

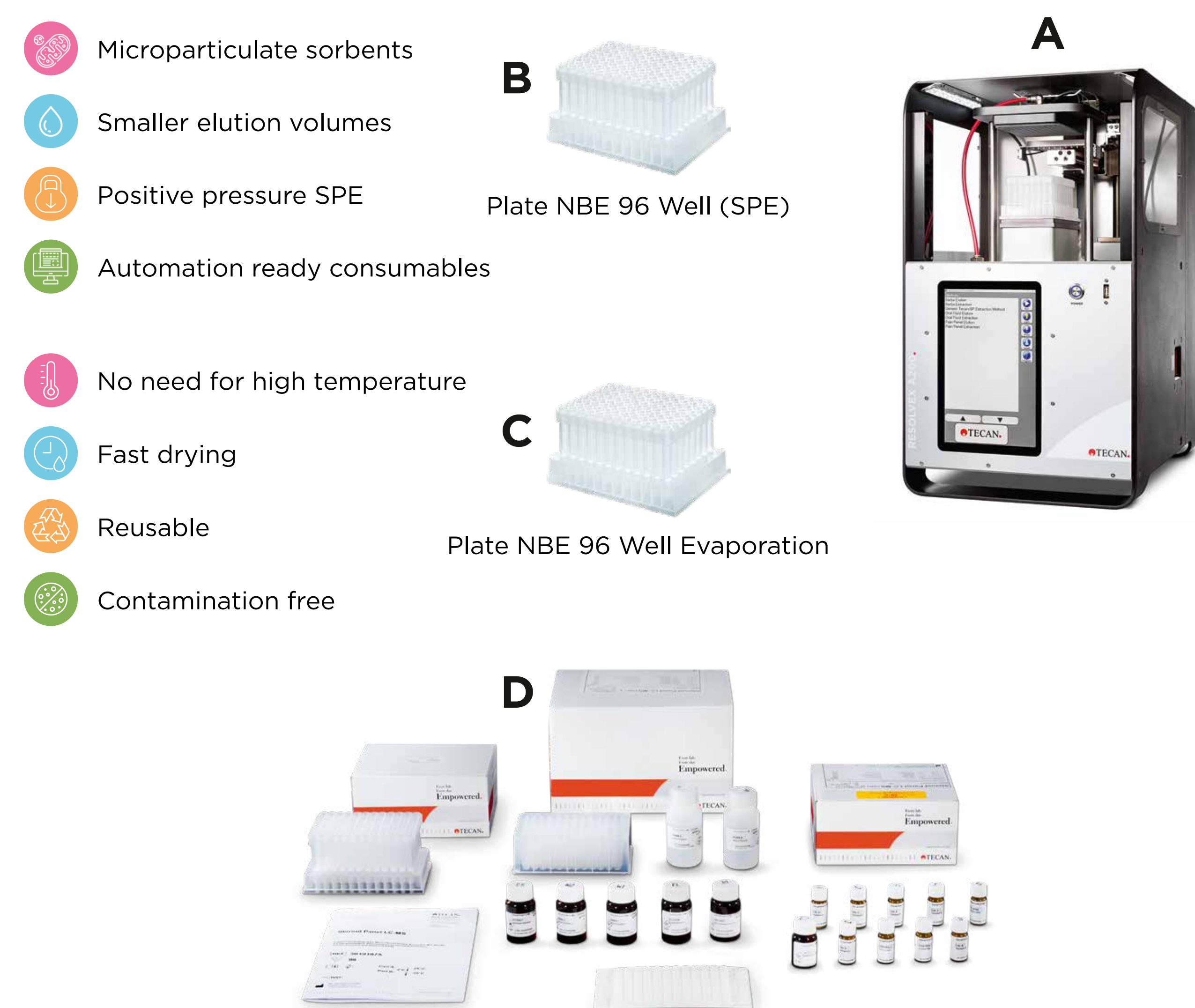
# CASE STUDY OF AN ALTERNATIVE DRYING METHOD FOR STEROID QUANTIFICATION USING THE TECAN STEROID PANEL LC-MS KIT.

