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Evaluation of emitter tip geometries to enhance the performance and robustness of ESI-MS

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Objectives

- The objective of this study was to investigate the holistic impact of all emitter tip specifications (tip angle, tip diameter, inner diameter, wall thickness) on the overall analytical performance (relative peptide count).
- Furthermore, we aimed to investigate and compare the performance of non-coated, metal-coated glass (MCG) and stainless-steel (SST) emitter tips in two common ion source configurations.

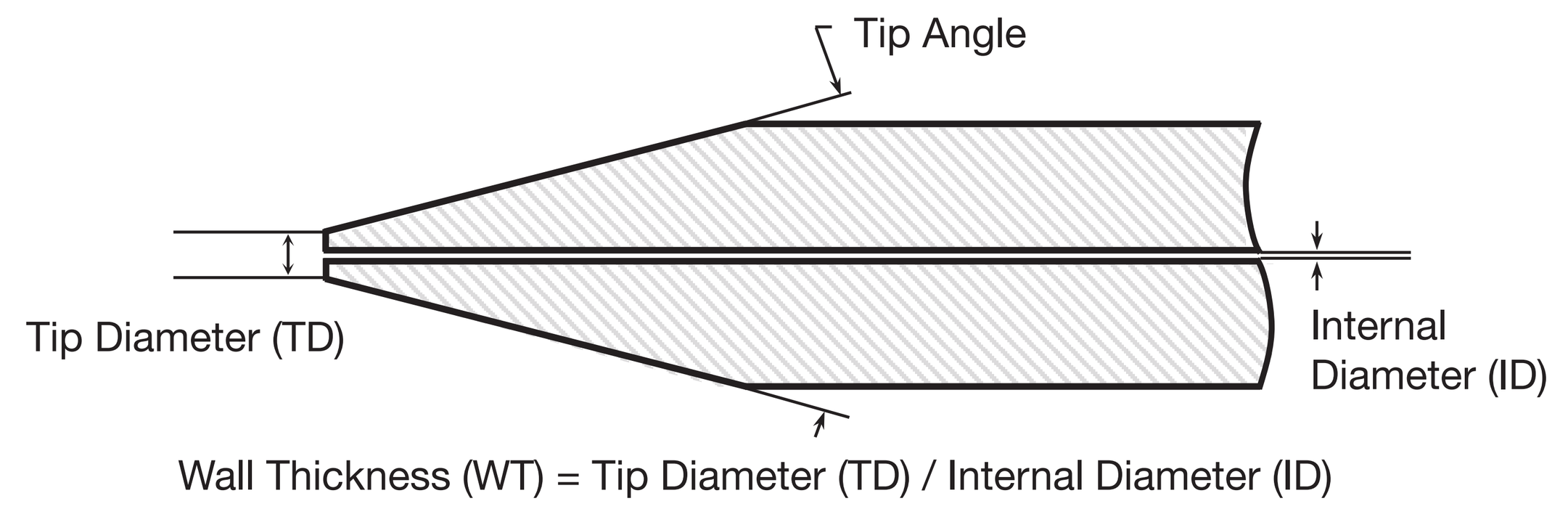


Figure 1. Emitter tip schematic with functional specifications.

