

**Silent in one assay, potent in another:
VHHs unlock a new way to block the cancer resistance target CDK2**

Joint case study profiles VHHs against CDK2/CycE1 in functional biochemical assays and finds binders acting through two distinct mechanisms

CAMBRIDGE, UK and MILAN, ITALY, 27 October 2026: A VHH antibody that had no effect on CDK2 kinase activity turned out to block the protein interaction that activates it, in a joint case study from **Isogenica Ltd.** and **Axxam S.p.A.** The study profiled VHHs against CDK2/CycE1, a cell-cycle complex linked to resistance to standard-of-care therapy in ovarian, breast and endometrial cancers. It found binders that inhibit the target in two ways: by blocking kinase activity, or by preventing the complex from forming.

CDK2/CycE1 regulates the G1 to S transition of the cell cycle. Selective small-molecule CDK2 inhibitors have entered clinical trials, but tumours can escape them by switching to the CDK4/6 pathway or by increasing CDK2 expression, which raises the dose needed and, with it, the risk of toxicity. According to the study, no VHHs against CDK2 or the complex had previously been reported.

Isogenica ran VHH selections against pre-activated CDK2 using their large, diverse synthetic libraries, discovering 192 VHH antibodies that bound the CDK2 antigen. Protein purification methods were introduced to Axxam's HTS automation, providing each VHH as a pure protein for testing in functional assays.

In the first assay, which measured CDK2 enzyme inhibition, four VHHs showed two-digit nanomolar potency. Three of these also stopped the complex forming in a second assay. But the two assays did not always agree. One potent VHH had no effect on complex formation, and one that looked inactive in the first assay blocked the complex, suggesting it binds where CycE1 docks rather than at the enzyme's active site. A third assay, using a different readout, confirmed the inhibition at lower potency, demonstrating the advantage of using multiple assays to deepen understanding through parallel datasets, especially when looking for binders to novel epitopes or with unique functionality.

"Binding tells you a VHH has found its target. It doesn't tell you what the VHH does there," said **Lurdes Rodrigues Duarte PhD, Director of R&D at Isogenica**. "Working with Axxam, we could profile a large, diverse set of binders in functional assays early. That showed us mechanisms a binding screen would have missed, including a molecule that demonstrated an unexpected way of operating."

"This study demonstrates how combining high-throughput VHH production with functional screening can transform the way we identify and characterize promising antibody candidates," said **Jean-François Rolland PhD, Senior Vice President, Early Drug Discovery at Axxam**. "By developing an automated platform capable of producing and purifying hundreds of VHHs in just a few hours, we can move rapidly from binder identification to functional characterization. This approach enables us to explore different mechanisms of action and identify promising candidates that might otherwise be overlooked."

The study is the first output of the partnership Isogenica and Axxam announced in September 2024, combining Isogenica's VHH discovery with Axxam's functional assays and, now, its automated VHH production. Following this proof-of concept study, significantly larger VHH panels can be explored directly following selections, enabling function-first discovery for rare functional binders such as agonists.

The full case study is available at <https://axxam.com/application-of-vhh-in-drug-discovery/>



About Axxam S.p.A.

Axxam S.p.A. is a leading provider of integrated discovery services across the life sciences industry. Within the drug discovery disciplines, we support pharma, biotech companies, start-ups, patient foundations as well as academic groups in their journey from hit identification to lead generation, regardless of the therapeutic area and target class. Our services include assay development, high-throughput screening, using either Axxam's high quality compound collections (synthetic and natural) or those provided by clients, hit validation, hit-to-lead and compound management. The same science-driven approach is also applied to identify new bioactive compounds for crop protection, animal health, food, beverage, pet food, cosmetic, and perfume industries. For more information, visit www.axxam.com.

About Isogenica Ltd.

Isogenica, The VHH Company™, is a Cambridge-based biotechnology CRO specialising in fast, customised VHH antibody discovery and engineering. Its synthetic VHH libraries enable high-diversity screening without animal immunisation, helping biotech and biopharma teams find high-quality, developable binders faster, including against challenging targets such as membrane proteins, GPCRs, ion channels and intracellular targets. With over 25 years of antibody discovery experience, more than 100 programmes delivered and multiple candidates progressed into clinical trials, Isogenica works as a responsive, science-led extension of its partners' teams. For more information, visit www.isogenica.com.