# Affinité's P4PRO: Precision Binding Kinetics Made Simple

## Introduction

Over the past decades, Surface Plasmon Resonance (SPR) has become a cornerstone technology for the biophysical characterization of protein interactions, playing a critical role in therapeutic development, diagnostics, and biosensing applications. While state-of-the-art systems have significantly accelerated scientific discovery, they often come with steep costs. Their complexity requires highly trained SPR specialists, and the cost is prohibitive for most academic and small-scale research labs. Accessing affordable, benchtop SPR instrument that delivers high-quality, reliable data without compromising usability remains a challenge as demand grows exponentially.

P4PRO is the reimagined benchtop SPR system by Affinité Instruments, delivering a miniaturized, user-friendly device that doesn't compromise on data quality. Designed to measure key binding parameters – including on-rate, off-rate, and binding affinity ($K_D$) – the benchtop P4PRO and AffiPump (Fig. 1) empower researchers with precision data previously reserved for high-end, centralized systems.


*Fig. 1. P4PRO (left) and AffiPump (right) for kinetic experiments with lensless SPR.*

At the core of Affinité's innovation is a proprietary lensless SPR technology, paired with a custom optical design and advanced data processing algorithms. This enables an ultra-compact, flexible SPR module capable of producing high-resolution kinetic profiles in real-time, without the need for labels or complex workflows.

Whether used as a rapid-access tool in individual research labs or as a complementary platform alongside existing label-free technologies in core facilities, Affinité delivers versatility across a wide range of applications. From traditional protein-protein interactions to more challenging targets such as aptamers, membrane proteins, peptides, exosomes, adeno associated virus (AAVs) and virus-like particles (VLPs), Affinité adapts to the needs of modern bioscience.

## How do the P4PRO and AffiPump work?

Affinité's P4PRO is a 4-channel SPR instrument designed to bring both throughput and experimental flexibility to the bench. Its intelligent dual-pairing architecture allows experiments to be conducted simultaneously in channel pairs (A & C or B & D), giving researchers the ability to optimize parameters in one pair while reserving the other for immediate follow-up studies or entirely different experiments (Fig. 2).

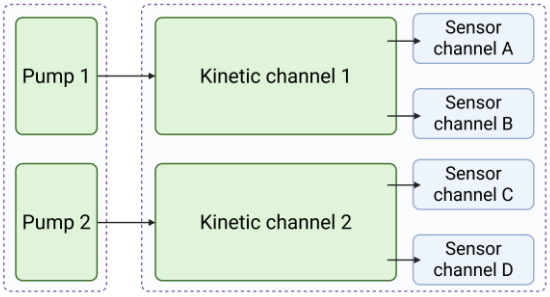


*Fig. 2. Schematic illustration of P4PRO and AffiPump.*

This modular approach not only streamlines experimental workflows but also doubles sensor utilization, significantly reducing the cost per data point and maximizing the value of each sensor chip.

To ensure high-quality kinetic measurements across all four channels, P4PRO is equipped with Affinité's proprietary AffiPump, a high-precision fluidic control system that delivers stable, consistent flow rates essential for accurate binding data. Whether you're optimizing complex interactions or running routine kinetic assays, P4PRO delivers performance that adapts to the researchers' pace and priorities.

## What does an SPR run look like?

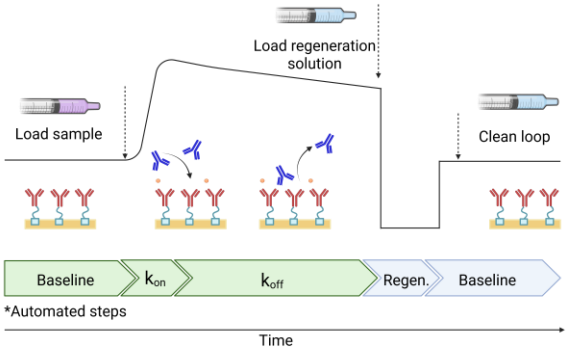


*Fig.3. Example of user's input during an SPR cycle for a single concentration on P4PRO.*

An SPR cycle generates a sensorgram, a real-time plot that captures the interaction between biomolecules across distinct phases: baseline, association, and dissociation (see Fig. 3). After immobilizing the ligand on the sensor surface, the user injects the analyte sample at a defined concentration. The association phase begins as the analyte binds to the ligand, followed by the dissociation phase, where binding is monitored after the injection stops and buffer flows across the surface.

To complete the cycle, a regeneration solution is injected to disrupt the interaction and restore the sensor surface for reuse. A buffer wash then cleans the injection loop to prevent cross-contamination before the next sample concentration is tested. This process is repeated across multiple analyte concentrations to generate a full kinetic profile, enabling accurate calculation of binding rates and affinity constants.