Method

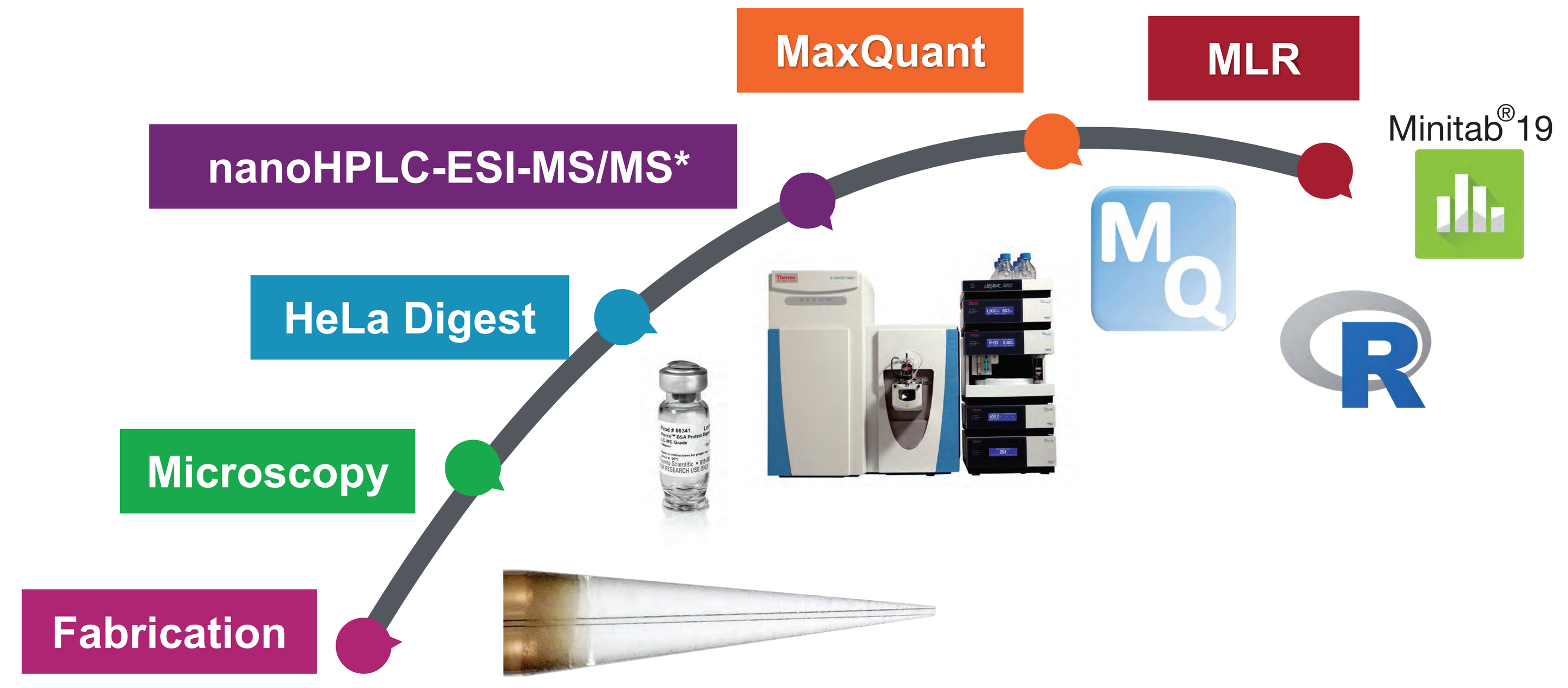


Figure 2. Emitter tip fabrication, testing and data analysis method (Note: 1hr LC gradient).

