22nd Annual Symposium
Clinical & Pharmaceutical Solutions through Analysis

Microsampling & Patient Centric Sampling

A journey through what it is and how you can incorporate it into your workflows

Monday 28th October 2019, 12:00-16:00 University Grille

Short Course Outline

12:00 Lunch, Welcome & Introductions
Neil Spooner & Joe Siple

12:30 Introduction to Microsampling
Presentation - Neil Spooner

13:00 Considerations for drug bioanalytical assay method development & validation, and clinical implementation strategies
Presentation & Discussion - Tim Olah & Enaksha R Wickremsinhe

14:00 Break

14:15 Considerations for Clinical Operations
Presentation & Discussion - Melanie Anderson

15:15 Future Directions
Discussion - Kevin Bateman & Neil Spooner

15:45 Wrap-up & next steps

16:00 End

Introduction to Microsampling

Neil Spooner PhD, CChem, FRSC (*neil@spoonerbioanalytical.co.uk*)

Founder & Director - Spooner Bioanalytical Solutions Ltd, UK

Senior Visiting Research Fellow - School of Life & Medical Sciences, University of Hertfordshire, UK

Senior Editor – Bioanalysis Journal

Perspective

For reprint orders, please contact: reprints@future-science.com

Bioanalysis

Microsampling: considerations for its use in pharmaceutical drug discovery and development

Neil Spooner*,[1], Kenneth D Anderson[2], Joe Siple[3], Enaksha R Wickremsinhe[4], Yang Xu[2] & Mike Lee[5]

[1]Spooner Bioanalytical Solutions Ltd, Hertford, UK & School of Life & Medical Sciences University of Hertfordshire, UK
[2]Merck & Co. Inc., Department of Pharmacokinetics, Pharmacodynamics & Drug Metabolism, West Point, PA 19486, USA
[3]New Objective Inc, 2 Constitution Way, Woburn, MA 01801, USA
[4]Lilly Research Laboratories, Eli Lilly & Company, Indianapolis, IN, USA
[5]Milestone Development Services, PO Box 178, Newtown, PA 18940-0178, USA
*Author for correspondence: neil@spoonerbioanalytical.co.uk

There is growing interest in the implementation of microsampling approaches for the quantitation of circulating concentrations of analytes in biological samples derived from nonclinical and clinical studies involved in drug development. This interest is partly due to the ethical advantages of taking smaller blood volumes, particularly for studies in rodents, children and the critically ill. In addition, these technologies facilitate sampling to be performed in previously intractable locations and occasions. Further, they enable the collection of samples for additional purposes (extra time points, biomarkers, sampling during a clinical event, etc). This article gives a comprehensive insight to the utilization of these approaches in drug discovery and development, and provides recommendations for best practice for nonclinical, clinical and bioanalytical aspects.

First draft submitted: 26 February 2019; Accepted for publication: 30 April 2019; Published online: 20 June 2019

What is microsampling?

Conventional Volumes
(200 µL – ? mL)

Micro-volumes
(≤ 100 µL)

Technologies for collecting & analysing smaller blood & plasma / serum volumes for the accurate determination of circulating concentrations of therapeutic drugs, metabolites & biomarkers in non-clinical & clinical studies

Spooner *et al* (2019) *Bioanalysis* **11(10)** 1015–1038

Non-Clinical – Why do this?

Ethical - 3Rs

- Reduction in rodent animal numbers
  - Elimination of TK satellites reduces number of animals by 30-40%
    - Effects primarily on reticulocytes; no affect in overt toxicity assessment, e.g., hepatotoxicity, renal toxicity*
  - Serial TK & PK sampling in mice
  - Discovery PK/PD, mouse TK & PK/PD & juvenile studies
- Refinement of bleeding technique
  - Reduction, or elimination of rodent warming
  - Sampling from more convenient / less disruptive location

*Powles-Glover *et al* (2014) *Reg. Toxicol. Pharmacol*. **68**, 325-331

Non-Clinical – Why do this, cont'd?

Improved data quality

- Exposure data in main study animals, rather than additional satellites
- Direct correlation of exposure with PD and toxicological outcomes

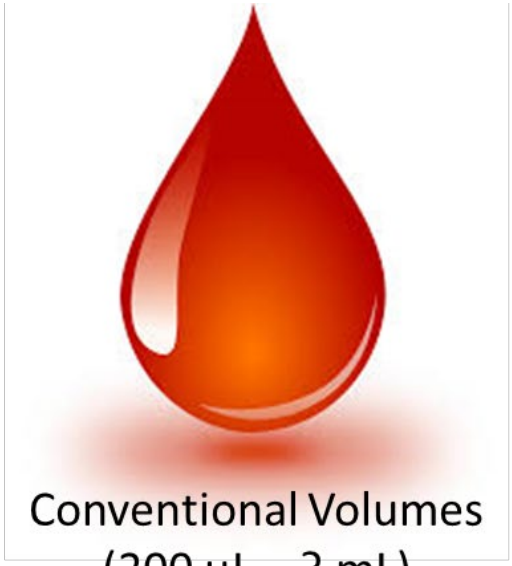
Enables samples to be taken for other purposes

- Additional PK/TK timepoints, biomarkers, metabolites, Clin. Path. determinations, etc.

Cost

- Reduced animal numbers, housing, drug substance
  - …..but, consumable costs are higher

See https://nc3rs.org.uk/microsampling



Micro-volumes
(≤ 100 µL)

However, this course is NOT simply about microsampling…

…..But it IS about moving beyond conventional clinical blood sampling

It’s about collecting……

…the appropriate sample…

…in a location that is most convenient for the patient…

…that provides high quality information

This may be blood sample volumes of 10 µL, or it may be 250 µL

With the **PATIENT** at the centre of our considerations

Patient Centric Sampling

Benefits of Patient Centric Blood Sampling

Quality	Obtaining a high quality blood / plasma / serum sample for accurate quantitative determination of drugs, drug metabolites & endogenous molecules
Patient	Minimising the impact on the human patient / consumer • Optimising blood volume sampled • Minimising pain • Facilitating convenience
New Data	Generating concentration data in situations that are currently difficult, or impossible to work with

