Biomarker (BMK) Sub-Group 2017 Report

Kitzbühel, Austria
October 10, 2017

Coordinator: G. L. Prasad
Secretary: Kirk Newland
Scientific Commission Liaison: Paul Harp
Objective 1: To review present knowledge of tobacco and smoking-related biomarkers of exposure and effect, and to document these in meeting minutes, CORESTA reports and scientific publications where appropriate

Objective 2: To undertake ring trials / proficiency tests for selected biomarkers as agreed by SC

Objective 3: To source and develop reference materials to support biomarker analysis for those biomarkers selected for ring trials / proficiency tests
October 08, 2016 Berlin, Germany
- 30 delegates attended

April 19, 2017 Orlando, FL USA
- 25 delegates attended

October 08, 2017, Kitzbühel, Austria
- 38 delegates attended

The Biomarker SG holds joint meetings with the Product Use Behavior Group
- BMK SG meets in the afternoon
**Biomarker**

- Generally refers to a measurable *indicator* of some biological state or condition (Wikipedia)
- Context-dependent
- Interim measures
- Decision making tools
- Biomarkers typically have two dimensions
  - The biology: What is that we are trying to measure and under what context?
  - Measurement: Analytical component
Two types of biomarkers

Broadly two types of Biomarkers exist in the context of Tobacco

- **Biomarkers of Exposure**: Measures of exposure to tobacco products. Examples include: nicotine, cotinine, TSNAs
- **Biomarkers of Effect**: Indicate the effect of tobacco use on the consumer.
  - Could be useful for assessing the health effects of the product and could inform the potential harm
  - Particularly useful in the absence of epidemiology
  - Very diverse in their characteristics: examples; a compound, a protein or its activity, a gene
Diverse topics for discussions include, analytical methods to cutting edge science

Methods discussion:

- **Flavoromics**, profiling for flavors in e-liquids and body fluids (ABF)
  - Library of flavor compounds, untargeted analysis

**Biomarkers**

- **Biomarkers in Clinical Trials (Inflamax)**
  - Focus on respiratory biomarkers, established (FEV1) and emerging (nasal mucociliary clearance)
Biomarkers

- Methodological considerations for identifying likely users of e-vapor products for ambulatory clinical studies (Altria)
  - Reduced cigarette use
  - Subject compliance and reporting issues noted in this fairly large clinical study

- Biomarkers for Cigars - Oral, inhalational, or a combination (Imperial Brands)
  - Fewer biomarkers/HPHCs in cigar smoke, lower levels in consumers relative to cigarette smokers, and need for additional work
Scientific Discussions

Biomarkers

- Biomarkers for tobacco products - proposal for a status review (Inflamax)
  - Writing committee formed: Victoria Nelson, Cherrie Small, Piyush Patel, Patrudu Makena, Michael McEwan, Krishna Prasad, Elizabeth Cerson, Michael Kong, Jeff Edmiston and G. L. Prasad

- Biomarkers of Effect review, with a focus on lung biomarkers (NWIP# BMK161)
  - Scope may be refined further, and publication costs resolved
Biomarkers

Biomarkers of effect for tobacco products (Altasciences)
  - Need for qualified biomarkers for tobacco studies
  - Two likely biomarkers-DNA adducts and non-coding RNAs discussed
  - Ensuing discussion led to exploration of a meta analysis of existing biomarker data, a potential NWIP

Biomarkers of effect in smokers who switch to electronic cigarettes (ITG)
  - Biomarkers of effect in a 2 year product switching study
  - Metabolomic data presented
Biomarkers

- Proteomic and lipidomic markers from an *in vivo* study involving THS2.2 product (PMI)
  - Distinct protein and lipid biomarkers from smoke exposed mice and cessation and product-exposed animals
Data

- **Data standards for clinical biomarkers of exposure (Celerion / BAT?)**
  - To standardize and streamline data management and reporting
  - Discussions underway to develop a new project

- **INTERVALS, a platform for transparent data sharing (PMI)**
  - Presentation on data transparency; opportunity for sharing and comparing
  - Role for/ involvement of CORESTA BMK SG?
  - To be discussed further

- **Meta analysis of biomarker data**
  - A likely NWIP (from discussion on biomarkers of effect)
  - More discussion needed
Reference standards

- Requirements for the Certification of analytical reference standards in tobacco biomarker studies (Objective 3)
  - CORESTA Technical Guide No. 20, published – September 2017
  - Guideline describes the desired content of Certificate of Analysis for reference standards
  - Writing Committee:
    - Frank Deschamps (Leader), Max Scherer, Mark Bentley, Krishna Prasad, Eckhardt Schmidt and G. L. Prasad
Requirements for the Certification of analytical reference standards in tobacco biomarker studies

- To have well characterized analytical standards so that quality data can be generated
- Analytical standards should be well characterized
  - Prove the identity
  - Purity of the analyte
  - Potency of the analyte

Next steps

- Development guidelines for standards for fit-for-purpose bioanalysis, a potential NWIP
Objective 2: Inter-laboratory Comparison: Bioanalytical Assay to measure total NNAL in human urine

- What is NNAL?
- Why NNAL?
NNAL is a major metabolite of the tobacco-specific nitrosamine NNK

- NNK is a known lung carcinogen (IARC Group 1 carcinogen) and is designated as HPHC
- NNAL (4-(methylnitrosamino)-1-(3-pyridyl)-1-butanol) is also considered as a carcinogen.
- NNAL is enzymatically conjugated as a glucuronide and excreted in urine
- Urinary levels of total NNAL (free + glucuronidated NNAL) are widely used as a biomarker of exposure to tobacco products
Several laboratories showed interest

- ABF GmbH
- SEITA – Imperial Tobacco
- China National Tobacco Quality
- Shanghai Tobacco Group
- Zhengzhou Tobacco Research Institute of CNTC
- KT&G Research Institute
- University of Minnesota
- Covance Laboratories
- Celerion, Inc., Organizing lab
Sponsors

- Altria Client Services
- British American Tobacco
- Imperial Tobacco Ltd
- Japan Tobacco
- Philip Morris International
- RAI Services Company

The cost associated with the study were shared by the sponsors.
Study Results

- Final data was received from 4 participating laboratories
  - ABF, SIETA, U Minnesota and Celerion

- Final data was not available from 5 of the 9 laboratories due to issues of:
  - Labs unable to receive biological samples from Celerion
  - Bioanalytical assay not available
  - Testing not completed/ Results were not provided
Study design

- Shared Quality Controls at 3 concentrations
  - 15.0 pg/mL (n=6)
  - 70.0 pg/mL (n=6)
  - 750 pg/mL (n=6)

- Individual Lots of Smoker Urine from 9 Volunteers (n=3)

- NIST Smoker Urine (n=3)

- Control Blank Matrix (n=3)

- 1 Set of Imbedded Standards
  - 9 Standard Concentrations
  - Range: 5.00 to 1000 pg/mL

- Original Participating Labs: 9

- Labs Completing Analysis: 4
When each laboratory used their own source of reference material, the bioanalytical results were not comparable within the standard bioanalytical acceptances (>30% R.E).

When the same reference material and standard set was used for quantitation, all 4 labs produced data within 15% of the expected concentrations.
With the 4 laboratories evaluated no analytical method issues were noted. The same variability was observed for aglycone and total NNAL samples.

The use of a single set of calibration standards resolved the bias observed for both aglycone and total NNAL samples.

Final Report anticipated Jan 2018
The inter-laboratory comparison study for the urinary acrolein biomarker, 3-HPMA was published.
Thank You

Berlin meeting