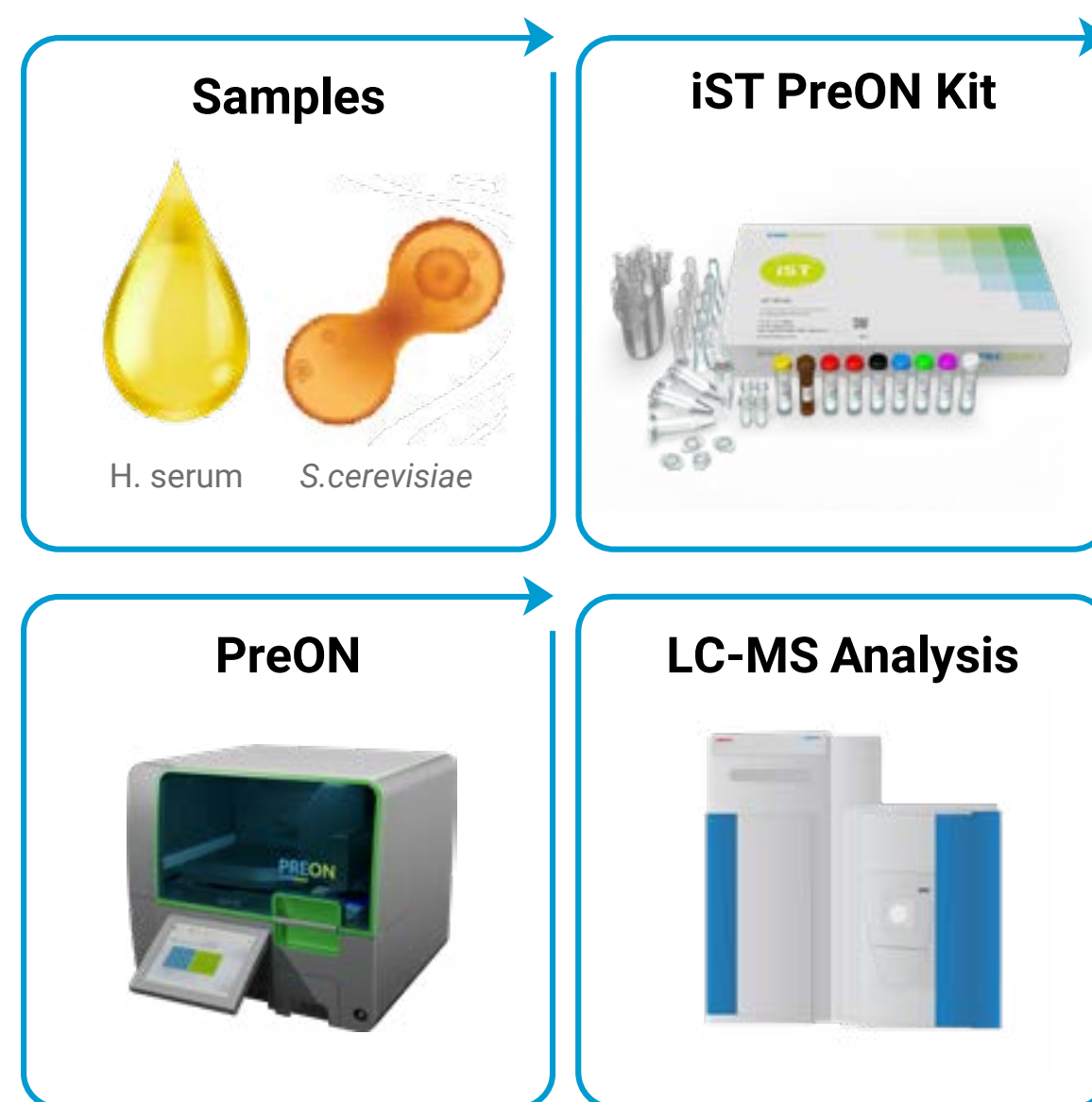
Workflow:

Mouse tissues (1-2 mg wet weight) were homogenized with the BeatBox (10 min, STANDARD mode).

Homogenates were processed with an iST and iST-fractionation (iST-frx) add-on kit (n=3).