Dried Blood Spots

Established for neonatal screening for 50+ years

Delivers all the benefits outlined

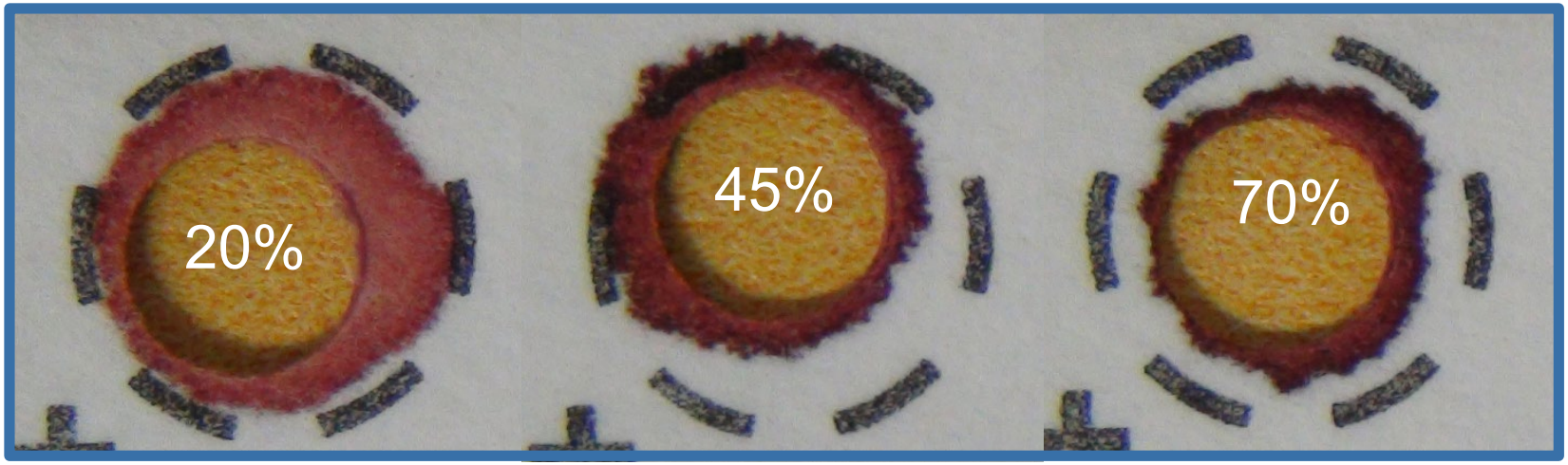
PLUS - Simpler process

- Removes need for centrifugation or sub-aliquots
- Dry ice and freezers not required
  - BIG cost savings on sample shipments

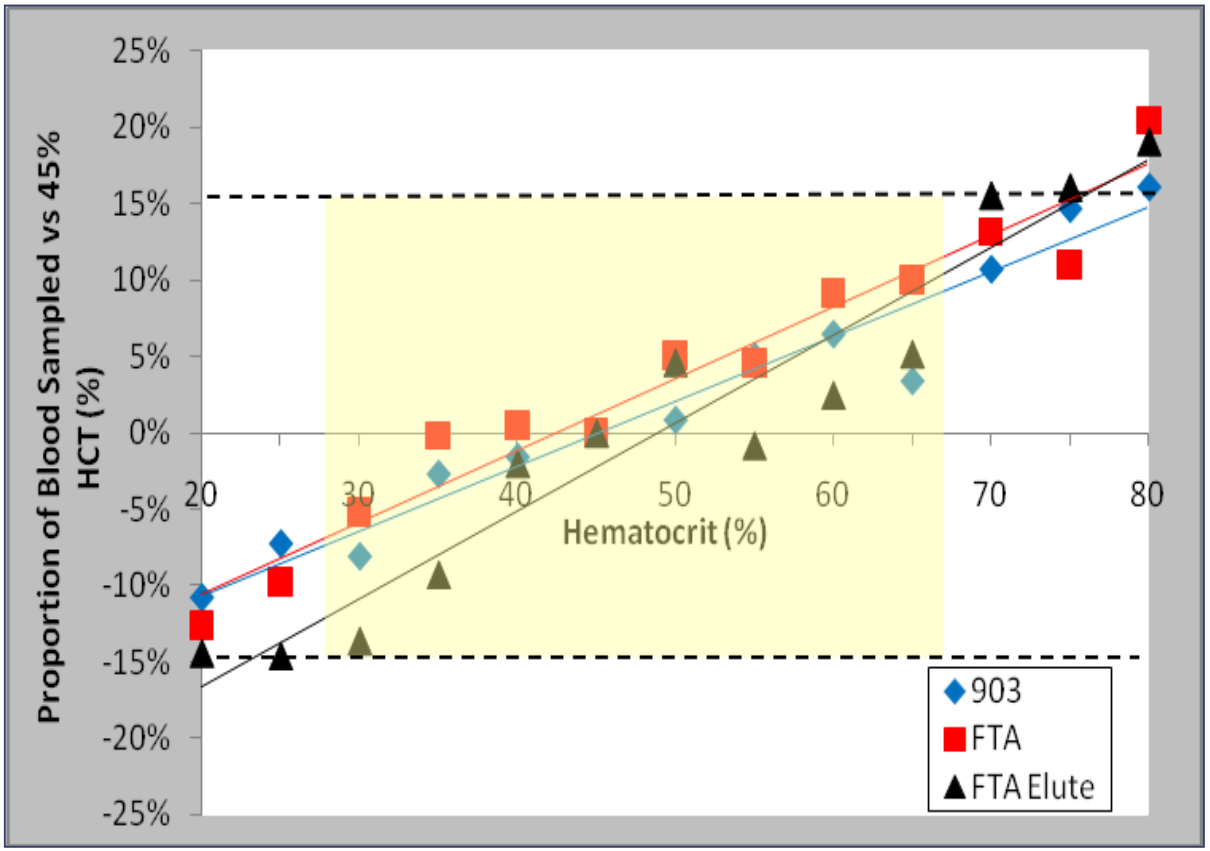
Barfield *et al* (2008) *J. Chrom. B* **870**, 32-37; Spooner *et al* (2009) *Anal. Chem.* **81**, 1557-1563; Spooner *et al* (2010) *Bioanalysis* **2(8)** 1515-1522; Pandya *et al* (2011) *Bioanalysis* **3(7)** 779-786; Stokes *et al* (2011) *Lab. Animals* **45**, 109-113;

However!!!

Blood hematocrit affects the size of the derived blood spot



Leading to a bias in the quantitative data!

