WHITEPAPER

Correcting SPR Signal Drift on Alto

SPR Signal Drift

Signal drift is one of the most common signal artifacts seen in SPR data. Drift is particularly troublesome when measuring tight binding kinetics, as the signal change due to drift may be greater than that of the off-rate. This makes the kd difficult to measure accurately. A typical method of overcoming signal drift is with a buffer blank subtraction, which involves subtracting a buffer baseline from the binding data in addition to reference channel subtraction. This method, known as "double referencing", requires buffer blank baselines to match the length of the binding curves, significantly extending the experiment duration. Applying a buffer blank subtraction to the data can also increase the resulting signal noise.

Traditional SPR instruments also rely on buffer blanks for subtracting injection artifacts in addition to drift from binding curves. Unlike traditional SPR instruments that rely on injections, Alto™ uses digital microfluidics (DMF) to move solutions in the form of droplets. In the absence of injections, Alto has no injection artifacts to remove, making drift the only undesirable environmental effect to require correction. Therefore, instead of using a full-length buffer blank, Alto is able to use a drift correction algorithm on a short representative baseline to correct for any baseline drift present in the data. This allows users to accurately measure tight off-rates without the need for them to add lengthy buffer blanks to their experiments.

Drift Correction on Alto

Nicoya® uses a linear baseline drift correction algorithm that allows users to subtract linear drift from the data without having to add considerable length to their experiments or introducing additional signal noise. The algorithm works by calculating the slope of a 200-second buffer baseline segment and extrapolating it over the duration of the subsequent binding curve. The extrapolated linear function is then subtracted from the binding curve, resulting in drift-free data.

To use the drift correction feature, users simply need to incorporate at least 200 s of buffer baseline prior to their first analyte association into their test design. Once the experiment run is complete and results are available, the user can toggle on the "Linear Baseline Drift Correction" option in the interaction menu as shown in Figure 1. This will apply drift correction to the displayed local interaction, or across all interactions in the dataset if applied from the Global Menu. If the baseline used for drift correction does not meet criteria for linearity, a warning message will be displayed to the user. Figure 2 shows an example of linear baseline drift correction applied to a single-cycle kinetics binding curve, significantly improving the data analysis fit.

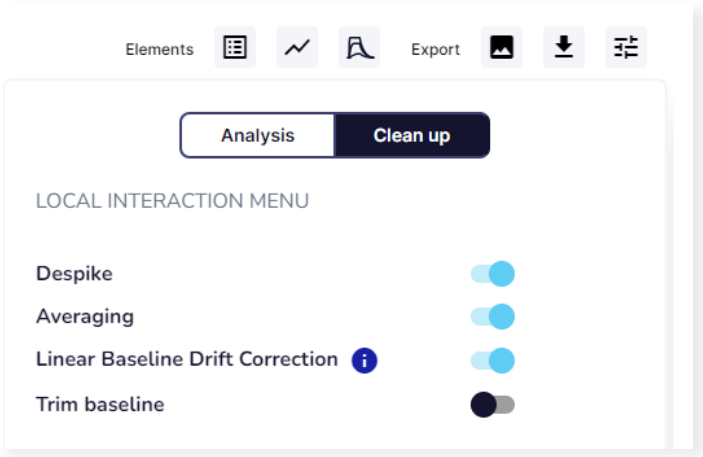
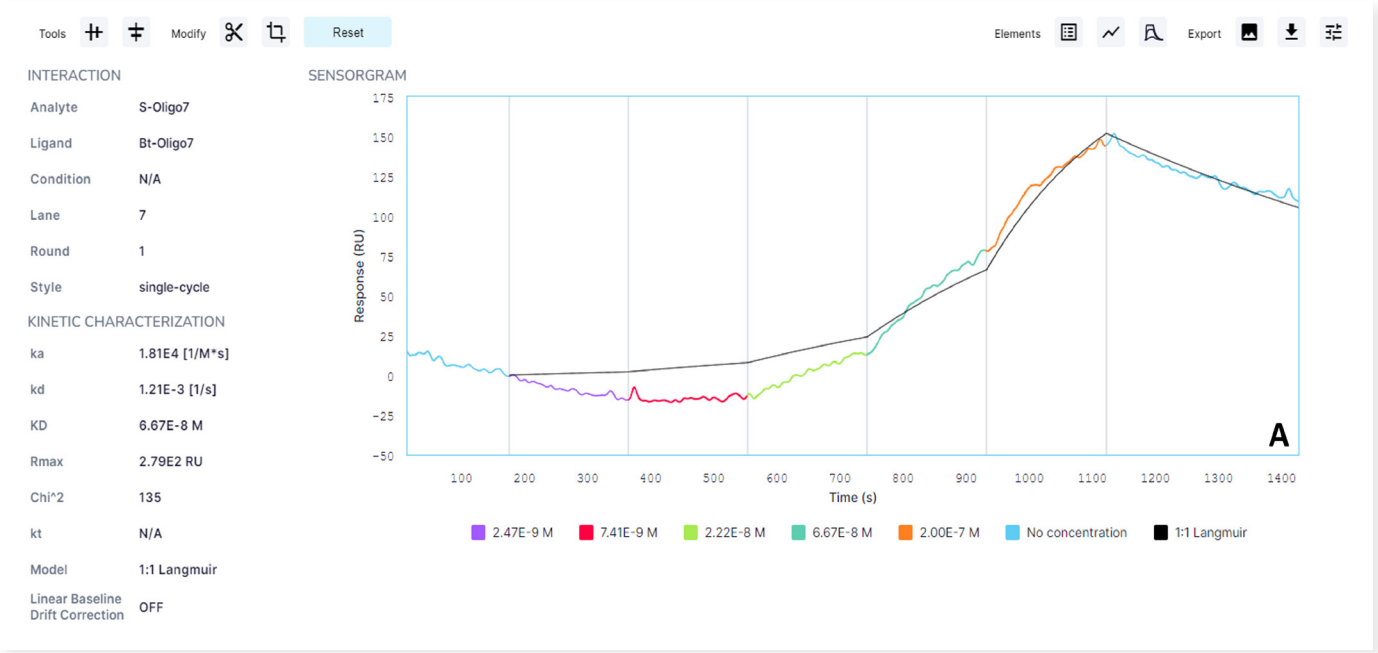


