

OUTSTANDING PERFORMANCE

The performance of Discovery Bioanalytics phosphocellulose cation exchange paper matches that of discontinued Whatman P81 paper across a range of substrates, enzyme activity levels, and concentrations of radiolabelled ATP.

SUPERIOR MECHANICAL PROPERTIES

The tensile strength of Discovery Bioanalytics phosphocellulose cation exchange paper exceeds that of discontinued Whatman P81 paper, making it easier to handle when wet.

Phosphocellulose Cation Exchange Paper

The human genome encodes approximately 520 protein kinases and 20 lipid kinases, which play a central role in health and disease. Protein kinases constitute the largest proportion of targets in current drug development programmes. To date, 121 kinase inhibitors have been approved for clinical use. For example, Imatinib/Gleevec was a game changer for patients with chronic myelogenous leukaemia and heralded the age of targeted therapies and precision oncology.

Kinase assays are crucial to studying kinase activity and protein phosphorylation, and there is an ongoing requirement for reliable kinase assays for the identification and profiling of kinase inhibitors, in addition to the performance of fundamental cell signalling research. Radiometric assays are the "gold standard" method of performing kinase assays because of their direct readout, high sensitivity, reproducibility, reliability, and very low background signals. Radiolabelled ATP kinase assays rely on Whatman P81 phosphocellulose cation exchange filter paper (P81 paper) to capture peptide and protein analytes, after which unreacted ATP is washed off and incorporated radiolabelled phosphate quantified *via* scintillation counting.

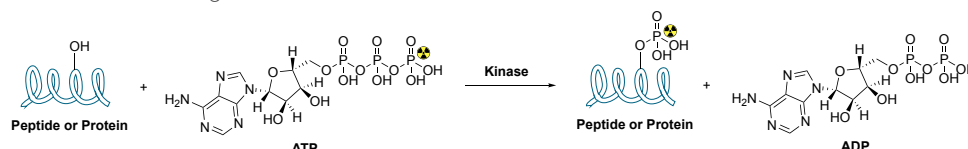


Figure 1: Radiometric kinase assay reaction scheme.

The production of P81 paper has been discontinued and stockpiled supplies are running out. Although alternative papers and assay methods have been reported, they do not match the performance of radiometric assays using P81 paper. Common issues experienced with alternatives to P81 paper include: 1) inefficient binding of both peptide and protein substrates across varied concentrations of radiolabelled ATP, 2) high background signals, 3) unavailability of a variety of formats, and 4) high cost. Furthermore, limitations of non-radiometric methods include: 1) ineffective for protein substrates, 2) high background signals, and 3) high cost.

Discovery Bioanalytics' phosphocellulose cation exchange filter paper matches the performance of discontinued Whatman P81 paper using peptide *and* protein substrates, a range of enzymes activities, and across various concentrations of radiolabelled ATP (Figures 2-4). Furthermore, our paper features superior mechanical properties (Figure 5).

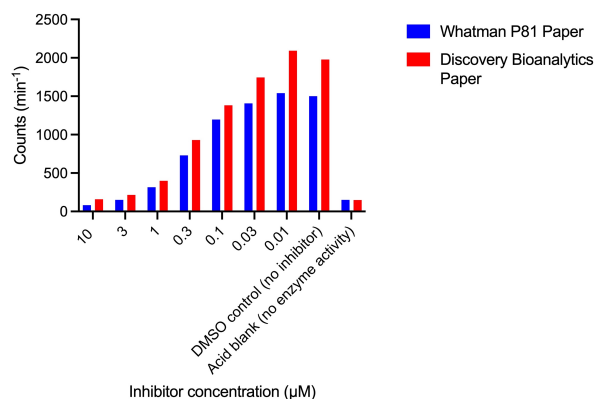


Figure 2: IC₅₀ curve of Sapk2b with inhibitor SB203580 (count time = 1 minute).

GREEN MANUFACTURING PROCESS

Discovery Bioanalytics phosphocellulose cation exchange paper is manufactured *via* an aqueous process using non-hazardous reagents.