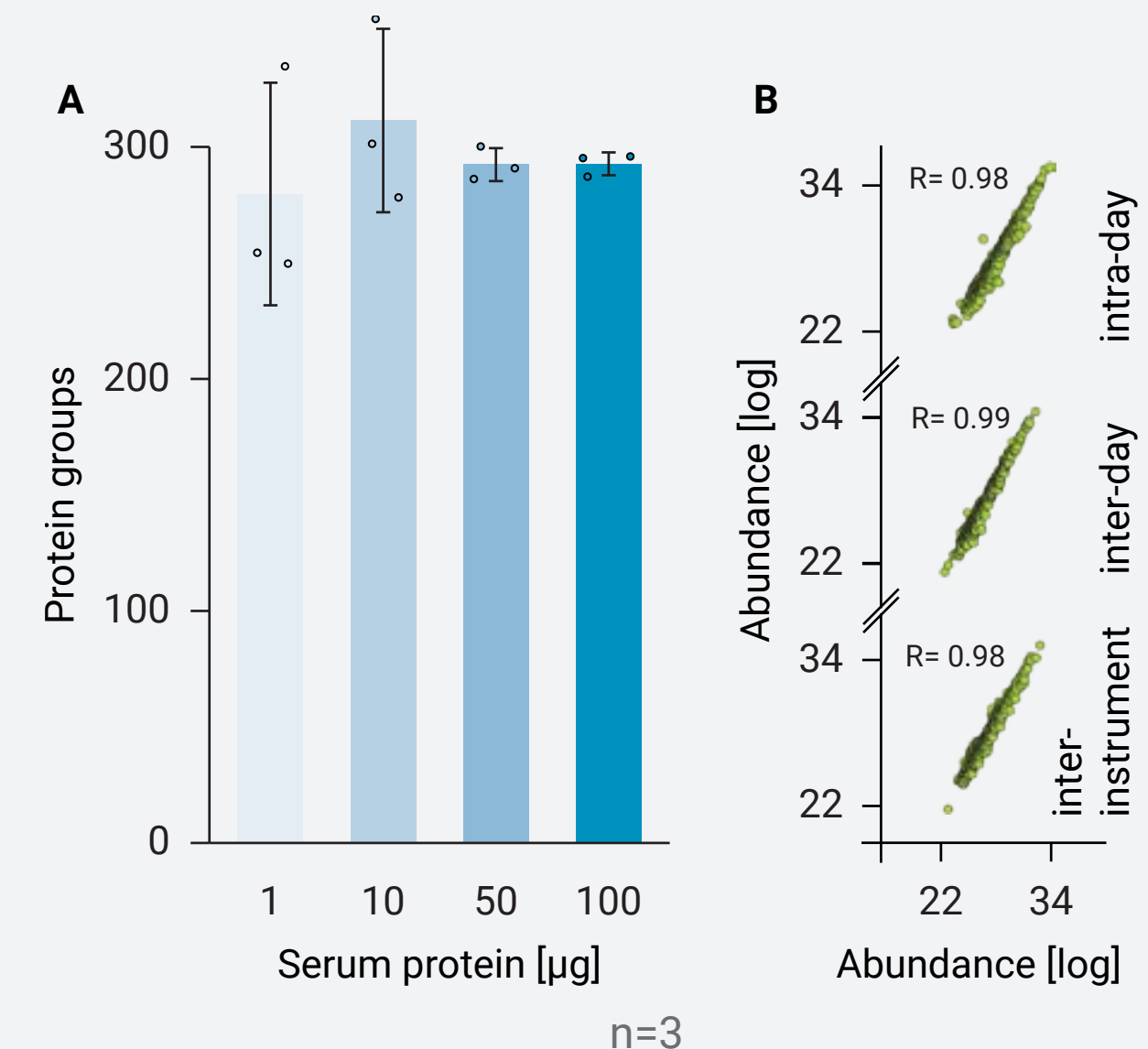Peptides were measured in a 45 min LC-gradient on an EASY nLC 1200 coupled to a timsTOF Pro.

Collaboration with Prof. Dr. Dieter Stoll's lab (University of Applied Sciences Albstadt-Sigmaringen, DE)

PREOMICS

Reliable analyses of serum and yeast samples by a sensitive and fully automated proteomic sample processing

Samples

H. serum   *S.cerevisiae*

iST PreON Kit

PreON

LC-MS Analysis

A

Protein groups

300

200

100

0

1   10   50   100

Serum protein [µg]

n=3

B

Abundance [log]

R= 0.98   intra-day

34

22

R= 0.99   inter-day

34

22

R= 0.98   inter-instrument
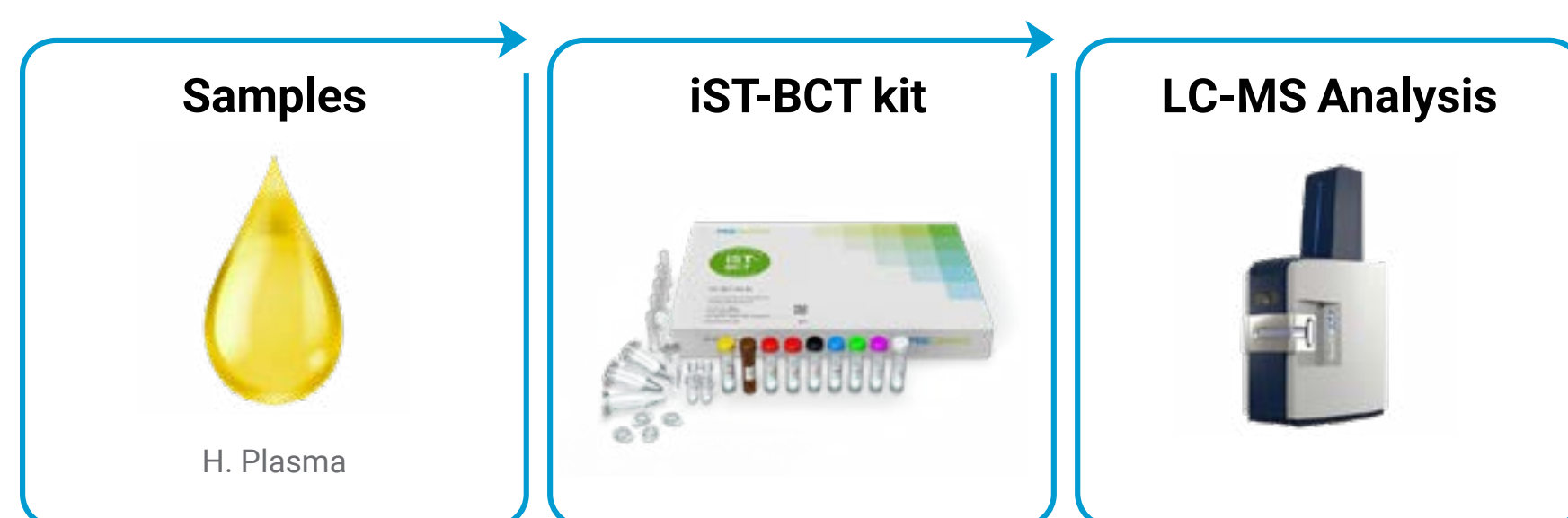
34

22

22   34

Abundance [log]

Easy-to-use, automatable, and standardized kit-based solution for a wide range of sample types.