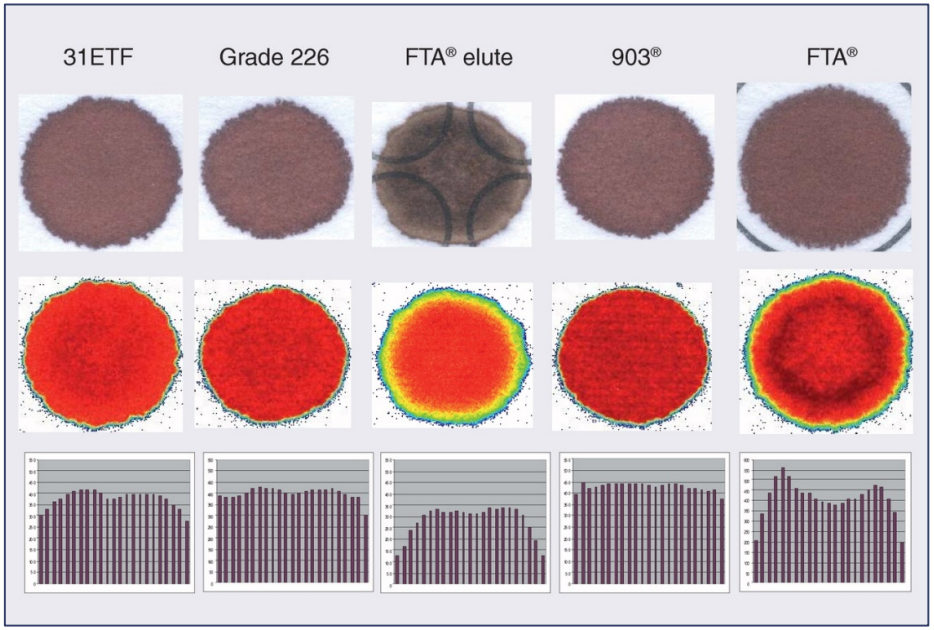


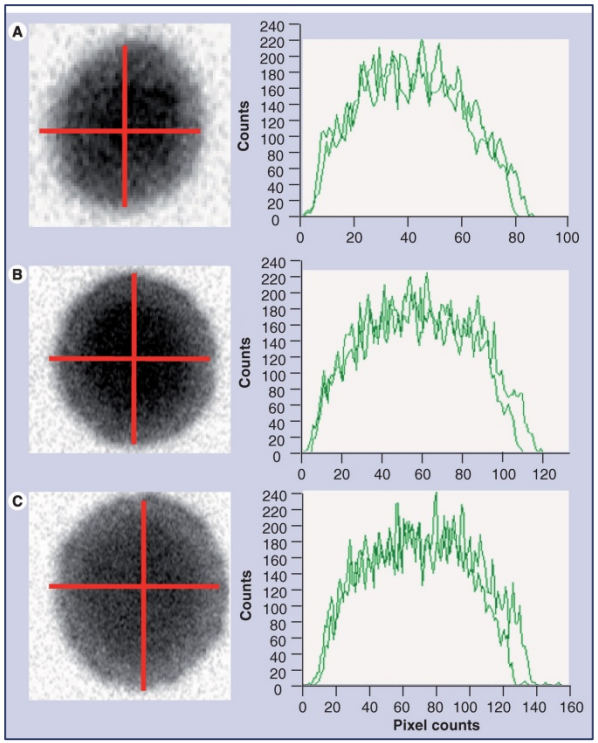
Fixed diameter disc collected from spot with varying HCT

- All data normalized to 45% HCT

Denniff & Spooner (2010) *Bioanalysis* **2(8)** 1385-1395

Spot homogeneity





Example radio histograms of the (A) 15-, (B) 30- and (C) 45-µl blood spots spiked with ^{14}C radiolabeled UK-414495

Ren *et al,* (2010) *Bioanalysis* **2(8)** 1469-1475; Clark *et al* (2010*) Bioanalysis* **2(8)** 1477-1488

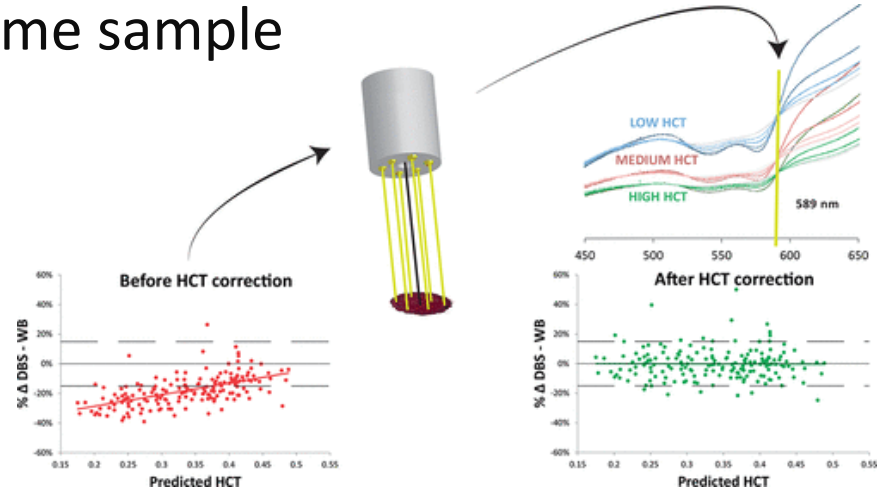
Resulting in……

Regulators (FDA & EMA) required collection & analysis of both wet and dry samples and demonstration of concordance in healthy volunteers and patient groups

Denniff & Spooner (2010) *Bioanalysis* **2(8)** 1385-1395; O'Mara *et al* (2011) *Bioanalysis* **3(20)** 2335-2347; de Vries *et al* (2013) *Bioanalysis* **5(17)** 2147-2160; Cobb *et al* (2013) *Bioanalysis* **5(17)** 2161-2169; Evans *et al* (2015) *AAPS J.* **17(2)** 292-300; Kothare *et al* (2016) *AAPS J.* **18(2)** 519–527

Ways forward for DBS sampling

- Closely match HCT of analytical calibrants & QC's to that of the clinical study samples
- Normalise data to another readily measured component of the same sample



- Collect accurate sample volume & analyse entire sample

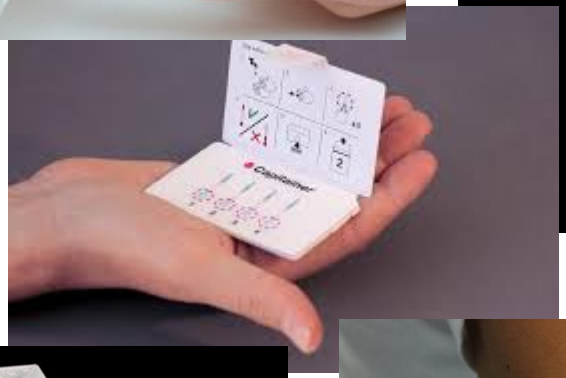
Capiau *et al* (2018) *Anal. Chem.* **90(3)** 1795-1804; Velghe *et al* (2019) *J. Pharm. Biomed. Anal.* **163,** 188–196