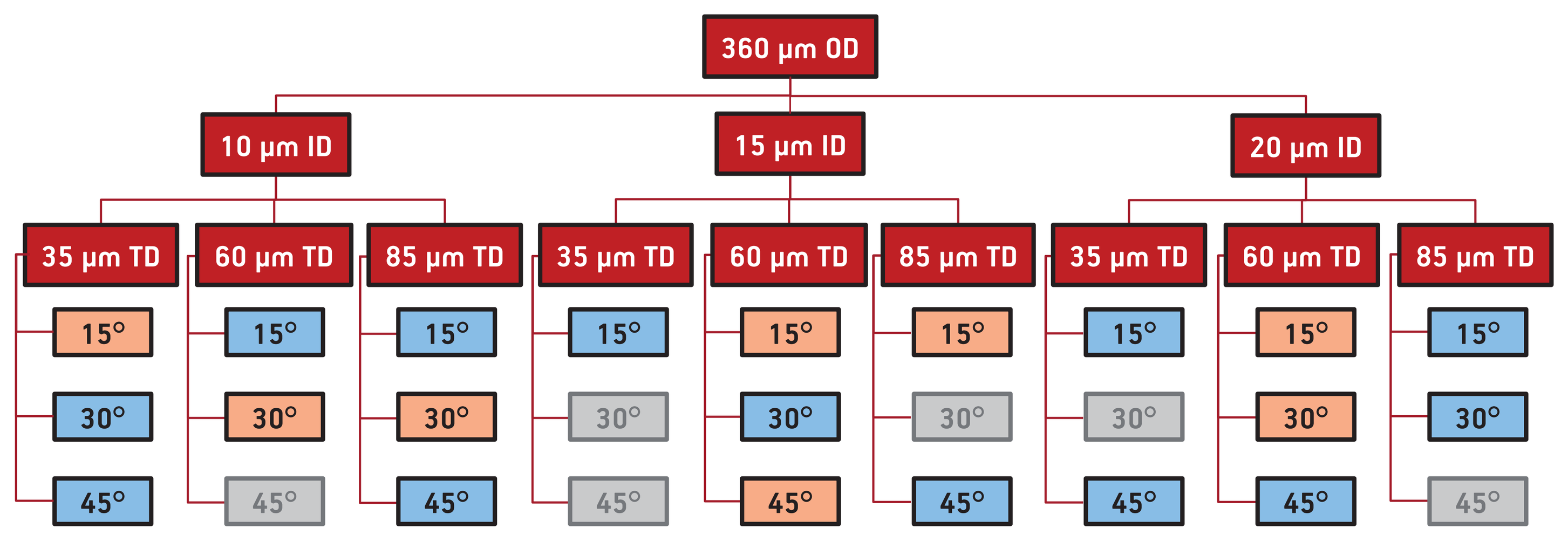
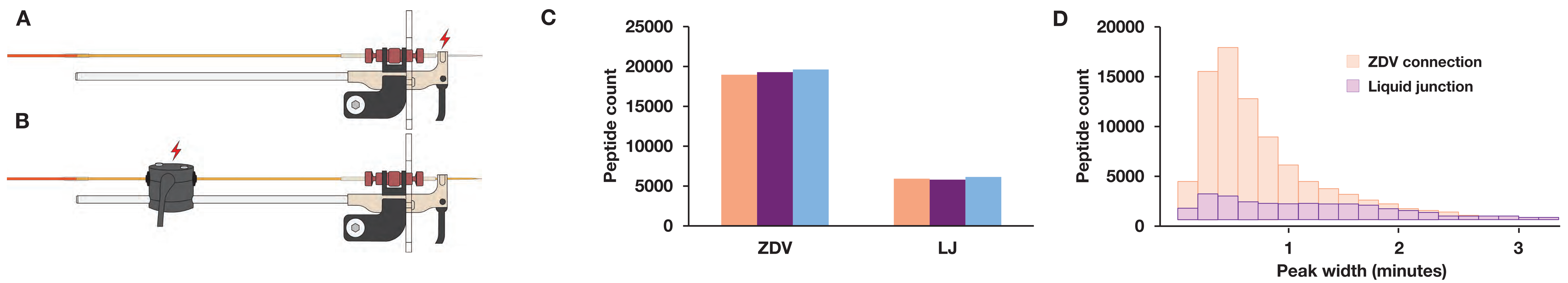


Figure 3. Design of experiment with initial MCG tip samples in blue and secondary samples (to improve model) in orange; ID (±1 µm), TD (±10 µm), TA (±1°).

Results

Effect of ion source configuration on analytical performance.



LJ configuration (Schematic B) reduced peptide coverage by 60-70% compared to ZDV setup, which was attributed additional dead-volume at the liquid junction interface.

Figure 4. (A) Zero Dead Volume (ZDV) with SST or MCG tip; (B) Liquid Junction (LJ) with SST or uncoated glass tip; (C) Peptide count versus configurations (2-hour HPLC gradient); (D) Histogram of peak width of peptides from each setup.

Multiple linear regression to evaluate the impact of variability in peptide count from external factors.

