Processed extract stability

Presenter: Amanda Wilson
on behalf of EBF

Focus Workshop

(*In collaboration with the AAPS and JBF*)

Industry input into ICH M10: Experimental data as the cornerstone for a science driven bioanalytical guideline

The Altis Grand Hotel Lisbon,
Portugal September 24-26, 2017

Problem statement: *Processed sample stability*

- Industry unclear on requirements
- To address health authority questions
  - Regulatory expected approach does not represent typical sample analysis procedures
- Experimental inconsistency across organisations
- Clarify nomenclature & experimental specifics

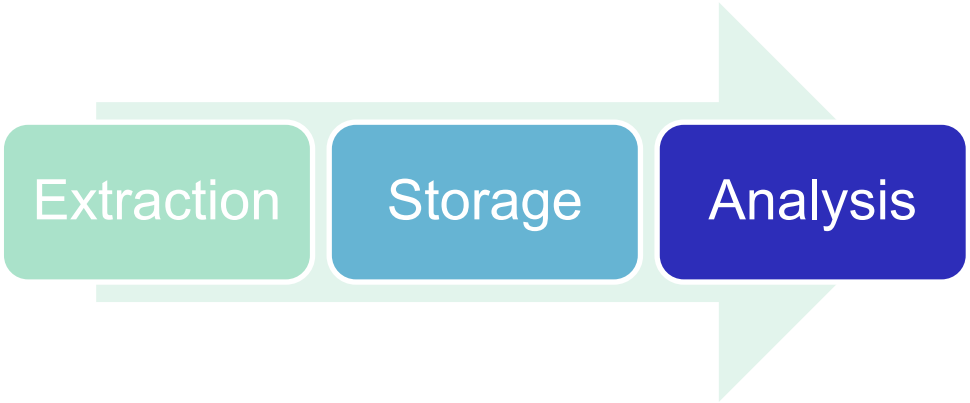
- Processed sample stability
- On extracts to be stored prior to analysis or after first injection

Content

- Industry & regulatory landscape
- European industry survey data – what does this tell us
- Recommendation

What is required?

- Understand the integrity of an analyte that has been extracted from a biological matrix for the duration of its storage following extraction to the time of being analysed



Limited specific experimental instruction within present guidance

- FDA 2001 post preparative stability.
  - processed samples, including the resident time in the auto-sampler (draft FDA 2013)
  - drug & internal standard assessed over the anticipated run time for the batch size
  - determining concentration on the basis of original calibration standards
- EMA 2012. The following tests should be performed if applicable:
  - Stability of the processed sample at room temperate or under the storage conditions to be used during the study (dry extract or in the injection phase).
  - On-instrument / auto-sampler stability of the processed sample at injector or auto-sampler temperature
- MHLW.
  - stability in the processed samples
  - All stability experiments should be performed on samples that have been stored for a time that is longer than the actual storage period

Addressing the absence of clear specific experimental instruction

- White papers e.g. Crystal City V workshop
- Industry reports on Health authority observations
- Stored extract stability requires extracts are stored for a period of time prior to injection with freshly prepared calibration sample extracts.

- Not representative of typical sample analysis procedures adopted in the lab
- Scientific value is unclear

Industry survey – what did it tell us about the most popular approaches

- Process sample stability by re-injecting a stored full A&P run
  - Typically in the autosampler
  - Part of method validation
  - A&P criteria used for assessment
- Significant number of companies perform combined stored / fresh extract stability assessments
  - Concerns of regulatory citation
  - *Less than half of companies thought this was a relevant experiment*

Industry survey – what did it tell us
Processed sample stability

- Failure rate <5%

- Most common causes for failure
  - Non specific binding
  - Evaporation of extract / insufficient volume remaining for re-injection
  - Degradation of analyte

EMA guidance

- All samples (calibration standards, QC samples and study samples) should be processed and extracted as one single batch of samples in the order in which intend to be submitted or analysed
- Suggests
  - Stored extracts vs fresh calibrators should not be performed
- Risk of introducing non-compliant behaviours of adding samples to an existing batch

Processed sample stability
What is the Purpose – 2 fold

- Ability to inject or re-inject a whole or partial run
- How long you can wait until you inject your samples post extraction

2 simple nomencletures

- Processed sample stability
- Re-injection reproducibility

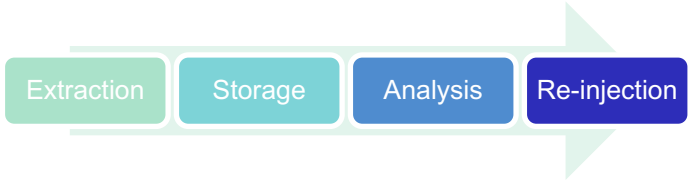
Processed sample stability

- Stability of the sample throughout the sample preparation process starting from the thawing of fresh sample and ending when the last sample of the batch is injected
- Document as part of validation
- Mimic the maximum batch size
- Prepare the run, store and analyse

Re-injection reproducibility

- Allows the possibility to re-inject a partial or full run following technical or chromatographic / Mass spec issue
- Document as part of validation
- Re-inject a prior injected validation run
- Calibration samples and at least one set of QCs (H/M/L)
- Assess accuracy and precision
- Note the timing of the re-injection to mimic anticipated real time situation

Summary = key messages



- Assessment of extract storage should mirror the conditions under which samples are stored prior to or during analysis
  - Use of fresh calibrants to assess extract stability is not considered reflective of typical situation with real samples
- Relevant experiments should be scientifically justified
- 2 experiments in method validation
  - Processed sample stability
  - Re-injection reproducibility

Acknowledgement

- EBF community
- Topic Team 41 members
  - Suzanne Barker
  - Ludovicus Staelens
  - Sirpa Laakso
  - Achim Freisleben
  - Steve White


Thank you


Questions
Answers