Collect accurate sample volume & analyse entire sample

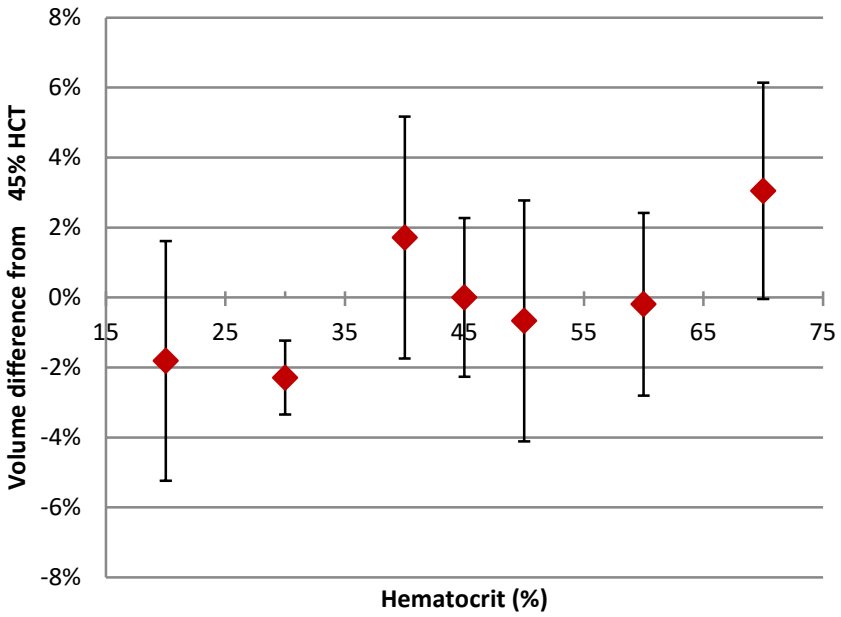
For quantitative analysis

- Technologies required that overcome the issues associated with
  - Blood hematocrit
  - Sample homogeneity
- Whilst delivering the benefits
  - Collecting smaller blood volumes (where appropriate)
  - Facilitating self/assisted sampling
  - Delivering cost savings through home sampling & room temperature sample shipments
  - Integrating with systems for sample shipping, tracking & analysis

A number of novel approaches are now commercially available



HCT independent volumetric sampling performance



- Human blood at different HCTs was spiked with ^{14}C caffeine
- Tip oxidised to CO_2

Denniff & Spooner (2014) *Anal. Chem.* **86**, 8489-8495, Denniff *et al* (2015) *J. Pharm. Biomed. Anal.* **108**, 61-69, Spooner *et al* (2015) *Bioanalysis* **7(6)** 653-659

This is NOT JUST about collecting the samples and data we do today!

Pediatrics

Critically ill

Remote areas

Additional data
- PK
- Biomarkers
- Compliance
- Therapeutic drug monitoring
- Medical event – migraine
- Longitudinal

