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Late Breaking Abstract - Validation of assays measuring functional alpha-1 antitrypsin (AAT) in patients with AAT deficiency (AATD)

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Introduction: AAT is a potent inhibitor of neutrophil elastase (NE) in the lungs. AATD is associated with low AAT levels and increased lung disease risk. In studies that informed FDA approval of plasma-derived AAT (pdAAT) for treating AATD, observed functional AAT (fAAT) increases in serum and bronchoalveolar lavage fluid (BALF) indicated biochemical efficacy. However, augmentation therapy with pdAAT targets an antigenic (ie, total) rather than a functional serum AAT level. fAAT levels are determined by measuring a sample's anti-NE capacity (ANEC), but reproducibility can be challenging. We developed an ANEC-based method for quantifying fAAT to meet regulatory agency expectations for method validation and trial use.

Methods: fAAT concentrations in human sera and BALF were determined by an enzymatic ANEC assay in which irreversible binding of fAAT to NE inhibits NE-mediated hydrolysis of chromogenic MeOSuc-AAPV-pNA. We evaluated our method using the FDA 2018 and ICH M10 bioanalytical method validation (BMV) guidelines.

Results: Our assay detected ANEC activity of endogenous fAAT and recombinant human AAT INBRX-101 in sera and BALF. Acceptance criteria for BMV parameters were met, including intra- and interassay precision, selectivity/matrix interference, dilution linearity, and sample stability. Dynamic ranges were 14-56 nM (BALF) and 1-5.6 µM (serum). Data on the normal range of serum fAAT in healthy volunteers will be presented.

Conclusions: This validated ANEC assay addresses the unique challenges associated with measuring fAAT in BALF and sera, making it well suited for evaluating biochemical efficacy of novel AATD therapies (eg, INBRX-101) in registrational clinical trials.