## Protein A-IgG binding kinetic setup

The Protein A–IgG interaction is a well-characterized binding pair commonly used as a reference standard across SPR platforms. In this experiment, Protein A was immobilized on a carboxyl-functionalized AffiCoat sensor in Channel A (Ch_A), while Channel C (Ch_C) was left unmodified to serve as a reference (Fig. 4). A series of increasing IgG concentrations (0.41 nM, 11.1 nM, 33.3 nM, and 100 nM) was then injected over both channels. Glycine regeneration was performed between each concentration to restore the sensor surface for subsequent cycles.

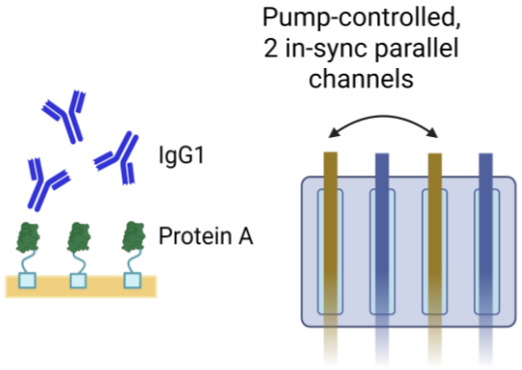


*Fig. 4. Sample sensor channel setup for protein A/IgG kinetic interaction measurement (left) and illustration of sensor channels active during the experiment (right).*

For each SPR cycle, reference subtraction was performed by subtracting the signal of the reference channel from the sample channel. This step corrects for temperature drift, bulk refractive index changes, and nonspecific binding, ensuring cleaner and more accurate data. The reference-corrected sensorgrams were then imported into TraceDrawer for visualization, fitting, and kinetic analysis (Fig. 5).

## High quality data with P4PRO

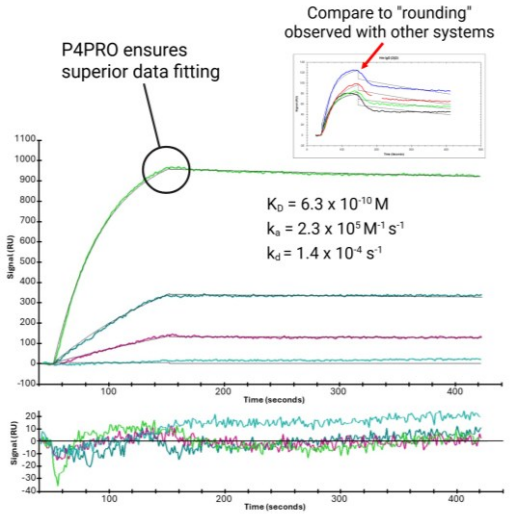


*Fig. 5. Processed P4PRO data in TraceDrawer (middle), the residual of the fit (bottom) and poor SPR data suffering from dispersion (inset).*

The entire Protein A-IgG experiment required less than two hours of hands-on time, demonstrating both the efficiency and ease-of-use of the P4PRO system. The resulting kinetic values $k_{on}$ of $2.3x10^5$ $M^{-1}s^{-1}$ and $k_{off}$ of $1.4x10^{-4}$ $s^{-1}$ gave a $K_D$ of 0.63 nM, consistent with values reported in the literature. The quality of the dataset is critical for generating accurate kinetic fits and accurate binding constants. In this example, the residual plot, which shows the difference between experimental and fitted data demonstrates excellent agreement with data points evenly scattered around zero. This indicates a high-confidence fit.

In contrast, the inset data produced by another benchtop SPR highlights a poor fit between experimental and fitted data. Such issues are often the result of suboptimal fluidic control. In many entry-level SPR systems, imprecise buffer switching leads to rounded sensorgram profiles, distorting the association and dissociation phases. This in turn produces kinetic parameters with high uncertainty and low interpretability. Affinité's high-performance fluidics and precise sample handling help eliminate these issues, ensuring clean sensorgrams, reproducible data, and reliable kinetic analysis.

The kinetic channels in the P4PRO are carefully designed to minimize flow artifacts, preserving the integrity of association and dissociation phases. When combined with the low-pulsation performance of the AffiPump, the result is data quality that competes with higher-end SPR systems typically found in core facilities.

By enabling high-quality, reproducible data at the benchtop, researchers can perform critical assay development and optimization in-house, saving both time and resources. When needed, these optimized conditions can translate to larger, more complex systems for final validation or extended studies, streamlining the entire workflow from early discovery to confirmation.

## Conclusion

The P4PRO is an affordable, lensless benchtop SPR instrument designed to deliver high-quality, reliable kinetic data. In addition to its precision performance, P4PRO offers several key advantages:

- ✓ AffiPump technology for rapid and precise buffer-sample-buffer switching
- ✓ 4-channel design for flexible, parallel experimentation and optimized sensor usage
- ✓ Wide sensor compatibility, including 4 essential surface chemistries and support for over 30 advanced, customizable surfaces
- ✓ Compact footprint with minimal maintenance requirements, ideal for individual labs or core facilities

P4PRO brings powerful SPR capabilities to the benchtop, empowering scientists to accelerate their research.