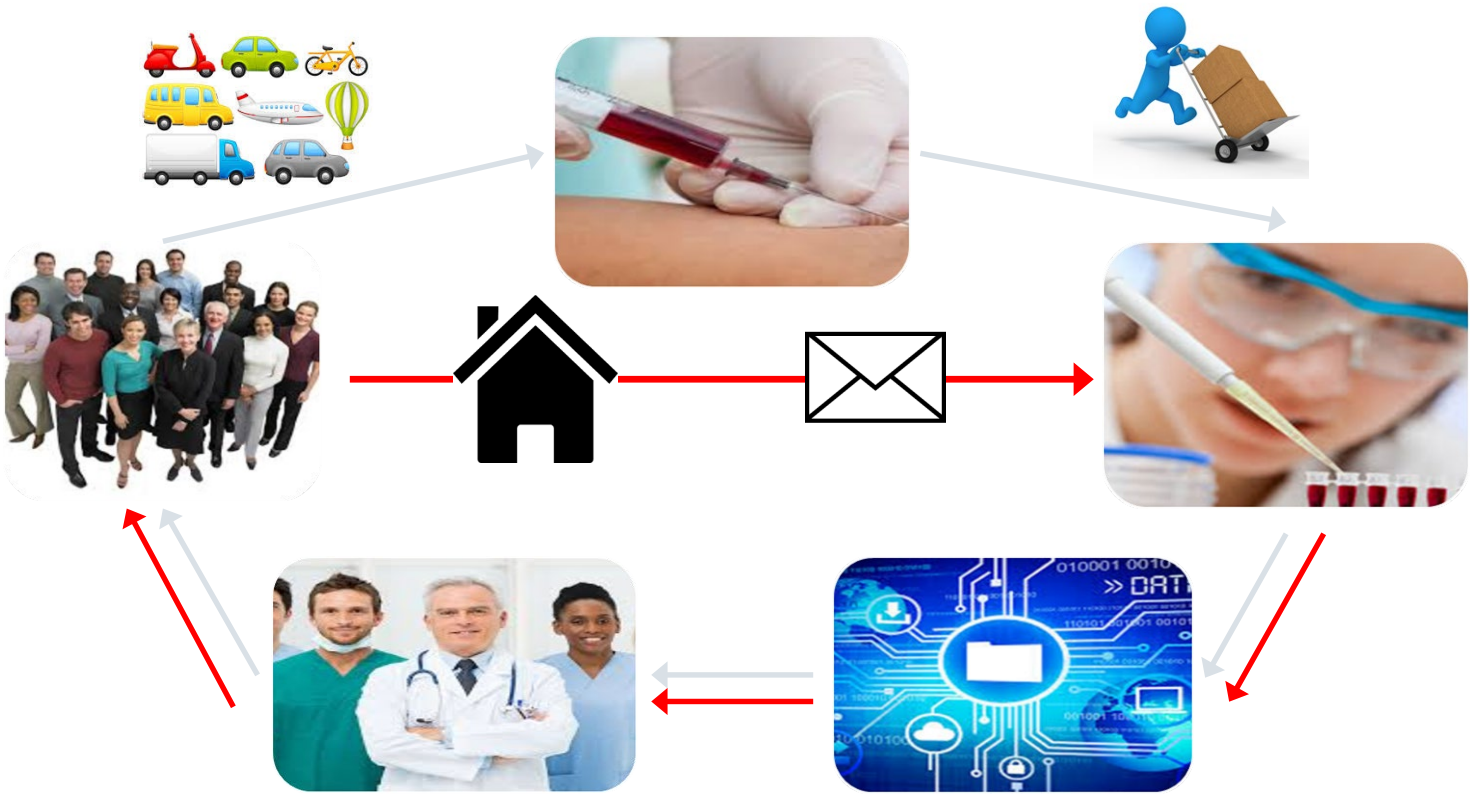
Convenience

Improved clinical trial recruitment & retention

Patient / Consumer driven healthcare

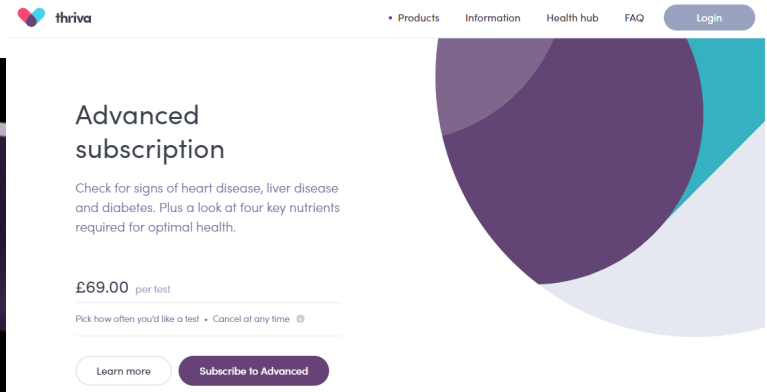
Leading to a new way of working? Current

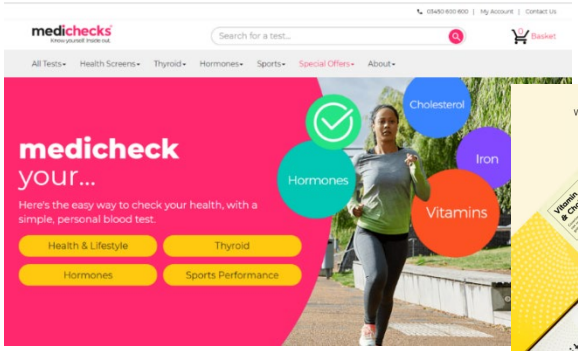
Future?

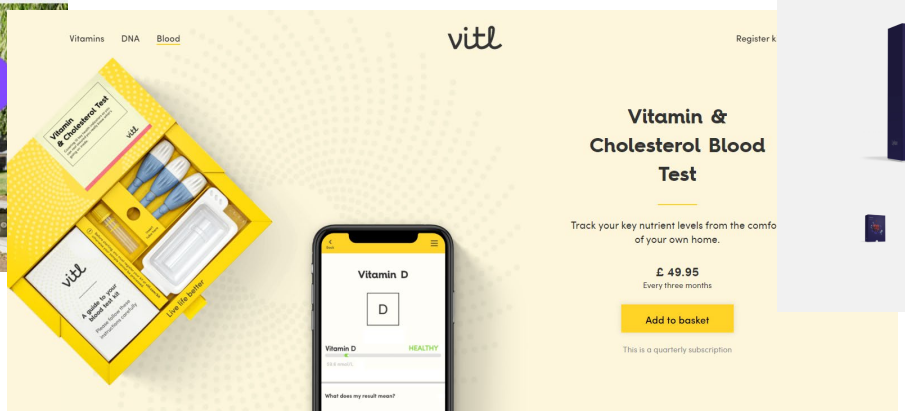


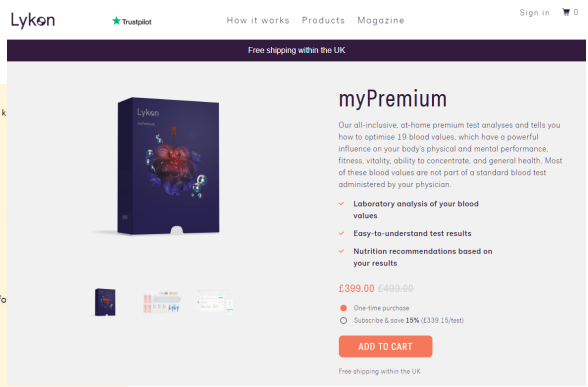
More often, for more analytes…..

Is the future already here? Consumer-led healthcare









Potential challenges……

Is the sample representative of the whole?

Non-conventional sample format
- Sample transfer
- Storage
- Analysis
- Automation

Bridging to existing data
- Blood / Plasma
- Venous / Capillary
- Wet / Dry

Additional assay validation steps

Data acceptability
- Regulators

Reluctance to change!!!

Regulatory Landscape

Non-Clinical

- ICH Q&A on Microsampling as part of ICH S3A Guideline (Nov 2017)
- Also See
  - Beharry (2010) *Bioanalysis* **2(8)**, 1363–1364
  - Viswanathan (2012) *Bioanalysis* **4(12)**, 1417–1419

Clinical

- FDA guidance provided in latest BMV document (May 2018)
- Draft ICH M10 Guidance on BMV (Feb 2019)
- Also see
  - Evans, *et al* (2014) *The AAPS Journal* **17(2)**, 292-300
  - Kothare, *et al* (2016) *The AAPS Journal* **18(2)**, 519–527

Further details on BMV will be given later in this course

Change is difficult!

It depends on how you look at it!

Working together to get “stuff” done

Patient Centric Sampling Interest Group

>90 members

>50 different organisations

- CROs, biotech, pharma, device innovators, instrument vendors, consumable vendors, consultancies, etc

Collaborate in non-competitive areas of interest

- Standardisation
  - Working with CLSI to build an industry standard
- Broad acceptance of patient centric sampling technologies and pathways to their implementation
  - Public, Scientific community, Medical community, Legislators, Payers, Regulators, Media, etc…….
  - Building external facing website

Other Forums for working together

CPSA EUROPE

USA CPSA

CPSA ANALYTICS

aaps®
American Association of Pharmaceutical Scientists

IQ INTERNATIONAL CONSORTIUM for INNOVATION & QUALITY in PHARMACEUTICAL DEVELOPMENT

EBF

Conclusions

New blood sampling & analytical technologies are emerging

Enables us to put the patient first & facilitates human wellbeing

Enables us to collect data that has previously been difficult / impossible to obtain