- Daily QC trials were performed using the ZDV-SST configuration and the **average peptide count was 9860 peptides** with a **standard deviation of 1423 peptides** over the course of the study; consistent peak widths indicated consistent chromatographic performance and emitter/column connection.
- Variation in the peak intensity correlated with variation in peptide count ($p < 0.001$, R-sq(adj) = 97.22%). The SST emitter remained constant and therefore further suggests that response variation was due to variability in the mass spectrometer.

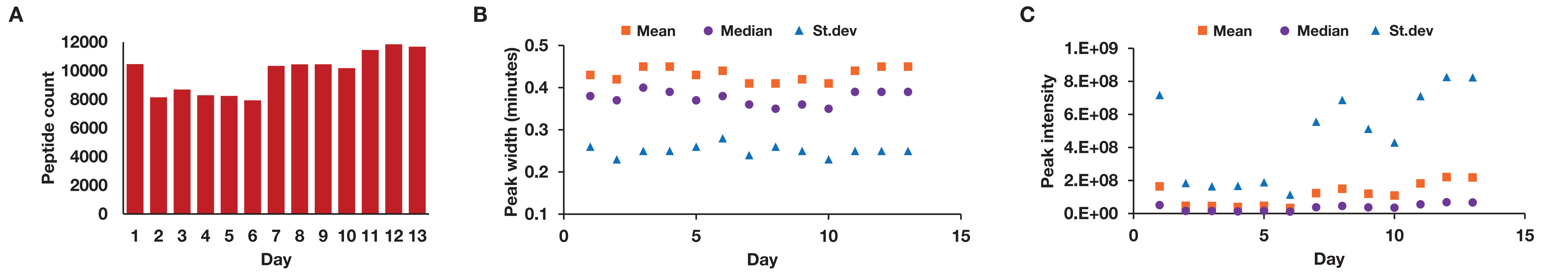


Figure 5. Variability in (A) daily QC peptide count, (B) peak width and (C) peak intensity using a ZDV-SST configuration (1-hour HPLC gradient).

Linear regression to evaluate the isolated effects of emitter geometry on peptide count.

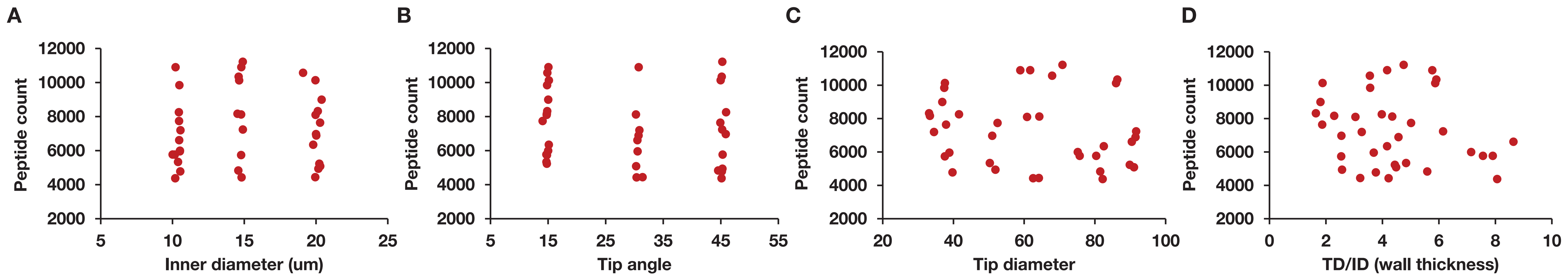


Figure 6. Single-variable results of the isolated effects of (A) Inner diameter, (B) Tip angle, (C) Tip diameter, and (D) TD/ID (wall thickness) on the analytical response.

Effect of tip geometry on peptide count and robustness at HV.

Table 1. Model summary and coded coefficients from multiple linear regression model of the average number of peptides versus tip geometries and instrument variation.

