Implementation of scalable sample preparation workflow solutions in a clinical laboratory

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Learning objectives

» Build from Julia’s discussion on fundamentals

» Workflow solutions to support strategies towards implementation
  » Processing options
    • Manual
    • Automation
    • Homogenizers – sample cleanup for solid phase samples
  » Extraction options
    • Cartridges
    • Plates
  » Evaporation
    • Cartridges
    • Plates

» What now?
» Conclusions
What if chromatographic separations had <1000 theoretical plates?

Acknowledgement to Brian

No leprechauns riding on unicorns

Sample prep can

• Add some resolution elements to an assay
• Protect your LC
• Protect your MS/MS
• Improve data quality

Good chromatography decision making is still required
New this year: Biotage BeadRuptor 24

Biotage® Bead Ruptor 24
Bead Mill Homogenizer

**SOFT SAMPLES**
- Ceramic beads 1.4mm
  - This matrix will provide a quick and reliable homogenization on small and soft animal, plant or human tissues sizes. It is optimal for brain, liver, skin, leaves, gonad etc. For sample volumes under 300mg and 1mL.

**MICROORGANISMS**
- Glass beads 0.5mm
  - Tough microorganisms and gram+ bacteria. It is optimal for spores, yeast, mold, algae, listeria, etc.
- Glass/Silica beads 0.1mm
  - Soft microorganisms and gram- bacteria. It is optimal for E-coli, mycobacteria, salmonella, etc.

**HARD SAMPLES**
- Carbide beads of 0.25mm
  - This matrix will provide a quick and reliable lysis when processing microbial RNA and soil DNA.
- Garnet beads 0.70mm
  - This matrix mix can be described as the Universal bead mix containing particles with sharp edge. This matrix will provide a quick and reliable homogenization of tough samples (skin, muscle etc.) and soil.

Aps in development: extraction of drugs of abuse from hair, bone, tissue
Positive Pressure\textsuperscript{+} Manifolds

Vs.
RapidTrace+ Automated SPE Workstation
New this year: Extrahera
Translation = extraction

» Simple robust & ready to run
» Compact 8 or 4 channel instrument
» Quick processing
» Dual format
» Easy software interface
EVOLUTE EXPRESS
method development plates

They do exist!

Presented at MSACL

Load, Wash, Elute
ISOLUTE SLE\textsuperscript{+}

SPE looks like this:

SLE looks like this:

It follows that it works a bit different

Also available in 96 well plates

No condition
No equilibration
No wash steps
Generic method development plan

**ISOLUTE SLE+ Supported Liquid Extraction Plates: Method Selection**

**ACIDIC DRUG**
- **Buffer 1**
  - Adjust plasma to pH 3.2
  - Mix plasma with 1% v/v aqueous Formic Acid, 1:1 v/v

**NEUTRAL DRUG**
- **Buffer 2**
  - Adjust plasma to pH 6.3
  - Mix plasma with 0.1% v/v aqueous Formic Acid, 1:1 v/v

**BASIC DRUG**
- **Buffer 3**
  - Adjust plasma to pH 8.0
  - Mix plasma with water, 1:1 v/v
- **Buffer 4**
  - Adjust plasma to pH 10.4
  - Mix plasma with 0.5 M Ammonium Hydroxide, 1:1 v/v

**Extraction Solvent 1**
- **MTBE**
  - Apply 1 mL for the SLE+200
  - Apply 2 x 900 μL for the SLE+400

**Extraction Solvent 2**
- **DCM**
  - Apply 1 mL for the SLE+200
  - Apply 2 x 900 μL for the SLE+400

**Extraction Solvent 3**
- **DCM/IPA, 95:5 v/v**
  - Apply 1 mL for the SLE+200
  - Apply 2 x 900 μL for the SLE+400

**Extraction Solvent 4**
- **Ethyl Acetate**
  - Apply 1 mL for the SLE+200
  - Apply 2 x 900 μL for the SLE+400
Patented Vortex Evaporation Technology

TurboVap® blowdown evaporators use a patented vortex evaporation system that increases the speed of evaporation by a factor of 10 compared with other techniques. The combination of patented nitrogen gas vortex shearing technology and temperature is used to optimize sample drying. Nitrogen is recommended due to its inert nature which minimizes sample oxidation.
Evaporation  
96 well plate options

**SPE Dry**

Designed for high throughput laboratories, the Biotage® SPE Dry 96 and Biotage® SPE Dry 96 Dual Sample Concentrator Systems provide efficient solvent evaporation in microplate format and are compatible with 24-, 96- and 384-well collection plates. Heated gas flow from above and below the collection plate ensures efficient solvent evaporation.

**TurboVap® 96**

TurboVap® 96 Concentration Evaporator Workstation is a microprocessor-controlled evaporation system for simultaneous, automated concentration of multiple samples with unattended operation, convenience and speed. It can accommodate one or two 96-well microplates or deepwell plates.
Implementing methods
I bought the technology, what do I do now?
Colleague versus customers

Applications support provided

25 published references with collaborators in 2014

Clinical applications include

• Drugs of abuse testing
• Alcohol abuse biomarkers
• Biogenic amine / Catecholamine metabolites
• Biomonitoring for exposure assessment
• Water soluble vitamins
Final thoughts

» Considerations for implementing should consider many variables including cost and throughput
  » Cartridges vs plate

» Automation should always be considered

» If you have a problem, please reach out
  » Sorbent selection
  » Issues with recovery
  » Method scalability

A grateful acknowledgement to the Biotage family

And thanks for everything Karl!
Thank You For Your Attention!

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