It's about so much more than the devices

Working across boundaries will enable the change to happen

Change will not be easy & there will be surprises

22nd Annual Symposium
Clinical & Pharmaceutical Solutions through Analysis

Microsampling Workshop

Considerations for bioanalytical assay method development/validation and clinical implementation strategies for drug candidates

Enaksha Wickremsinhe & Tim Olah

Selecting a Microsampling technique

- **Multiple techniques**: enabling collection of reduced blood volumes
- **Stage of assay implementation**: Discovery or Development
- Discovery: minimal validation, fit for purpose
- Development: must meet BMV guidance (EMA 2011, FDA 2018, ICH M10, etc)
- What is different when compared to routine bioanalytical methods
  - **Dried** sample (blood or plasma) vs **wet** sample (blood or plasma)
  - **Sample volume**: capability to handle small samples/volumes
  - **Collection device/format**: selection, ease of use, cost
  - **Source** of blood: IV draw, finger-stick, subcutaneous

Bioanalytical challenges for microsampling

- **Preparation of Standard Curves and QCs**
- **Assay sensitivity:** can you achieve the required LLOQ?
- **Additional validation experiments:** depends on technique employed
- **Account for stability during collection/transit/storage**
  - temperature, humidity, drying time, shipping conditions, etc.
- **Addition of Internal Standard:** in extraction solvent or on pre-dried device?
- **More time and effort required in BioAnalytical lab**
  - Samples are **not** in 96-well format, AUTOMATION not currently possible
- **Sample storage and related logistics**: physical change or analyte degradation
- **Overall BioAnalytical cost higher than current practices?**

Other Bioanalytical challenges

which may require unique experiments or assessments

- Cost to perform cross validation
- Cost of “novel” devices needed for method dev and validation
- Impact of mismatched data sets (poor correlation?)
- Understanding assay efficiencies, operational errors?
- Impact of shipping, storage, and handling temperatures
- Assess homogeneity of samples (especially DBS)
- Multiple “aliquots” of microsamples

Considerations for Clinical Implementation

1. *In Vitro* data: B:P ratio and Fu (protein binding) should not be conc dependent. No Hct effect over clinically relevant range

2. BioA feasibility: LLOQ, Stability, Hct, Homogeneity, etc.

3. Establishing concordance (Bridging): relationship between microsampling conc data and traditional sample (plasma/serum) concentrations data (such that PK conclusions can be drawn across studies)

Is Microsampling data = IV blood/plasma/serum data?

Typically, the biological matrix is either liquid plasma (SM) or serum (LBA)

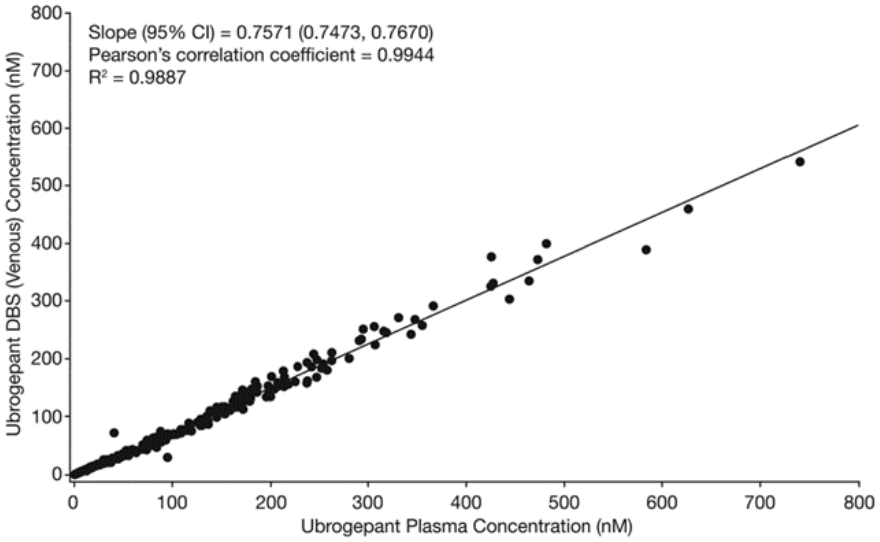
- Microsampling introduces additional matrices
  - Plasma collected in capillaries
  - Liquid blood
  - Blood diluted in water
  - Dried blood (DBS, VAMS, etc.)
  - Dried plasma
  - Other dried matrices: urine, CSF, etc.

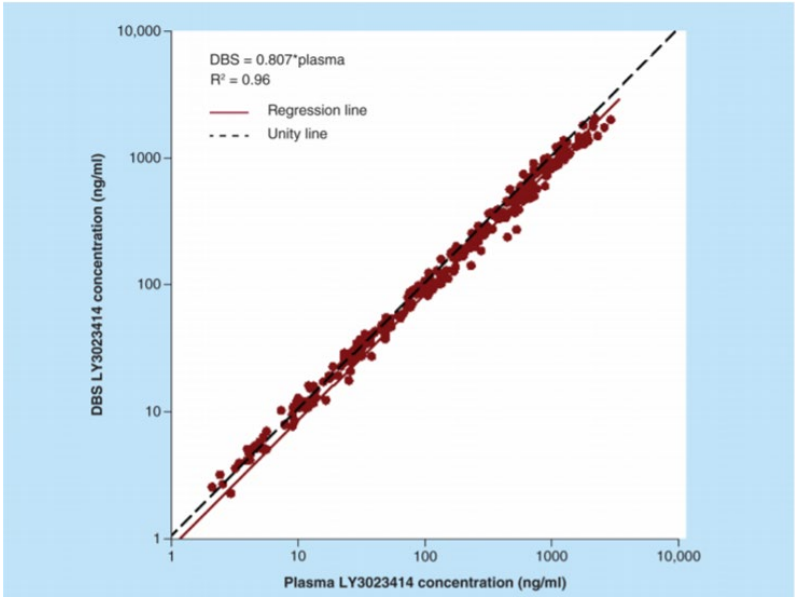
- Also provides alternative sampling sites (compared to IV draw)
  - Finger stick
  - Sub-cutaneous (Tasso, TAP etc)
  - Arterial blood: via umbilical catheter in neonates

Establish concordance (cross validation) with traditional method (plasma/serum)

- 2018 FDA BMV provides guidance on demonstrating correlation between the microsampling method and traditional method
  - **Wet vs Dry**
  - **Plasma vs Blood**
  - **Venous blood/plasma vs finger stick blood/plasma**

- Use incurred samples (n $\geq$ 20)?
- Blood from Healthy volunteers? Patients?
- Acceptance criteria?
- Seek feedback from Regulatory agency (FDA).