P-value	R-sq (adj)
< 0.001	62.81%

Explanatory variables	Estimated slope	P-value
Tip angle	-183	0.605
Wall thickness	-460	0.074
Voltage (kV)	333	0.301
Intensity mean	1640	0.000
Intensity CV	1386	0.000
Intensity mean *Intensity CV	1528	0.000

When all emitter tip data is isolated from instrument variability, wall thickness at the tip of the emitter has the greatest impact on the analytical response; thinner walls generally provide improved peptide counts.

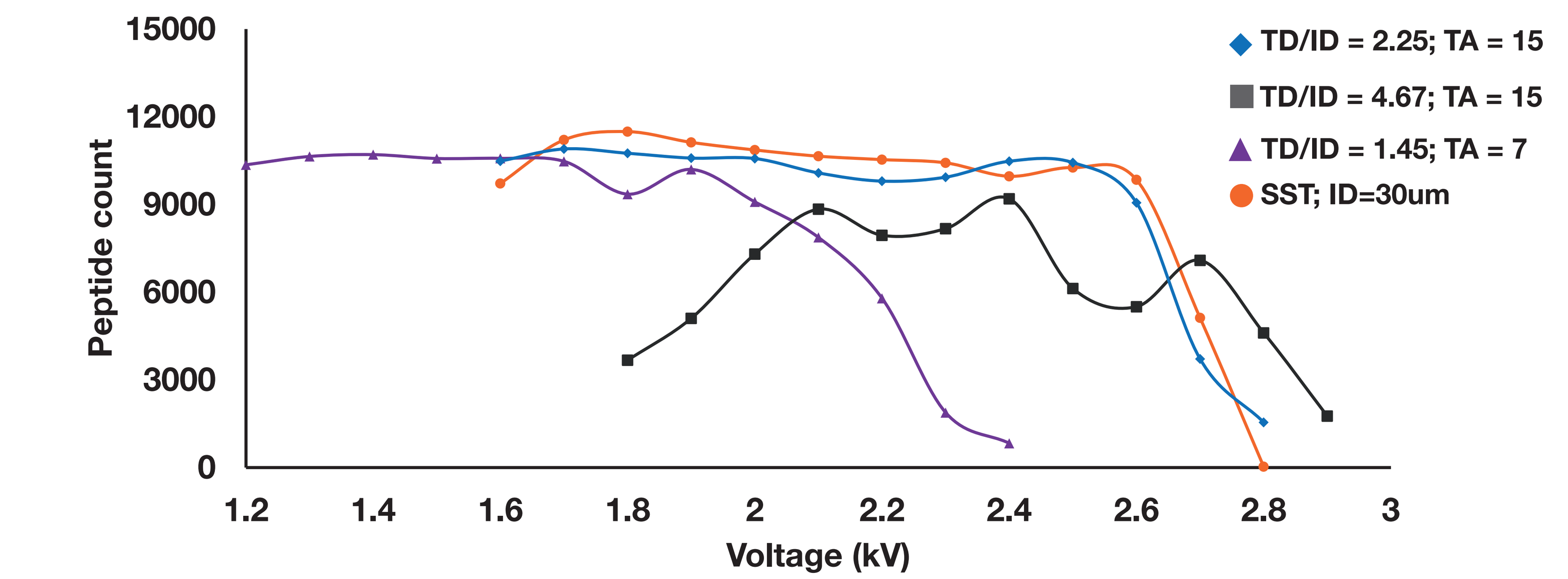


Figure 7. Effect of emitter tip material, geometry and applied voltage on peptide count (1-hour HPLC gradient).

- MCG emitters with thinner WT had lower onset voltages, consistent with that predicted by theory and previous reports[1,2].
- MCG tips with WT = 2.28 provided comparable performance to SST over a wide range of applied voltages, while MCG tips with WT = 4.67 had less stable and poorer performance at most voltages.

Conclusions

- MLR predicts that emitter tip wall thickness has the greatest impact on analytical response and thinner wall thicknesses generally provided higher and more reproducible peptide counts over a wide range of operating voltages.

[1]Mass Spectrom. Rev. 2009, 28, 898–917.; (2) Electrophoresis. 2014, 35, 1484–1488.; (3) Anal. Chem. 2006, 78, 2658–2664.

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