

# Increased Throughput, Walk-Away Time and Efficiency with Tecan® Automated CIEF Sample Preparation



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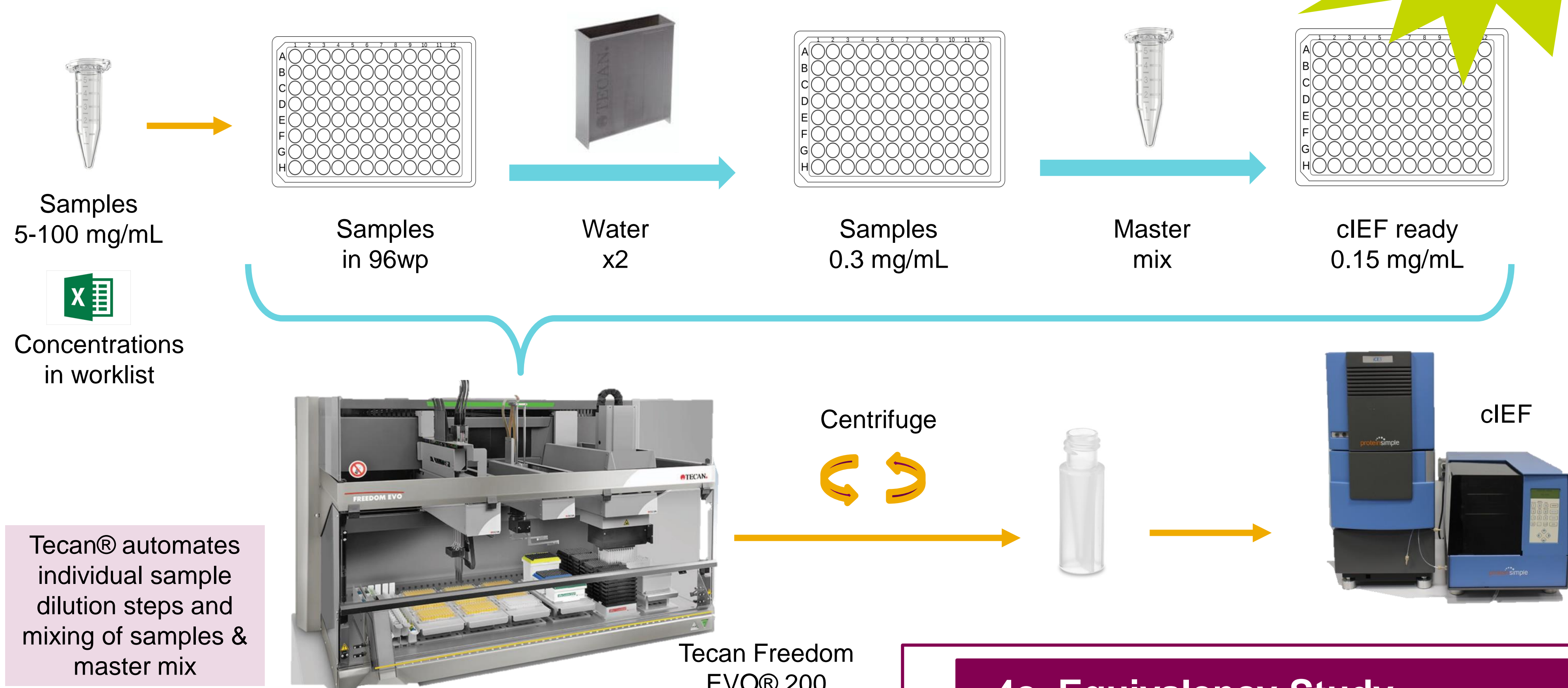
## 1. Overview

- We have developed an **automated sample preparation** method for monoclonal antibody (mAb) capillary isoelectric focusing (cIEF) analysis using **Tecan Freedom EVO® 200**
- The method was designed with **individual sample dilution** control over a **wide range of initial concentrations (5-100 mg/mL)**
- It is capable of dispensing and mixing **water and viscous urea-based solutions** (i.e., master mix)
- This automated method shows good **workflow performance** and **equivalency to manual sample preparation**
- It was successfully applied to mAb formulation development activity with **no repeats** and **3 hours of hands-on time saved**

## 2. Sample Preparation for cIEF

- Capillary isoelectric focusing (cIEF) is the gold standard method for monitoring **charge heterogeneity of biopharmaceuticals**
- The method is routinely used across all stages of biopharmaceutical development which results in **high sample load**
- Manual sample preparation is a multi-step, time-consuming and **labour-intensive** process, with high repeat analysis rates (> 10%)

## 3. Automated cIEF Sample Preparation Workflow



## 4a. Efficiency improvements

- Automated workflow was implemented to test 21 cIEF samples for a mAb formulation development activity.
- Preparation required 1 hour of analyst hands-on time, no repeats were needed
- Tecan® workflow improved efficiency by reducing time and repeats compared to manual preparation