AVAILABLE IN A VARIETY OF FORMATS

Discovery Bioanalytics phosphocellulose cation exchange paper is readily available in A4 sheets and can be made available in other formats.

For more information on any of our products or services, please visit us at:
www.discoverybioanalytics.com

Phosphocellulose Cation Exchange Paper

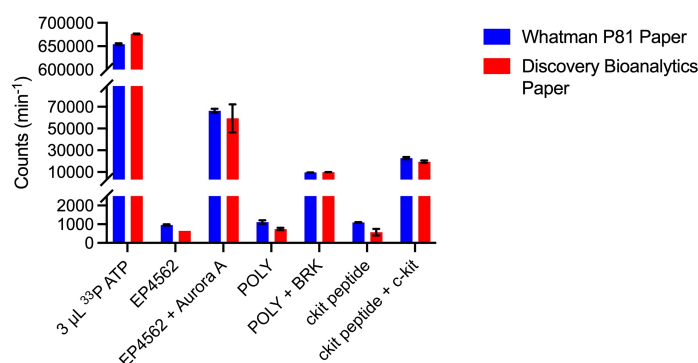


Figure 3: Kinase assays of: **a)** EP4562 using Aurora A, **b)** POLY using BRK, and **c)** c-kit peptide using c-kit (count time = 1 minute).

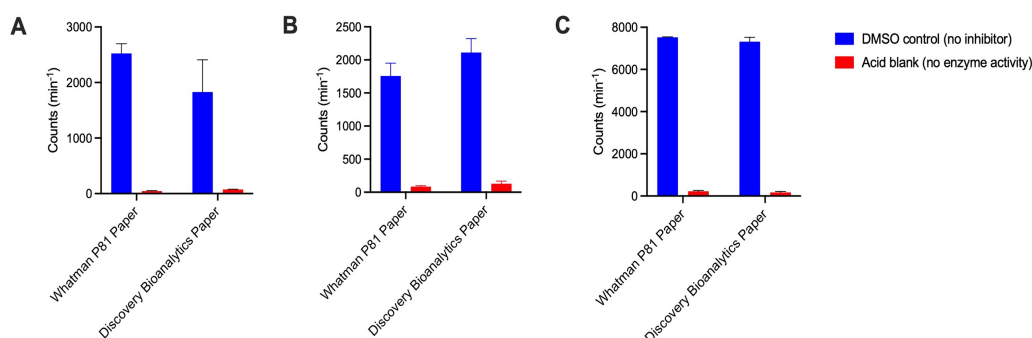


Figure 4: Quantitation of radiation on cation exchange papers *via* scintillation counting for the kinase assay of **A:** PKBα using 5 μM ³³P ATP, **B:** Sapk2b using 20 μM ³³P ATP, and **C:** SRPK1 using 50 μM ³³P ATP (count time = 1 minute).

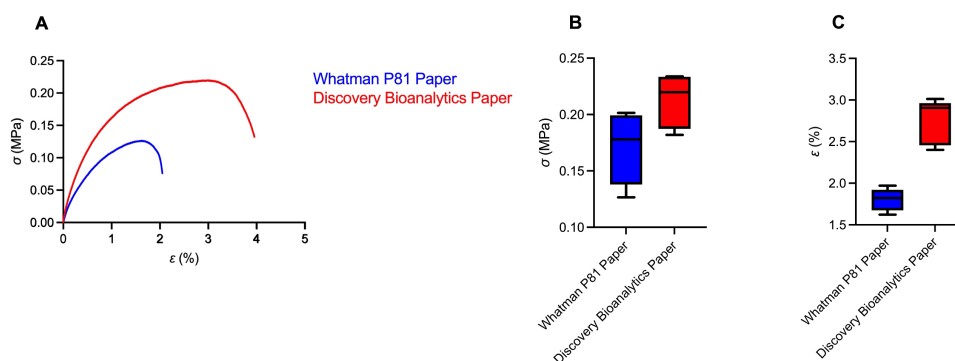


Figure 5: Wet filter paper **A:** stress-strain curves, **B:** tensile strength (σ) values, and **C:** percent tensile strain (ϵ) values.