

Why Are Kinetics Better, Even For Low Affinity Interactions?

Kinetics considers the entire interaction over time, not just a single point. That means it extracts the on-rate (how fast binding happens) and the off-rate (how fast it falls apart) and then computes K_D from those using multiple concentrations. In low affinity interaction systems, that extra information matters, because two molecules can look “the same” by end-point affinity yet behave very differently in real biological settings (fast-on | fast-off vs slower-on | slower-off).

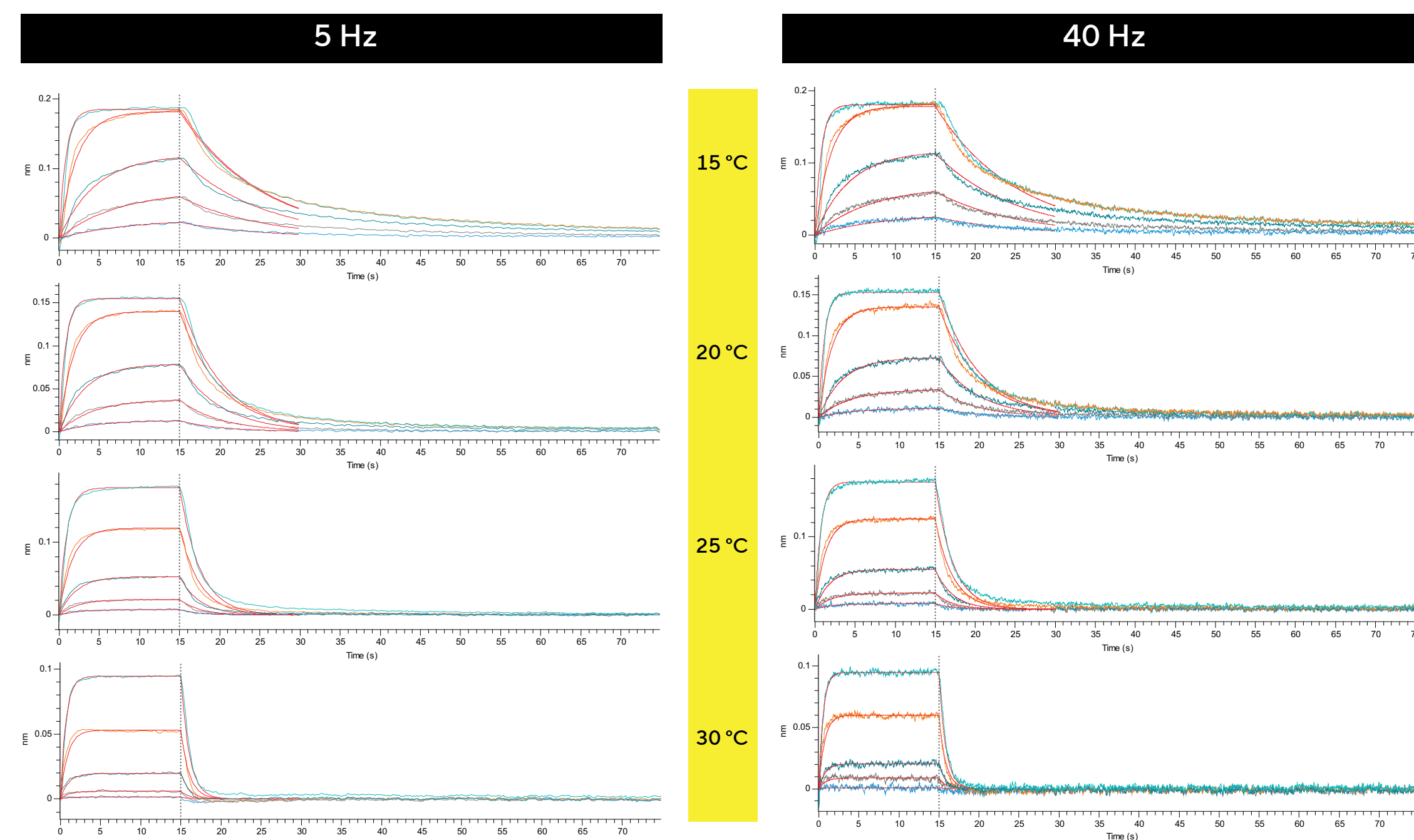


Figure 1: CD16b NA1 sensorgrams at different temperatures and 5 Hz and 40 Hz acquisition rate.