Highly confident protein identification rate achieved from 1 to 100 µg of serum protein input.

Excellent proteomics workflow reproducibility using a fully automated iST sample preparation on the PreON instrument.

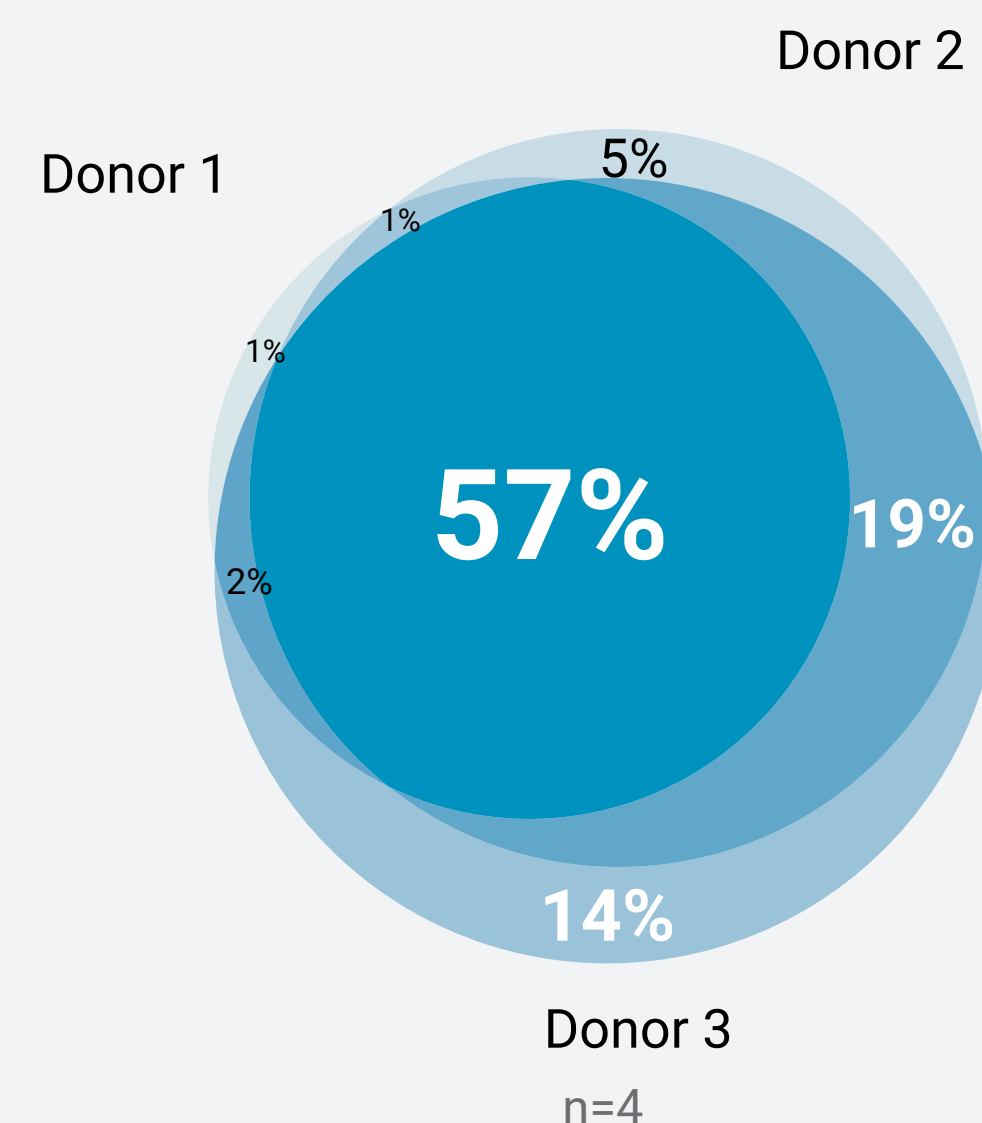## Workflow:

Human serum (1-100 µg of total protein) and yeast pellet (100 µg of total protein) were processed fully automated with the iST kit (n=3) on the PreON platform.

Peptides were measured in a 90 min LC-gradient on an EASY nLC 1000 coupled to a Q Exactive Plus.

Collaboration with Prof. Dr. Bernd Wollscheid's lab (ETH Zurich, CH)

PREOMICS

Optimized sample preparation workflow for biological fluids

Samples → **iST-BCT kit** → **LC-MS Analysis**

H. Plasma

Donor 1

Donor 2

Donor 3

5%

1%

1%

2%

57%

19%

14%

n=4

Easy-to-use 3-step sample preparation to analyze a broad range of biological fluids, from complex plasma to challenging Cerebrospinal fluid.