Annalena Keitel, Ayham Al Ahmad, John Rohloff, Maïke Arndt

Tecan, IBL International GmbH, Flughafenstraße 52a, 22335 Hamburg, Germany

## INTRODUCTION.

The Tecan Steroid Panel LC-MS Kit is used for the quantitative determination of a broad range of steroids in human serum and plasma. The standard protocol, as described in the Instructions for Use (IFU), includes a validated solid phase extraction (SPE) and a drying step using an evaporator at elevated temperature. This study explores an alternative drying approach using the Resolvex® A200 system. The aim was to evaluate the feasibility of this method in comparison to the standard protocol, with a focus on maintaining consistency with international standards. Figure 1 highlights the semi-automated sample processing workflow with the Tecan® Resolvex® A200 system.



**Figure 1.** Semi-automated sample processing workflow with the Resolvex A200. **A:** Resolvex A200 instrument; **B:** NBE 96W SPE plate; **C:** NBE 96W Evaporation plate; **D:** Tecan Steroid Panel LC-MS kit with reagents and consumables.

## METHODS.

A different drying approach was applied to the Tecan Steroid Panel LC-MS Kit (RUO Cat. No. 30220266\*), using the Resolvex A200. Feasibility was tested in six independent runs with calibration standards, controls, and spiked samples. For method comparison, parallel processing was performed: one set of samples was dried at 40°C on an external evaporator according to the IFU, while the other set was processed using the alternative drying method on the Resolvex A200 at room temperature with the NBE 96 Well Evaporation plate (not part of the kit, Cat. No. 30138271). Quantification by LC-MS/MS, followed by Passing-Bablok regression and correlation analysis, demonstrated strong agreement between both methods, consistent with international standards.

## RESULTS.

The alternative drying approach resulted in complete evaporation within 58 to 90 minutes at room temperature, with the duration depending on both the matrix and the temperature. Feasibility studies showed high reproducibility, with all controls within acceptable ranges.

Method comparison demonstrated good agreement across 18 steroid analytes, with Passing-Bablok regression coefficients (R) 0.947-1.00, most were found above 0.99. All intercepts were within 95% confidence intervals (Table 2).

For key analytes, regression lines matched the line of identity, confirming equivalence. (Figure 2).

**Table 1.** Key Advantages of the NBE 96 Well Evaporation Plate used with the Resolvex A200 System.

| Benefit                              | Description  |
|--------------------------------------|--|
| <b>No Need for High Temperatures</b> | Evaporation can be achieved at room temperature, preserving sensitive samples.                           |
| <b>Contamination-Free</b>            | Design protects samples from external contaminants during evaporation.                                   |
| <b>Fast Drying</b>                   | Samples dry quickly, eliminating long waiting times.   |
| <b>No Extra Equipment Needed</b>     | No need for additional instruments, saving lab space and simplifying workflow.                           |
| <b>Reusable</b>                      | The evaporation plate can be used multiple times, making it cost-effective and environmentally friendly. |