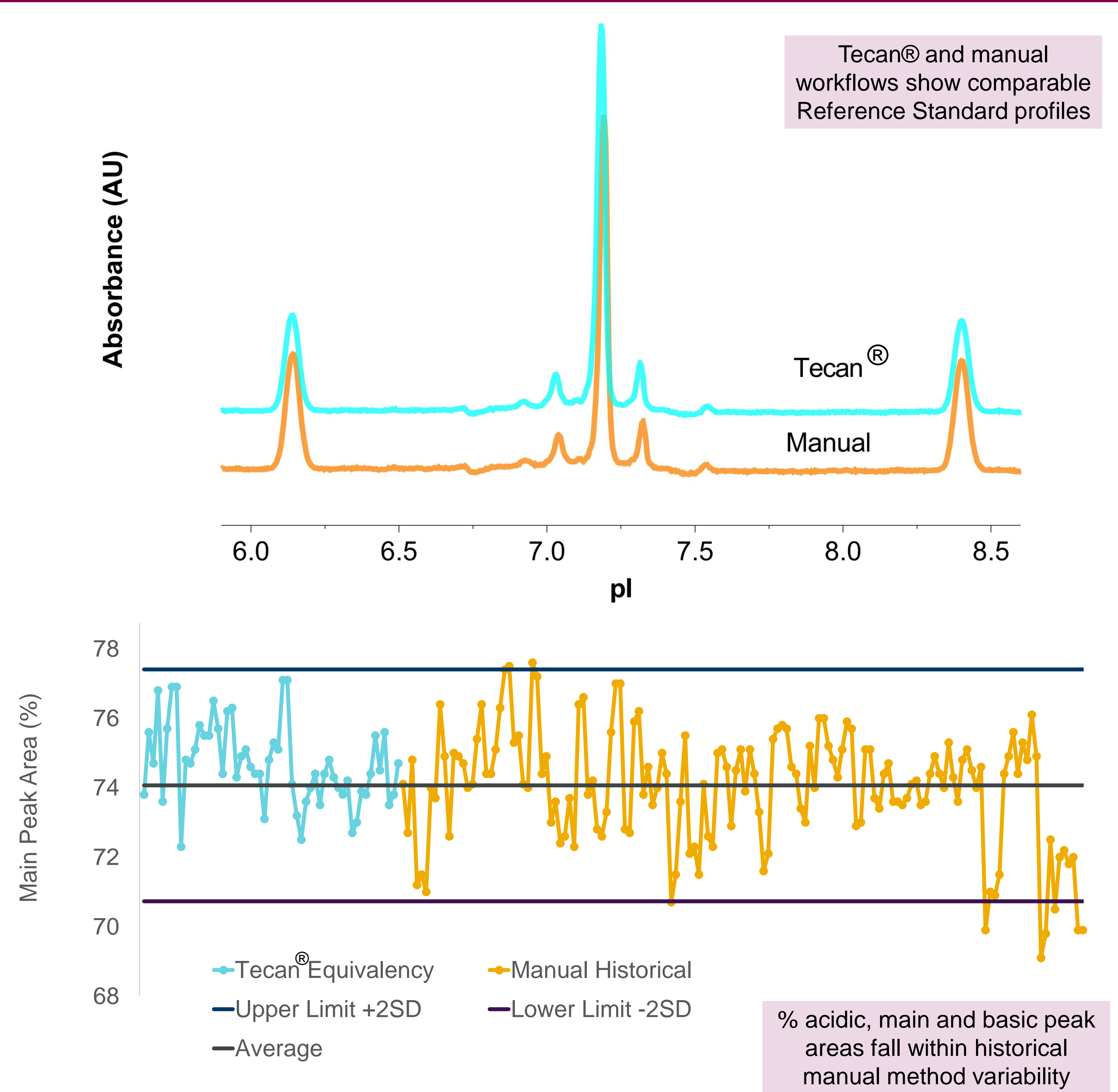
20 samples	Manual	Tecan
Hands-on analyst time	4 hours	1 hours
Repeats	10% (expected)	No repeats

## 4b. Performance Assessment

- Automated method passes performance criteria agreed in Equivalency Plan
- Accurate over the initial concentration range of 5 to 100 mg/mL

		< 7% CV		
		% Acidic	% Main	% Basic
<b>Repeatability</b>				
6 replicates single run	Average	15.3	76.5	8.2
	%CV	2.7	0.7	2.9
	Target	10	10	20
<b>Intermediate Precision</b>				
6 replicates over 2 days	Average	15.6	75.8	8.5
	%CV	3.7	1.3	6.2
	Target	20	20	30
<b>Accuracy Over Initial Concentration Range</b>				
Initial concentrations 5-100 mg/mL (3 replicates each)	Average	18.7	71.6	9.7
	%CV	3.5	1.7	6.6
	Target	10	10	NA

## 4c. Equivalency Study



## 5. Conclusions

- Automated cIEF sample preparation workflow using Tecan® liquid handler shows good performance which compares to a manual workflow
- Implementation of automated workflow for mAb development activity saved 3 hours of analyst time and no repeats were needed
- Tecan® workflow for cIEF sample preparation will be expanded to include current mAb products and new modalities

**References**  
Freedom EVO Operating Manual, Tecan Trading AG

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