Nearly 60% of the human plasma proteome is shared between three healthy patient donors.

iST-BCT workflow reduces artificial modifications and improves alkylation rate to enhance proteomic coverage for biological fluids.

Workflow:

Human plasma from 3 different donors (100 µg of total protein) was processed with the iST-BCT kit (n=4).

Peptides were measured in a 60 min LC-gradient on an EASY nLC 1200 coupled to a timsTOF Pro.

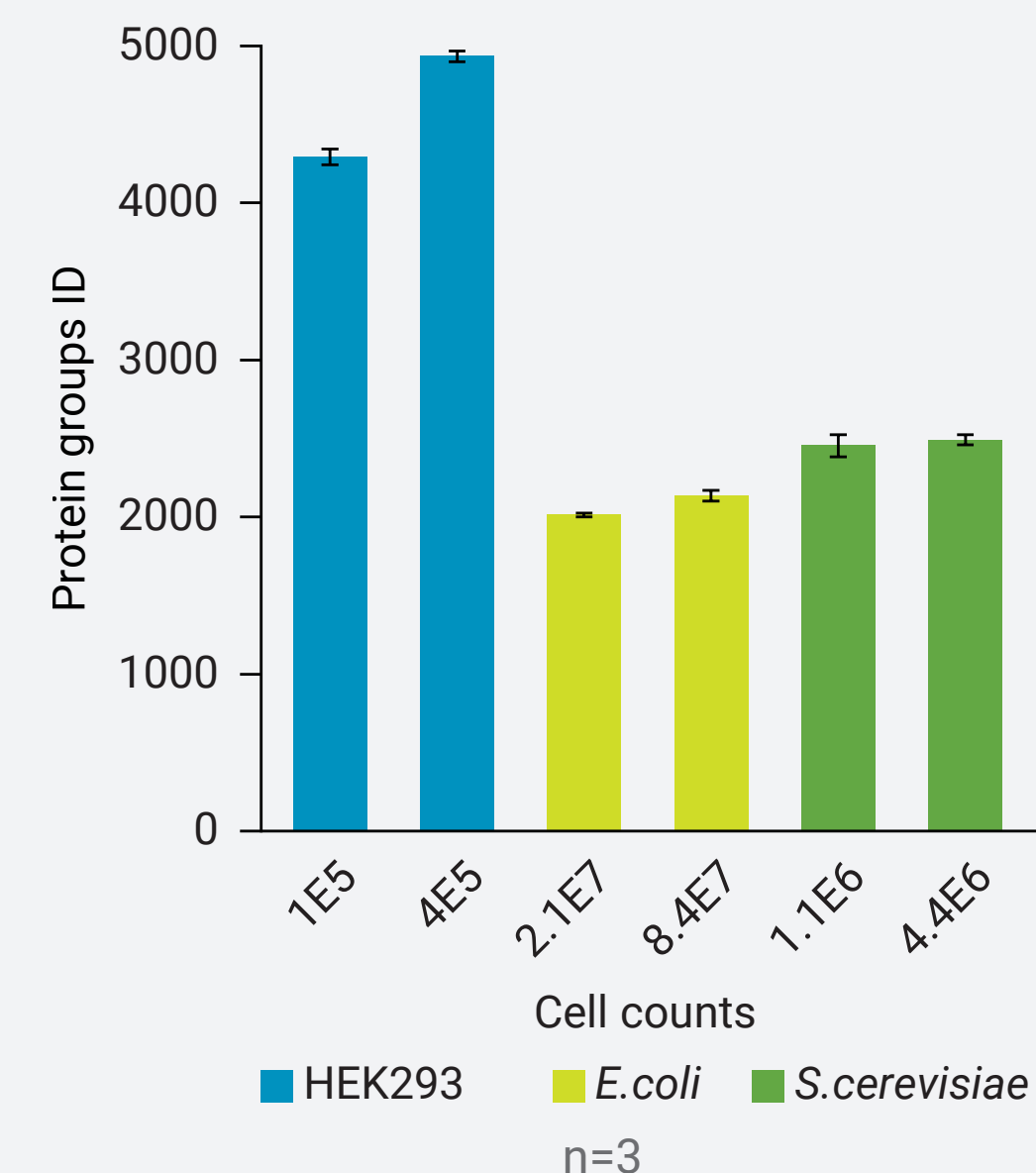# Efficient, reproducible, high-throughput cell lysis on the BeatBox platform
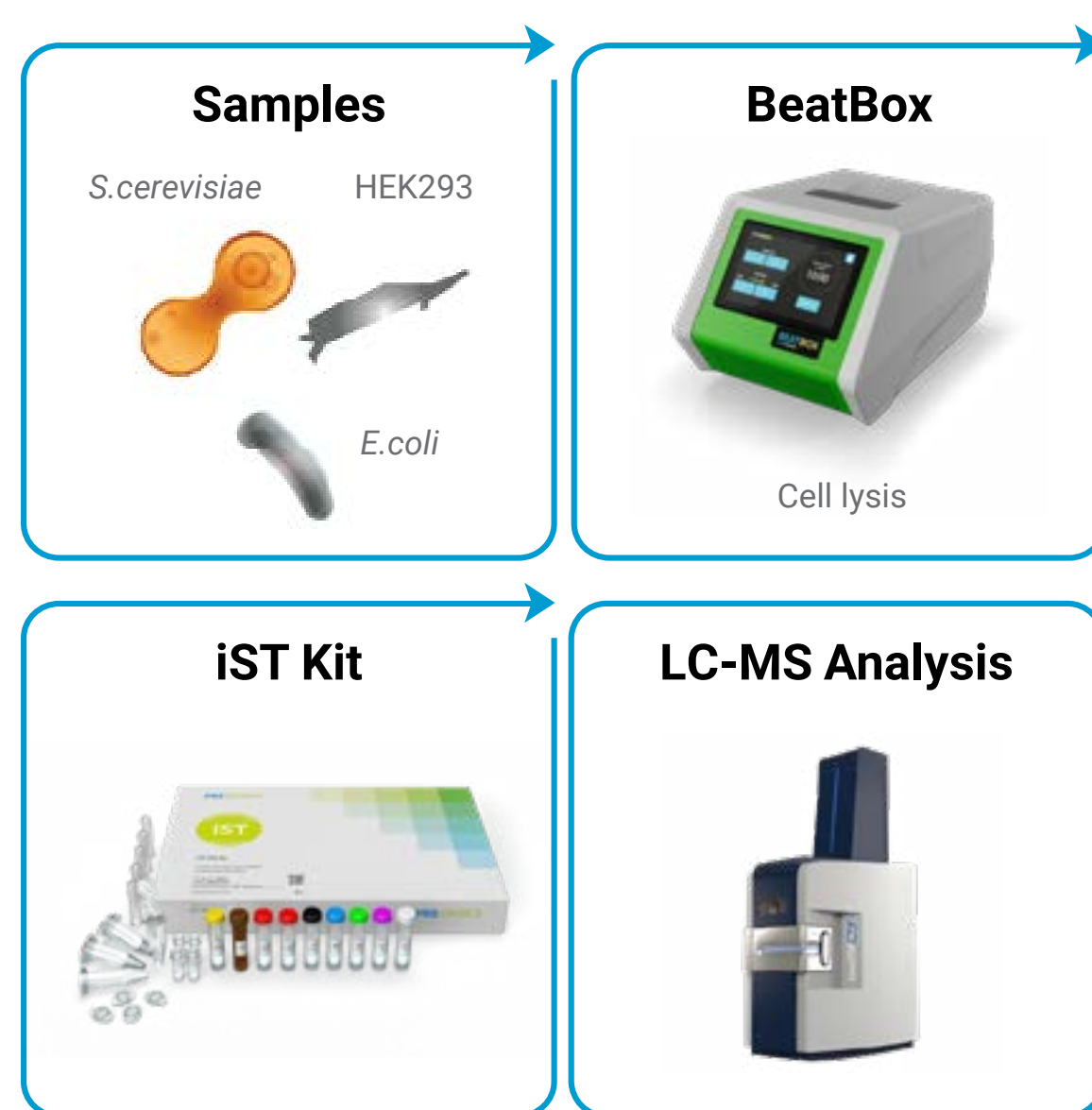
Workflow:

E. coli (2.1E7 and 8.4E7 cells), *S.cerevisiae* (1.1E6 and 4.4E6 cells) and HEK293 (1E5 and 4E5) were lysed with the BeatBox (10 min, STANDARD mode).

Cell lysates were processed with the iST kit (n=3).

Peptides were measured in a 45 min LC-gradient on an EASY nLC 1200 coupled to a timsTOF Pro.
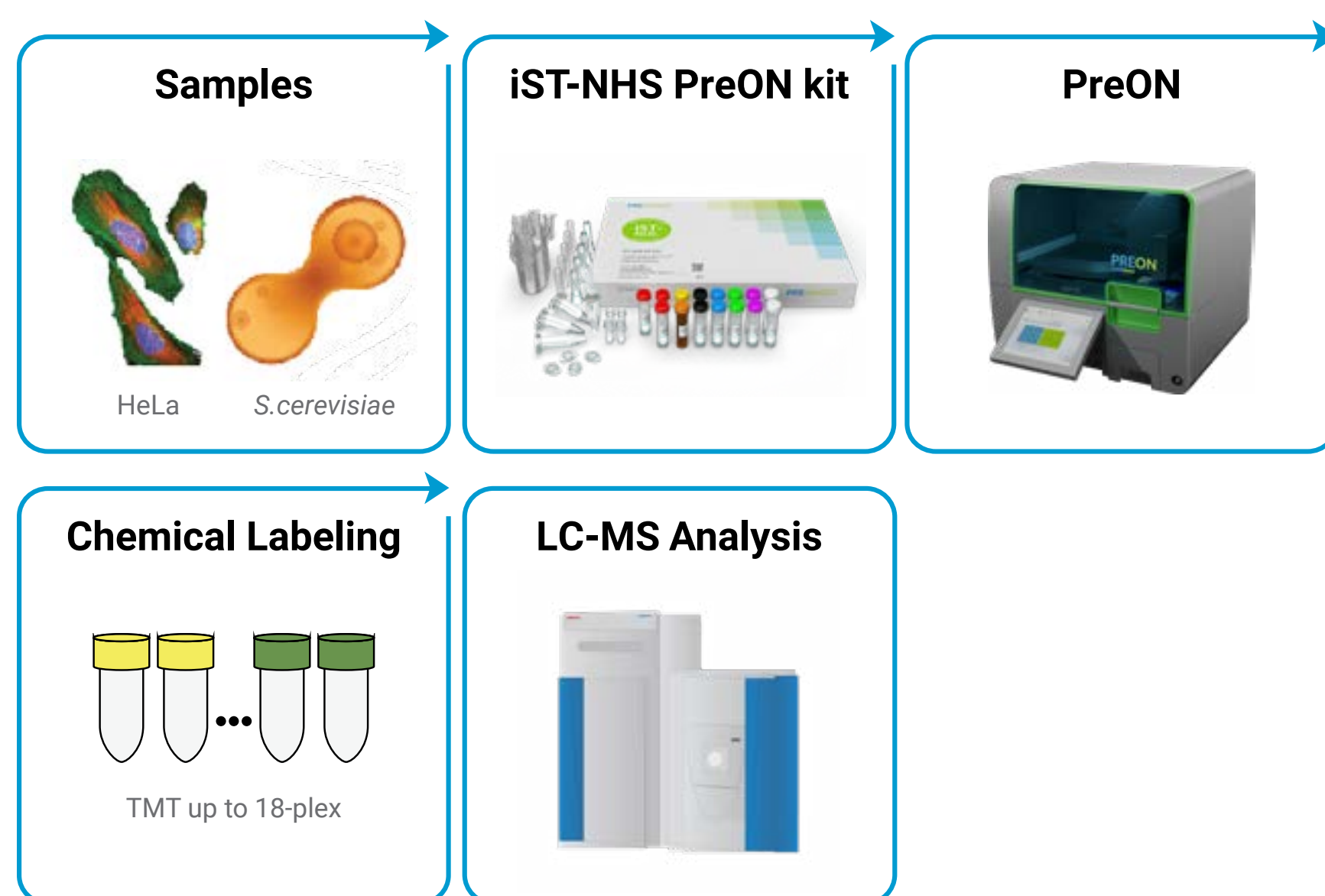
BeatBox enables high-throughput, efficient, and reproducible lysis across many cell types.