**Table 2.** Summary of Passing-Bablok regression and correlation results for 18 steroid analytes.

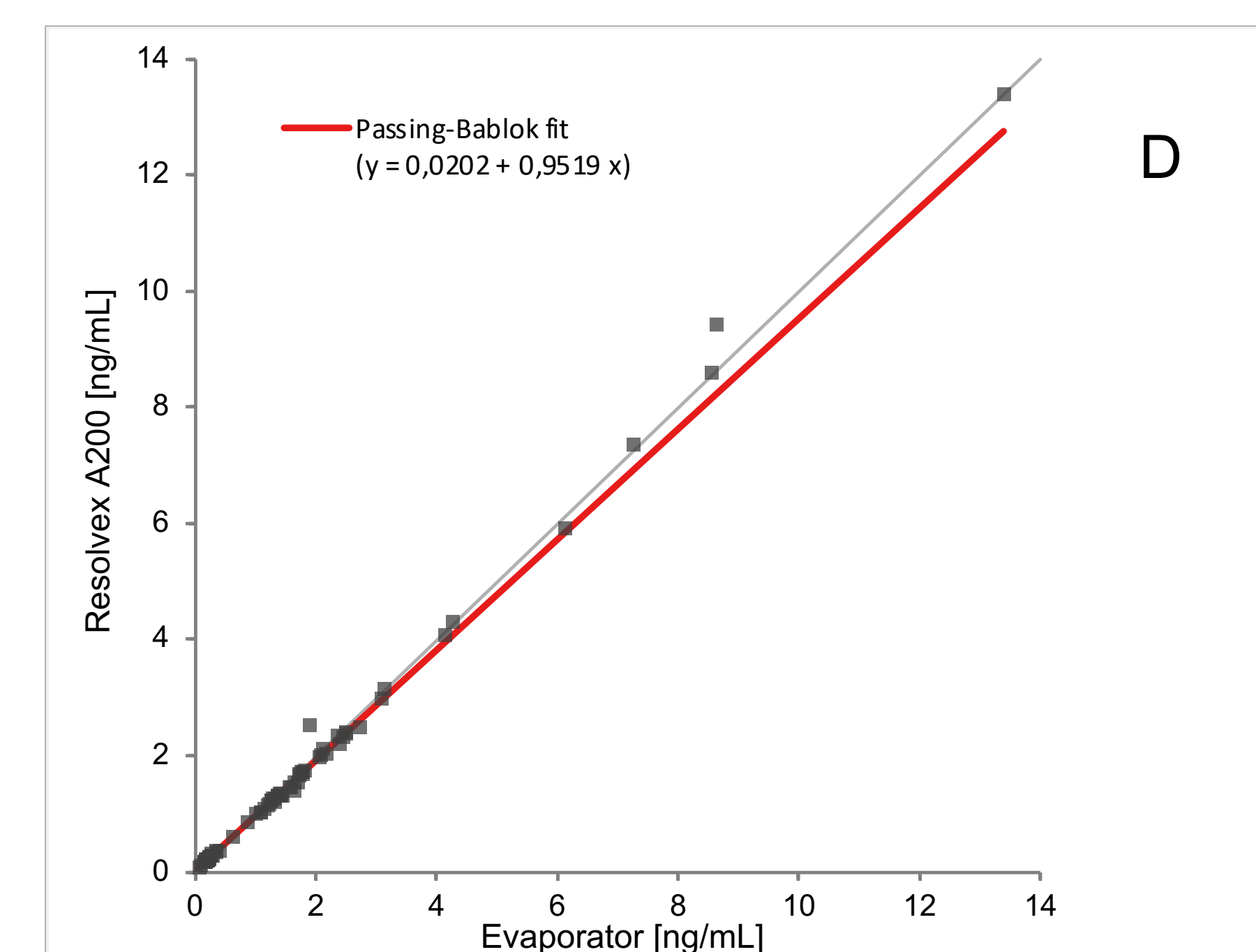
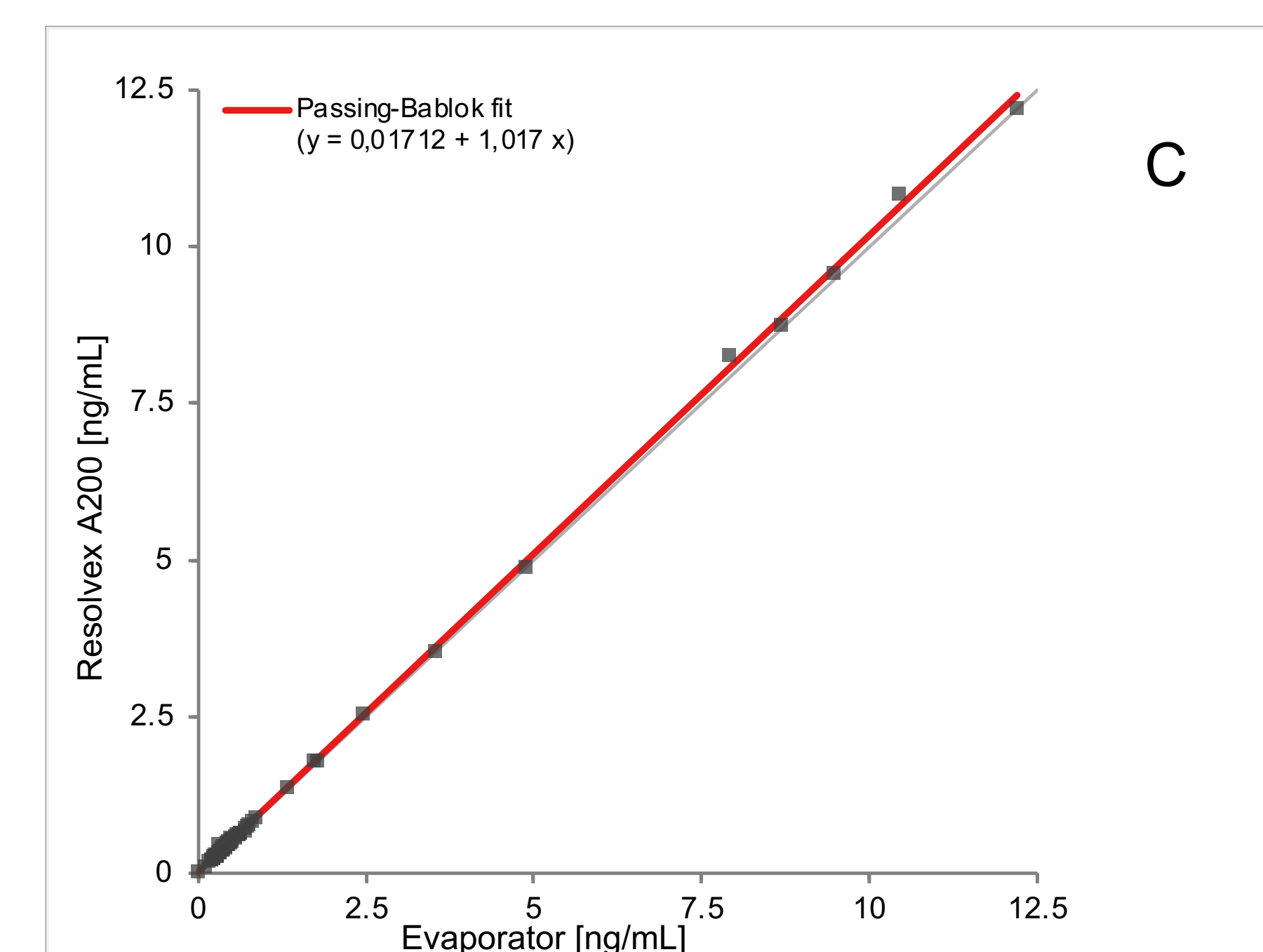
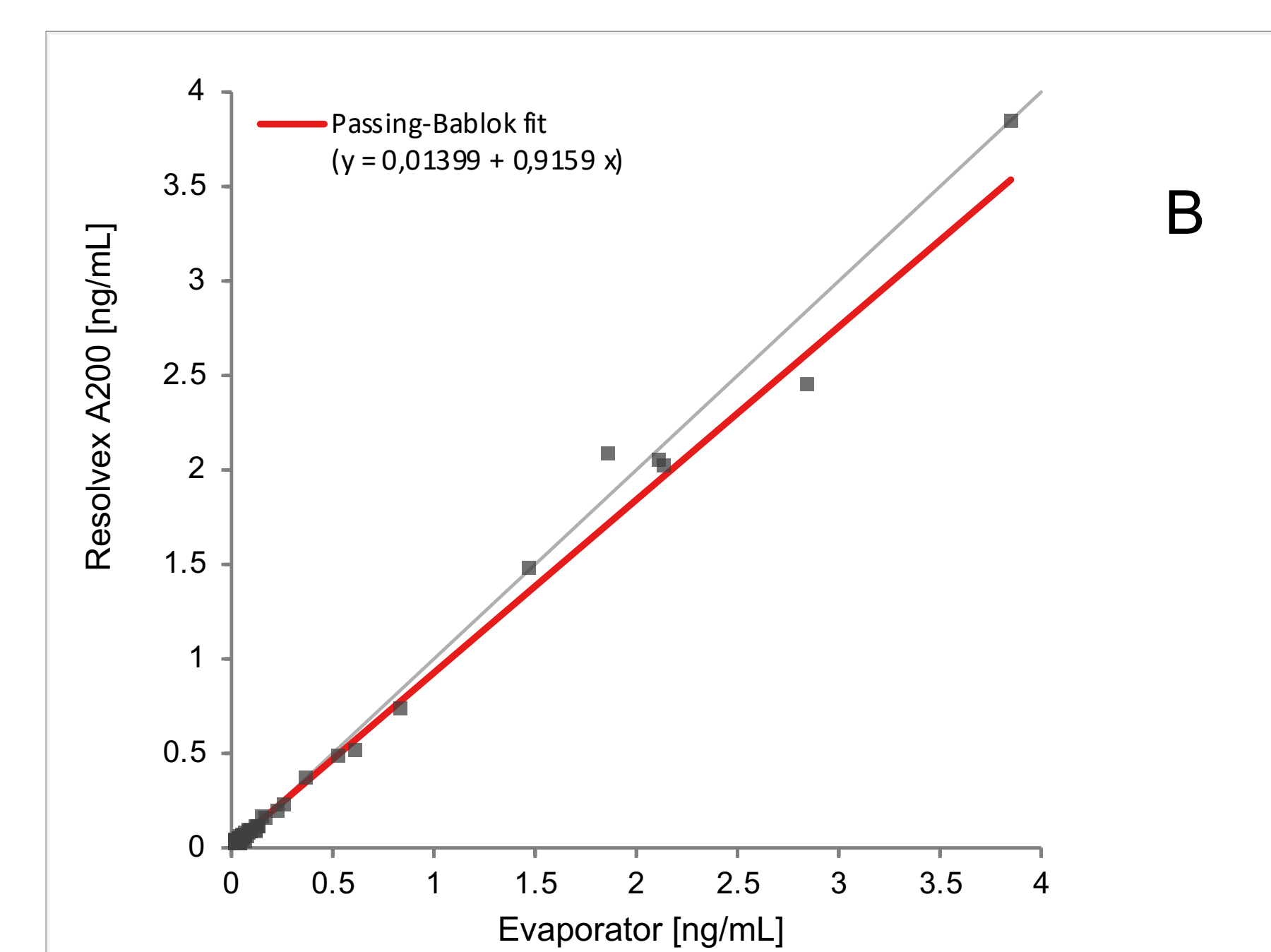
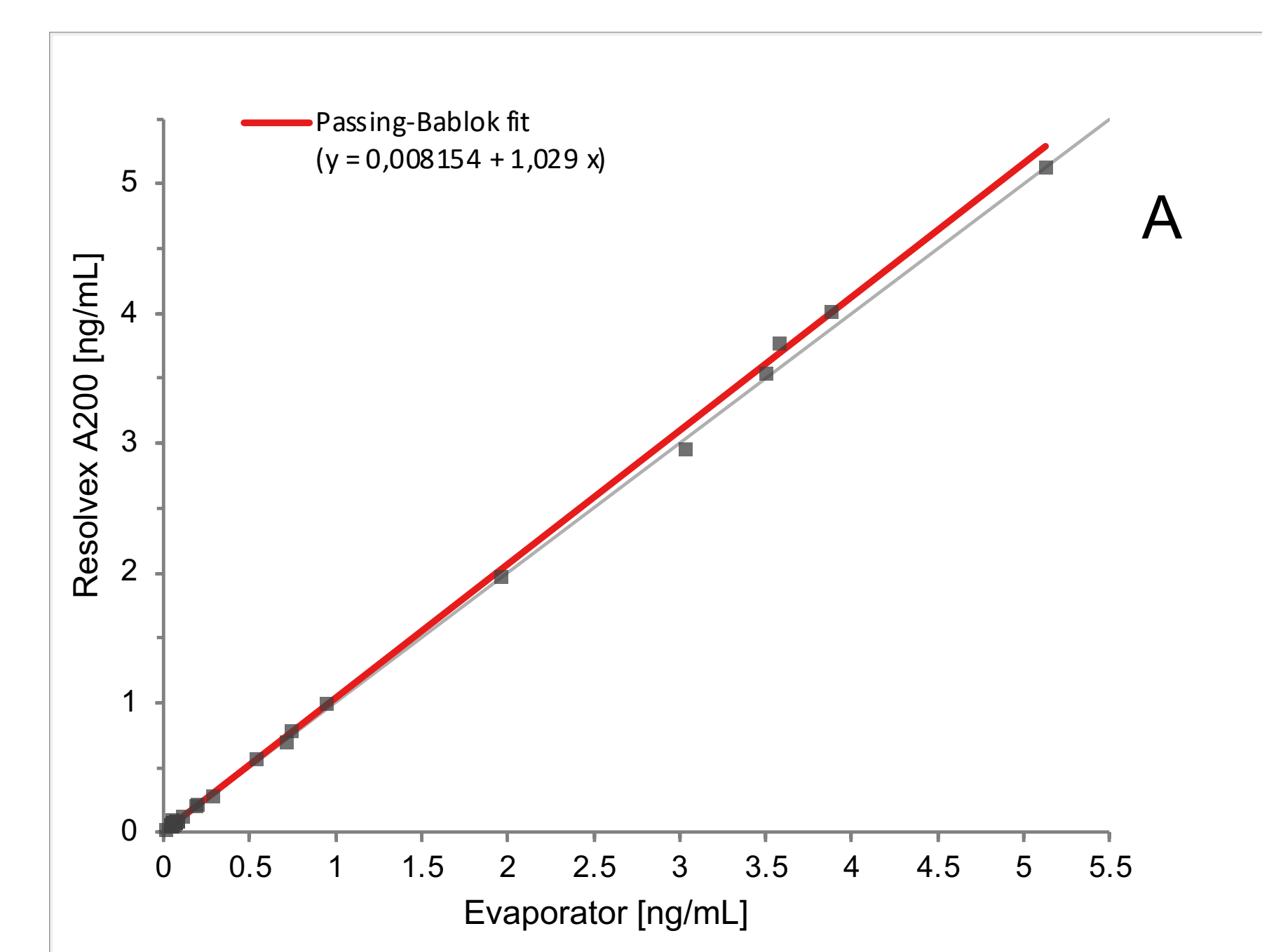
| Analyte                        | Passing-Bablok fit | r     |
|--------------------------------|--------------------|-------|
| 11-deoxycorticosterone         | 0,008154 + 1,029   | 1.00  |
| 11-deoxycortisol               | 0,0123 + 1,174     | 0.996 |
| 21-deoxycortisol               | 0,02319 + 1,139    | 0.995 |