Figure 1: *Interaction tools showing the "Linear Baseline Drift Correction" post-processing option.*

(A)



(B)

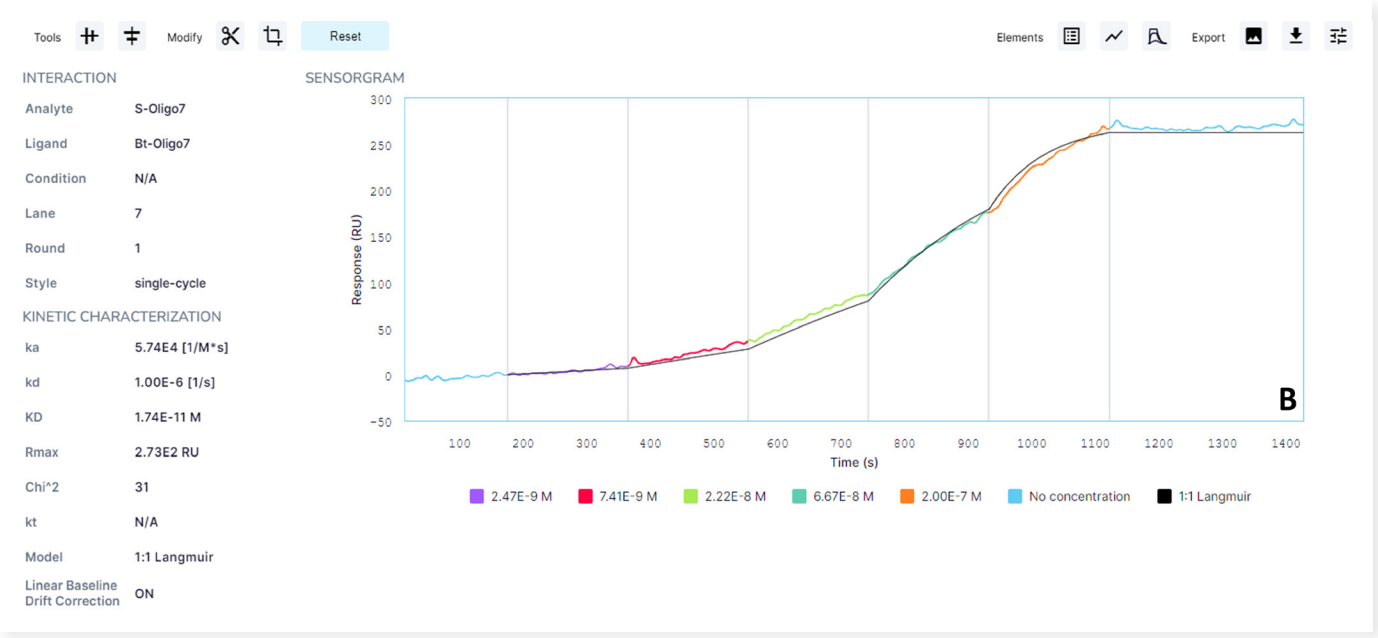


Figure 2: *Alto data before (A) and after (B) linear baseline drift correction.*