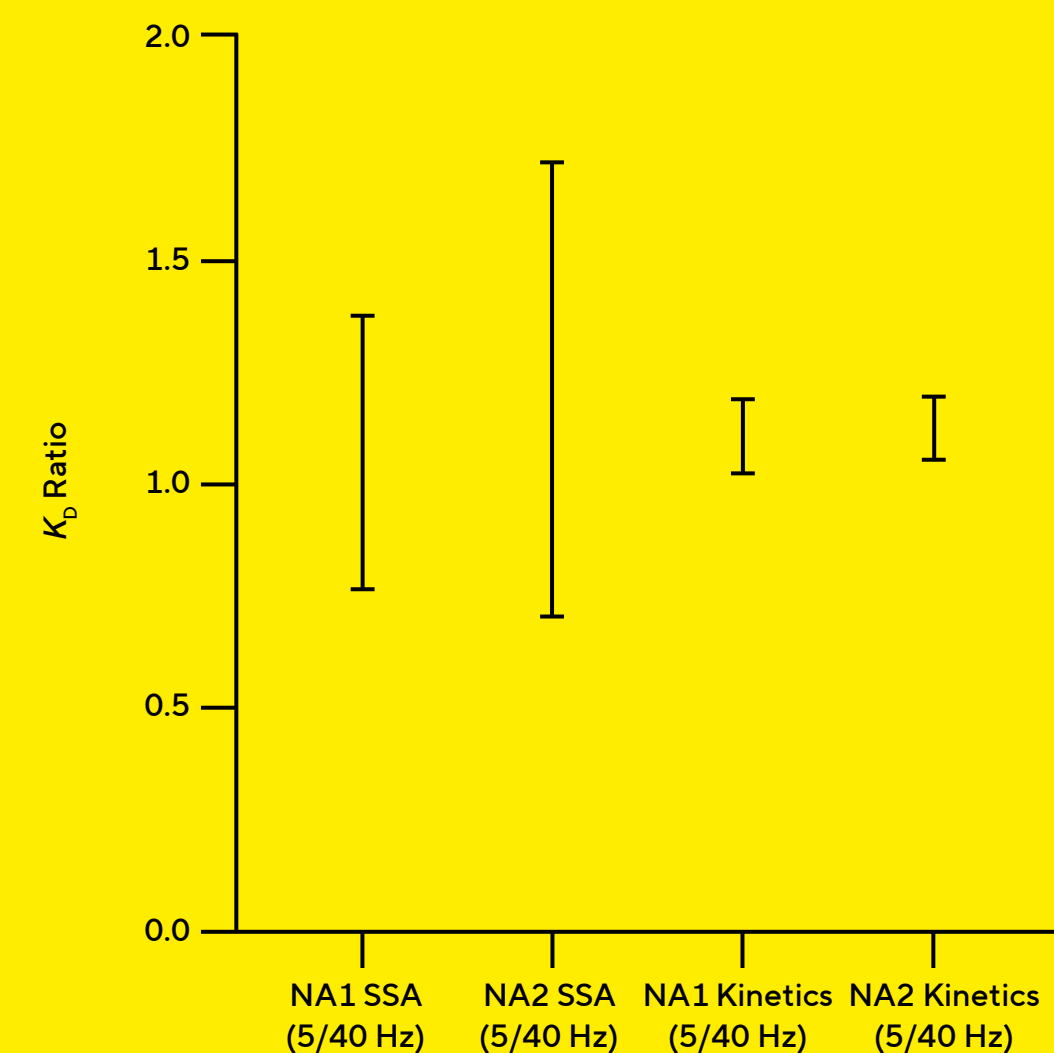


Figure 2: Comparison of the observed kinetic and steady state affinity (SSA) affinities for CD16b NA1 and NA2 isoforms collected at 5 Hz and 40 Hz data across a temperature range of 15 - 30 °C.

Why Do the Octet R8e’s Fast Data Acquisition Capabilities Matter?

The Octet® R8e’s 40 Hz acquisition essentially turns fast, low affinity binding into a data-rich kinetic curve (8x more timepoints than 5 Hz). In this assay format (15 s association, 60 s dissociation), 40 Hz allows capture of those all-important early critical data points, which improve kinetic fitting of low affinity interactions and reduce error.

 Want to learn more?

Read the full application note here:

Challenging the Dogma: The Use of Kinetics Versus Steady State in Low Affinity Interactions.

Application Note Overview

Challenging the Dogma: The Use of Kinetics Versus Steady State in Low Affinity Interactions

What this Overview Shows

When an interaction is weak (“low affinity”), the current trend is to default to steady-state (end-point) analysis because the curves can look “too fast” to capture kinetics or “too small” to fit reliably. But steady-state uses only one value per concentration (a single end-point), so small experimental differences (signal level, drift, uncertainty in the true maximum signal) can shift the calculated K_D . This is why steady-state K_D can change simply because you collected data at a different rate.