| androstenedione                | 0,01712 + 1,017    | 1.00  |
| cortisol                       | 0,2135 + 1,129     | 0.985 |
| cortisone                      | 0,2722 + 0,9911    | 0.996 |
| progesterone                   | 0,01407 + 1,093    | 0.999 |
| testosterone                   | 0,0202 + 0,9519    | 0.998 |
| 17-hydroxyprogesterone         | 0,04729 + 1,414    | 0.991 |
| dehydrotestosterone            | 0,02903 + 1,003    | 0.992 |
| corticosterone                 | 0,08657 + 0,9417   | 0.997 |
| dexamethasone                  | 0,07204 + 1,07     | 0.998 |
| dehydroepiandrosterone         | 1,231 + 0,635      | 0.955 |
| dehydroepiandrosterone sulfate | -36,36 + 1,278     | 0.947 |
| aldosterone                    | 0,02976 + 1,003    | 0.994 |
| estradiol                      | 0,01399 + 0,9159   | 0.997 |
| estrone                        | -0,00469 + 1,105   | 0.999 |
| 17-hydroxypregnenolone         | -0,2852 + 1,272    | 0.992 |

## CONCLUSION.

This study demonstrates that the workflow can be streamlined. The results confirm that data quality is preserved, as shown by the Passing-Bablok regression plots. The drying option utilizing the Tecan Resolvex A200 is effective for LC-MS steroid quantification and offers users flexibility, without the need for additional equipment for evaporation. The data are provided for research and scientific discussion only.



On our homepage you will find further information on the Steroid Panel LC-MS, including three interesting videos.



**Figure 2:** Passing-Bablok regression and correlation coefficients (r) for selected steroid analytes: A) 11-Deoxycorticosterone, B) Androstenedione, C) Estradiol, D) Testosterone.

\* For research use only. Not for use in diagnostic procedures.