Refresher:
Conventional Flow, Spectral Flow, CyTOF

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Conventional flow cytometry
“one detector, one color”
Spectral Overlap

Our high speed analyzers

3 BD LSRFortessa, 2 BD LSRII, 1 NxT Attune = conventional

3 Cytek Auroras (2 in Unified Core, 1 at HCC) = spectral
Spectral flow cytometry: Cytek Aurora

collect ALL of the light \(\rightarrow\) increased sensitivity

discriminate fluors with overlapping spectra \(\rightarrow\) not possible with conventional flow cytometry

64 channels
Spectral cytometry rivals CyTOF capabilities

**FACS**
Antibodies labelled with *fluorochromes*

Conventional cytometry, 10-15 colors; beyond that gets tough

Spectral technologies = game changer, 64 channels

**CyTOF**
Antibodies labelled with *elemental isotopes*

40-50 parameters in a single tube

Ex. gadolinium (Gd), samarium (Sm), terbium (Tb), thorium (Th), ytterbium (Yb)
Pitt Aurora Flow Panels Repository

• Panels for each mouse and man
•Designed/validated by our Pitt colleagues
•Excel spreadsheet located in same PittBox that contains this ppt

Thanks to our users for sharing their panel designs!
Next Tutorial: Harness the Full Information Content of Your Data with Algorithmic Analysis

- **High-dimensional analysis** – Human brain is not so hot at >3 dimensions.

- **Highlight patterns in the data** – Manual analysis with iterative bivariate plots is highly inefficient.

- **Facilitates population discovery** – Populations can be overlooked b/c biases and a priori knowledge dictate analysis.

- **Fun!**
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