Effective cell lysis followed by iST sample preparation for different cell counts resulted in a similar protein group identification rate.

Outstanding reproducibility (CV ~3%) is achieved using the BeatBox – iST workflow.

PREOMICS

Fully automated workflow to enable a highly efficient isobaric labeling

Samples

HeLa   *S.cerevisiae*

iST-NHS PreON kit

PreON

Chemical Labeling

TMT up to 18-plex

LC-MS Analysis

Labeling efficiency [%] — TMT zero, TMT 11-plex, TMT 16-plex

■ *S.cerevisiae*   ■ HeLa
n=4

Workflow:

Cells (50 µg of total protein) were processed with the iST-NHS PreON kit (n=4).

Peptides were labeled with TMT zero, TMT 11-plex and TMTPro 16-plex.

Labeled peptides were measured using a 100 min LC-gradient on an EASY nLC 1200 coupled to a Q Exactive HF-X or Orbitrap Fusion Lumos Tribrid.

Collaboration with Prof. Dr. Bernd Wollscheid's lab (ETH Zurich, CH)
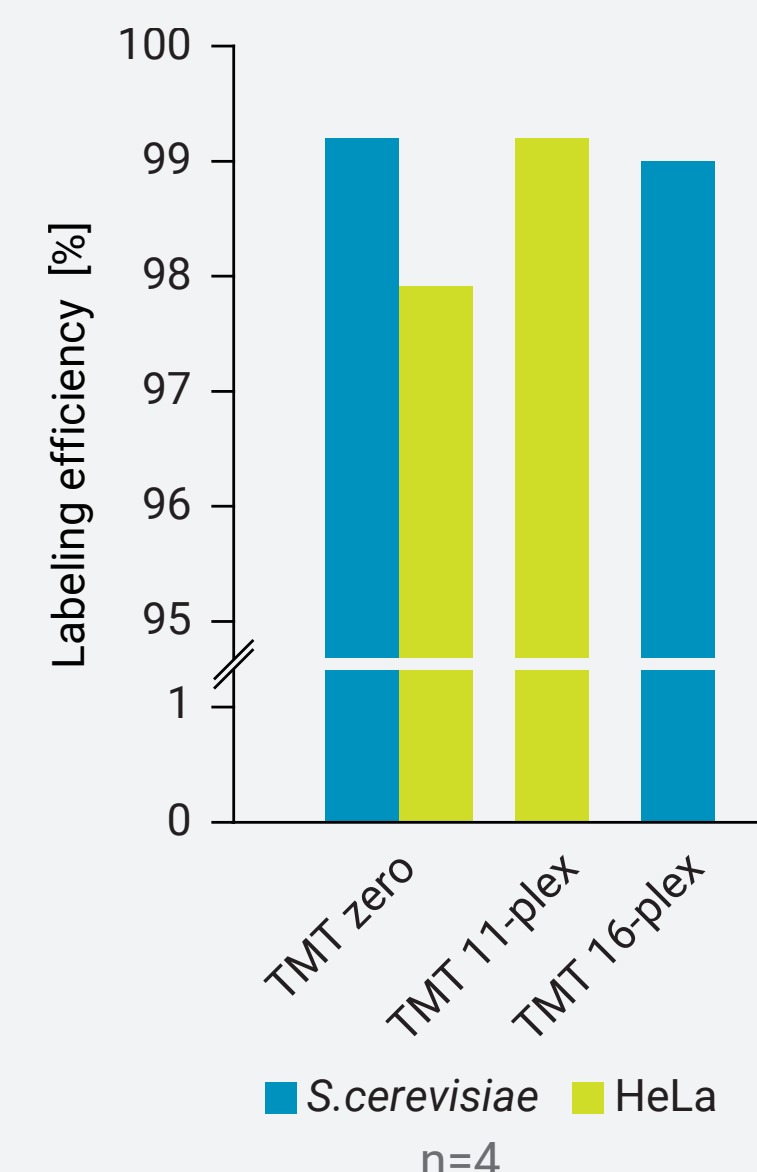
Automation of the iST technology on the PreON enables isobaric labeling of up to 18 samples* in parallel, from cells to labeled peptides in less than 4 hours.
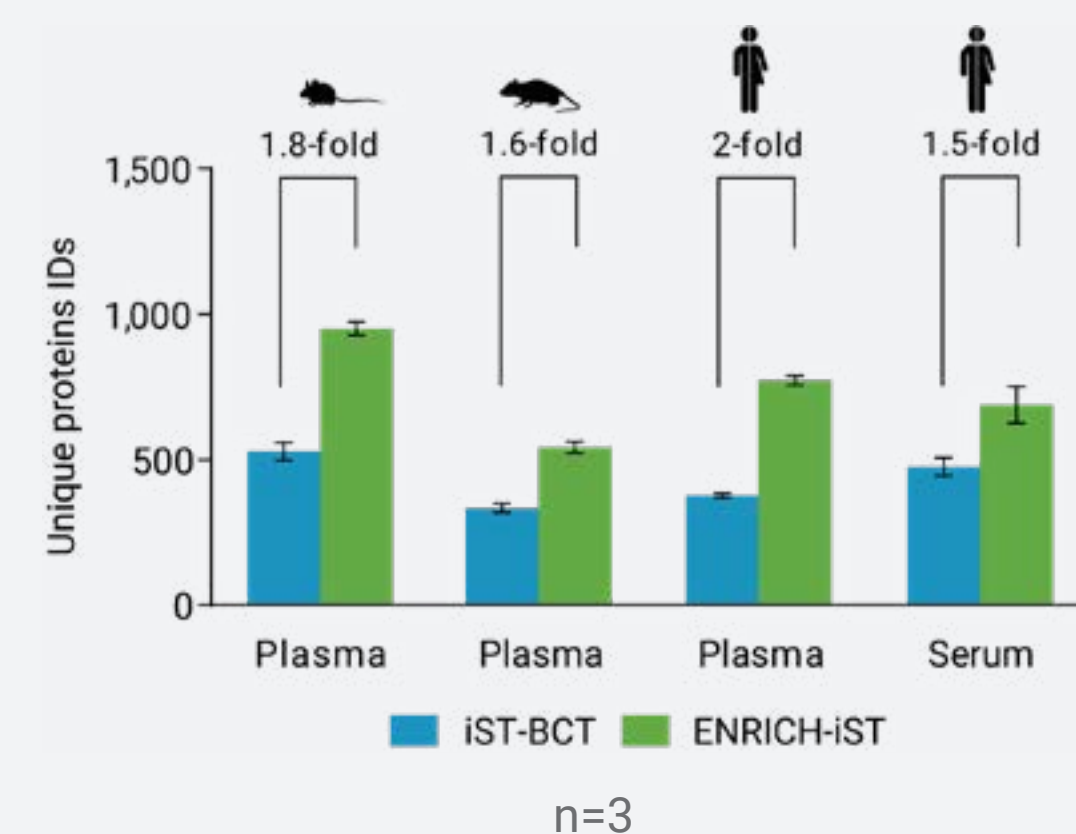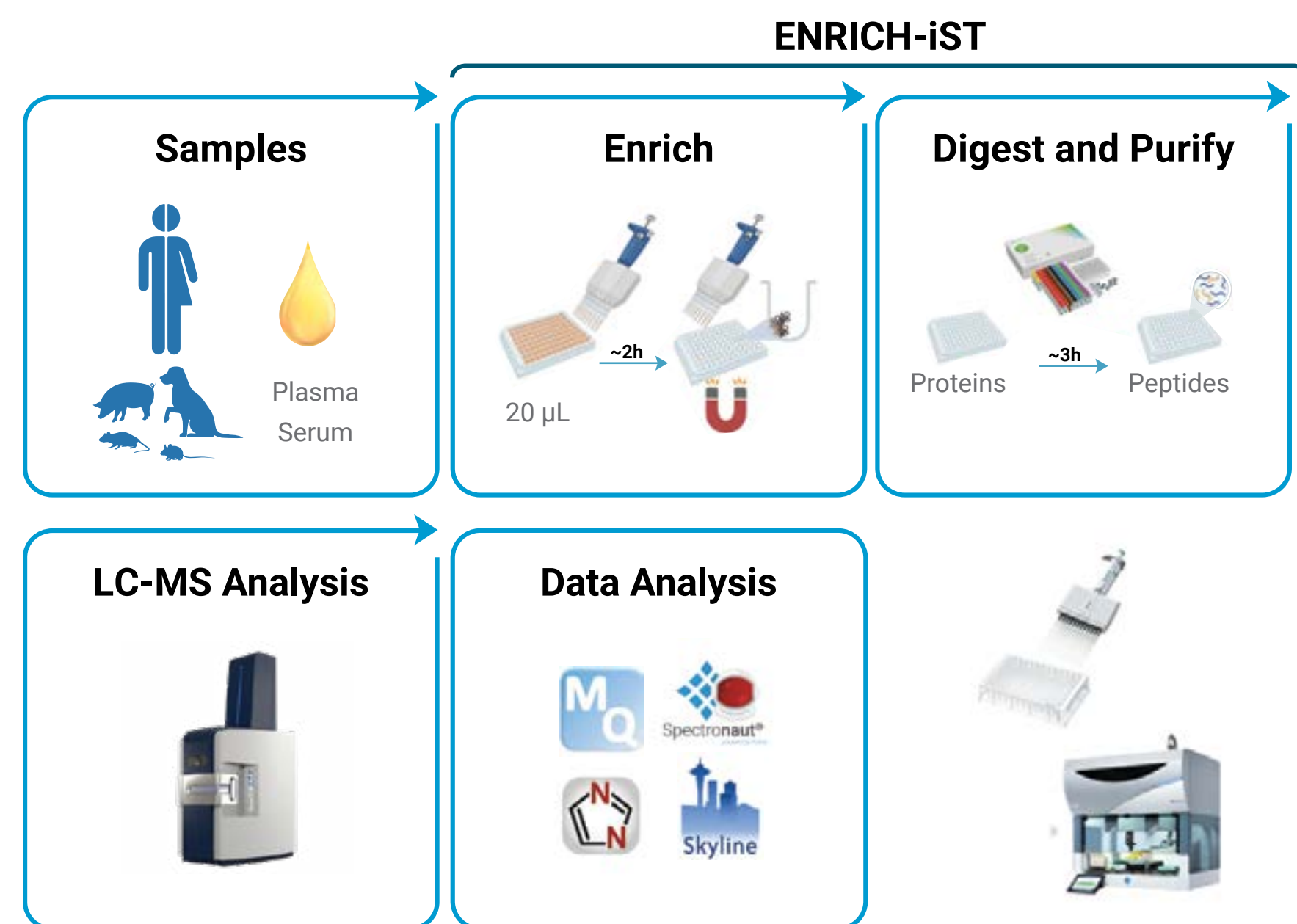
The fully automated workflow increases the throughput and reproducibility of quantitative proteomics experiments using isobaric tags.