Li, Bateman, Kothare, et al, 2018. J Clin Pharmacol, Vol. 58, No. 3, 294-303

Wickremsinhe et al., Bioanalysis (2018) 10(5), 341–356

How do you decide: to microsample or not!

It's not a panacea: Just another tool in the BioA tool box.

Make sure it's the right one for the right study

Needs input from the following:

- Medical
- Clinical operations
- PK
- ADME/DMPK
- BioAnalytical

Discussion Questions

22nd Annual Symposium
Clinical & Pharmaceutical Solutions through Analysis

Break

Monday 28th October 2019, 12:00-16:00 University Grille

22nd Annual Symposium
Clinical & Pharmaceutical Solutions through Analysis

Clinical Operations

How to implement patient centric sampling in clinical trials

What is the need?
What question are we answering?
Can micro sampling meet this need?

- Program/Institutional Needs
  - Matrix requirements
  - At home sampling benefits
  - Specific indications – pediatrics, migraine, cancer, therapeutic drug monitoring

The technology is disruptive to existing workflows across the organization – the need must be great

Logistical

- Training Clinical site and Patient – this can involve several clinical sites all over the world and require language translation.
- Sample Integrity/Quality – how do we ensure the right person gets sampled at the right time in the correct way?
- Environmental Exposure in the wild
- Technology access for use in remote/underserved geographies if using an eDiary/App based data collection approach is used.
- Supply – scaling up manufacturing for device availability, lot-to-lot variability.
  - How do you track various lots across large clinical studies, make standards and QCs with matching lots, etc.?
- Shipping requirements within a country and country to country?

Logistical

- How do we reliably collect a time stamp and how will the data flow?
  - No preassigned barcodes for sample collection, new process needs to be developed and proven to work.
  - How do you match the concentration values with the date and time of collection?
  - What about time zones?
- Patient population may not be appropriate for use of invasive technologies (i.e. shipping HIV infected blood).
- Patient compliance and sample collection reliability, at home sampling needs to be as simple and straightforward as possible.

- Records time and temperature every 10 min for 2 weeks
- Starts when button is pressed
- Wireless communication with smartphone or smartbox (to be design in partnership with Merck)

Business/Regulatory Related

- "If it can't be used at 100% of sites, it can't be used at all" attitude.
- Increases the cost of conducting the trial.
- Requires bridging from liquid plasma to dried blood.
- Increases the complexity of the protocol for the trial and this will impact enrollment.
- No definitive data that shows return on investment for Patient-Centric Sampling.
- How are devices treated and what regulatory approval is needed in each country?
- How do we engage with regulators to minimize issues during filing?
- How do you show the sample is from the person enrolled in the trial?
- Can you define inclusion/exclusion criteria using adherence data from at home sampling? What about intent to treat criteria?
    - Potential for un-blinding when using these approaches

Program Needs

Data Enrichment at Home Capability – late stage, Migraine, compliance

Bioanalytical Feasibility

Sensitivity, Selectivity, Stability, Automation

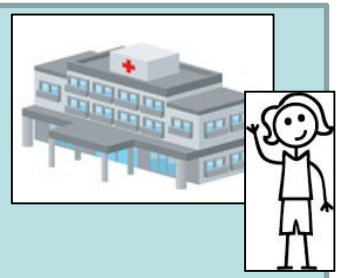
Validation on Device

Supplies

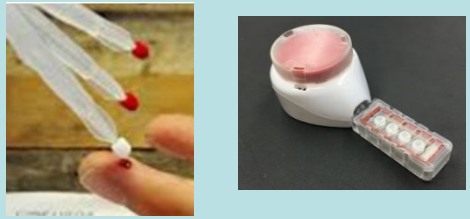
Storage, Shipping, eDiary

Training Materials

Operations Manual, Site Materials, Patient Handout, Video

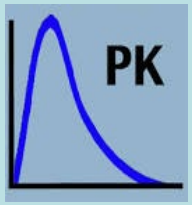


Training: Site and Subject



Smart Sampling and Management

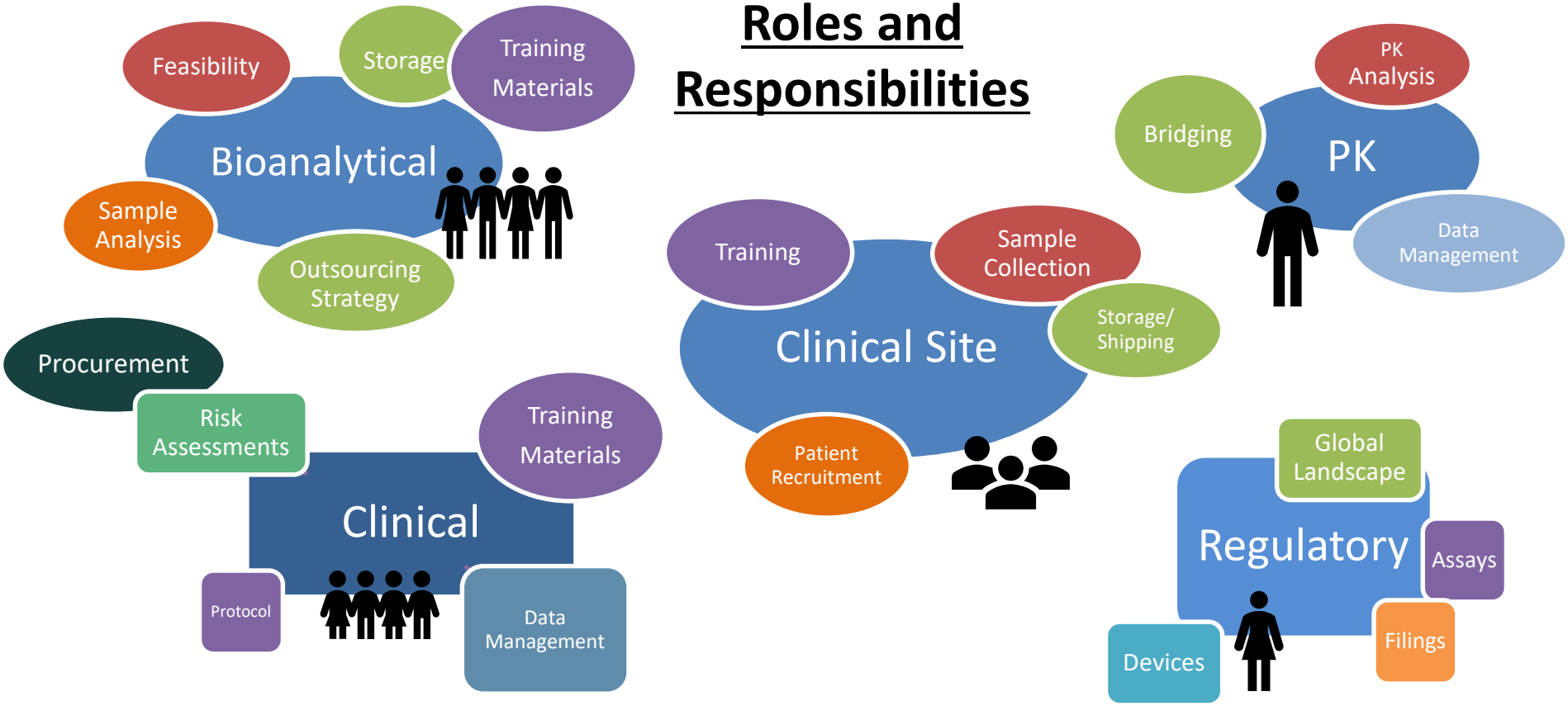
Home based microsampling for PK, Labeling, time stamps



