Table 6: Integration of Electropherograms

Facilitator: Timothy Blanc, *Eli Lilly and Company, Branchburg, New Jersey, United States*

Scribe: Lin Zeng, *Amgen Inc., Thousand Oaks, CA, United States*

Scope:

The desirable throughput of rapid capillary electrophoresis methods can be obliterated if electropherogram integration is manual and exceedingly time-consuming. While CE separations can be rapid, CE baselines are prone to irregularities often resulting in tedious and time-consuming manual integrations. So, CE user are abundantly motivated to use auto-integration to function efficiently and regulators are requiring auto-integration to assure data integrity. But, is Auto-Integration possible for all capillary electrophoresis methods…. 100% of the time?

Questions for Discussion:

Part I: Where We Have Been and Where We Are Now

First, we will go around the table to introduce ourselves and briefly describe the integration practices, problems and solutions (work arounds) we have encountered. We will then discuss topics such as:

1. Working under SOPs requiring auto-integration or other mechanisms to assure objective integration and data integrity.
2. Throughput and sample turnaround challenges due to intensive integration.
3. The impact of "analyst to analyst" integration difference as significant source of variability (as related method validation, method transfer, and/or stability testing).

Part II: Towards a Better Future

Regulatory guidance on data integrity and recent emphasis on auto-integration during inspections.

4. The desire for auto-integration is understandable, but is it faster or more accurate than manual integration of electropherograms from CE methods with irregular baselines? Is software the answer for the future? (e.g. new algorithms, artificial intelligence/machine learning)
5. Is new hardware, like alternate detection systems, the answer for the future? (e.g. moving beyond low UV to Fluorescent derivatization for improved baseline and greater signal leading to significant improvement in integration).

6. Brain-storming and suggestions for the CE Pharm community to work on.

Discussion Notes:

Eleven scientists participated the discussion. Key points were captured as following:

1. Common challenges in electropherograms integration:
    a. Integrate peaks which are below LOD
    b. Obvious baseline shifting, wobble baseline during run
    c. No regulation or standardized practice respect to integration. The way people integrate peaks varies from user to user and company to company.
    d. Integration is different from molecule to molecule, making it is hard to have a specific platform to cover most molecules
2. Common things that govern interaction is done:
    e. Follow validated methods, clear instruction for specific integration parameters is given
    f. Separate work instruction to properly zoom to identify peaks
    g. Use "Custom Field" at Empower to filter out peaks below LOD.
    h. When having baseline shifting issue, perform blank injection before each sample as the background to avoid identify unrelated peaks
3. Methods to help decrease background shift:
    i. Add mineral oil on top of gel buffer to help prevent gel matrix evaporation as the run goes on.
4. What companies do in terms of dropping a peak on a shoulder where not able to see a very clear valley but see some inflection?
    j. Characterize the shoulder
    k. Only include a dropdown when see a clear consistent inflection
    l. Use "Tangential" instead of "Drop down" in Empower. "Tangential" will ensure only the shoulder is captured, not necessary all the areas that beneath it.
5. How to solve the analyst variability problem?
    m. At the training process, specify how to drop the shoulder and line.
6. Future desires for CE software and hardware:
    n. Software advances: automatically compare sample chromatograms with blank chromatogram, simple subtraction
    o. Hardware is updated to improve baseline, making integration easier
    p. Hardware is modified to be able to change laser intensity or correction factor that change the output signals, more flexibility to set the dynamic range
    q. Have UV and fluorescence detector for CE at the same time
    r. Standard integration method to make analysis easier for different molecules