Excellent TMT tag labeling efficiency (>98%) even with a label to peptide ratio of 4:1.

*With the TMTpro™ 18-plex upgrade set.

Optimized plasma and serum sample preparation for deeper, high-throughput proteome profiling

ENRICH-iST

Samples → Enrich → Digest and Purify

Plasma Serum

20 µL   ~2h   ~3h   Proteins → Peptides

LC-MS Analysis → Data Analysis

n=3

Workflow:

20 µL of plasma and serum (mouse, rat and human, respectively) was processed with the ENRICH-iST (n=3).

Peptides were measured using a 45 min LC-gradient on a nanoElute® 2 coupled to a TimsTOF Pro HT.

A fast, easy-to-use, and standardized protocol for high reproducibility and less hands-on time, automatable high-throughput processing of up to 96 samples in parallel possible in just 5h.

Species-independent and flexible enrichment technology optimized for plasma and serum samples that enhance proteomics depth compared to the iST-BCT workflow.

ENRICH-iST workflow provides superior robustness with overall reproducibility of CV<15%, which is essential for the reliable execution of large-scale studies.

PREOMICS

Selection of publications and products listing

Selection of publications

- Proteomic analysis distinguishes extracellular vesicles produced by cancerous versus healthy pancreatic organoids, Buenafe et al., 2022, Scientific Reports, doi: *https://doi.org/10.1038/s41598-022-07451-6*
- Supramolecular arrangement of protein in nanoparticle structures predicts nanoparticle tropism for neutrophils in acute lung inflammation, Myerson et al., 2022, Nat. Nanotechnol. 17, 86–97,
doi: *https://doi.org/10.1038/s41565-021-00997-y*
- A clinically compatible drug-screening platform based on organotypic cultures identifies vulnerabilities to prevent and treat brain metastasis, Zhu et al., 2022, EMBO Mol Med 14:e14552, doi: *https://doi.org/10.15252/emmm.202114552*
- A lysosomal biogenesis map reveals the cargo spectrum of yeast vacuolar protein targeting pathwa, Eising et al., 2022, J Cell Biol 221 (4): e202107148, *doi: https://doi.org/10.1083/jcb.202107148*

Products listing

Product	Manufacturer	Product Code	Product	Manufacturer	Product Code
iST 8x	PreOmics GmbH	P.O.00001	iST-NHS 12x	PreOmics GmbH	P.O.00026
iST 96x	PreOmics GmbH	P.O.00027	iST-NHS 96x	PreOmics GmbH	P.O.00030
iST HT 192x	PreOmics GmbH	P.O.00067	iST-NHS-HT 192x	PreOmics GmbH	P.O.00083
iST-BCT 8x	PreOmics GmbH	P.O.00084	ENRICH-iST 8x	PreOmics GmbH	P.O.00163
iST-BCT 96x	PreOmics GmbH	P.O.00099	ENRICH-iST 96x	PreOmics GmbH	P.O.00164
iST-BCT-HT 192x	PreOmics GmbH	P.O.00124	ENRICH-iST 96x HT	PreOmics GmbH	P.O.00165

Want to know more?

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to learn more about the iST technology

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