PK Analysis

Microsampling for PK

Roles and Responsibilities

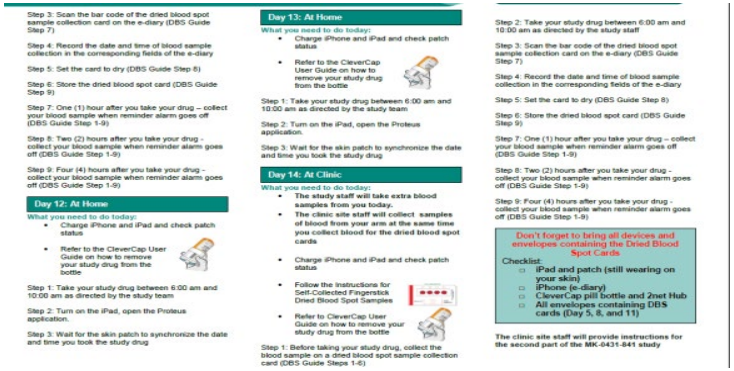


Supplies/Kits

Patient Supply Kit

- User guide
- Alcohol wipe
- Lancet
- Sterile gauze pad
- Band-Aid®
- VAMS tip clamshell
- Foil Envelope
- Desiccant
- Mailing Envelope

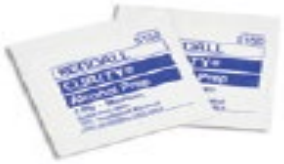
User guide



Collection Kits

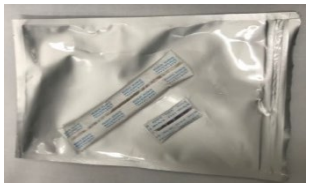
Lancet

Alcohol wipe



VAMS Tip Clamshell

Foil Envelope and Desiccant



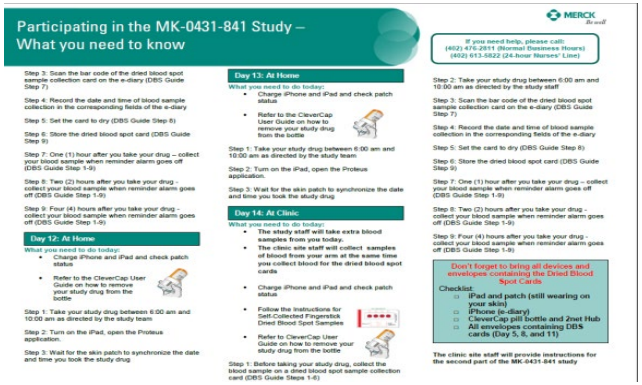
Band-Aid®

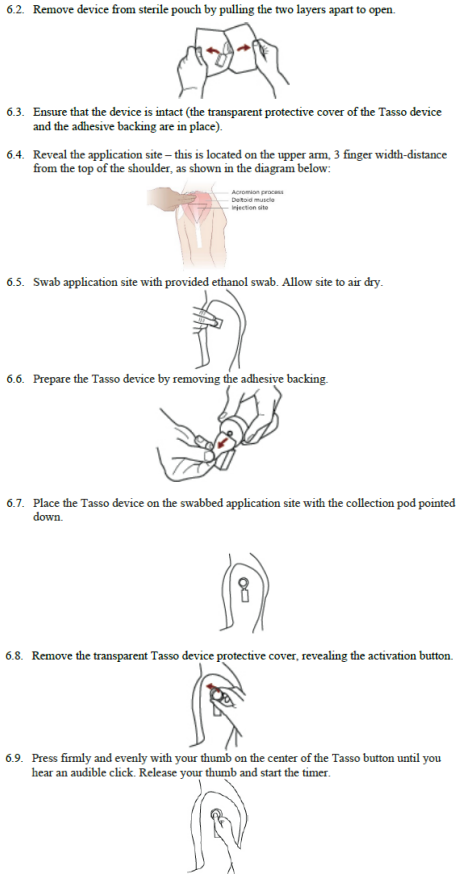
Sterile gauze pad

Mailing Envelope

Training Materials

- Brochures for Patients
- Brochures for Clinical Staff
- Study Operations Manual
- Videos
- Investigator Meeting Presentations





Future Directions

Kevin Bateman & Neil Spooner

What do we need to do in order to progress?

- Technology
  - Sampler features
    - Ease of use – end user vs analytical scientist
    - Sample volume
    - Sample format
      - Wet, dry, plasma, serum, blood
      - Single vs replicates
    - Sample processing at point of collection
    - Time, date & location
    - Traceability
    - Recycle / re-use
  - Analytical workflows
    - Centralised vs decentralised
    - Skilled analysts vs push button
    - Compatibility / integration with analytical methods
  - Standardisation

What do we need to do in order to progress?

- Implementation
  - Organisational change
    - Within group
    - Between groups
  - Regulatory acceptance
  - Acceptance by society
  - Training
    - Patients
    - Clinicians
    - Analytical Scientists

Where else can we use these approaches?

- Large molecule bioanalysis
  - LBA and/or LC-MS approaches
- Vaccine research
  - Epidemiological studies
  - Track response/protection over time
- Non-drug analytes
  - Dynamics of disease signatures (RNA)
  - Longitudinal studies of human health

22nd Annual Symposium
Clinical & Pharmaceutical Solutions through Analysis

Wrap-up and Next Steps

Monday 28th October 2019, 12:00-16:00 University Grille