

KINEXUS

Kinex™ Reverse Lysate Microarray

KRLM-1.0

Case Study

Probing with an Anti-phospho-ERK1/2 Antibody



www.kinexus.ca

The Kinex™ Reverse Lysate Microarray (KRLM) is a powerful tool for rapid characterization of a protein for which a highly specific antibody is available. This unique microarray features about 250 different lysates prepared from several human tumour cell lines that have been treated with diverse hormones and drugs in time course and dose response experiments as well as tissues from monkey, rat and mouse. Each lysate has been printed in triplicate at four different concentrations to ensure that detection of the antigen is within the linear range. For this presentation, we have provided the results from the probing of one KRLM-1.0 microarray with an phospho-site-specific antibody that targets the activations sites of the MAP kinases ERK1 and ERK2. These kinase are phosphorylated in response to diverse growth factors and during oocyte maturation. For comparison, we have provided some of the results from parallel immunoblotting studies with similar cell and tissue extracts. Any interesting results from the KRLM-1.0 analysis can be confirmed with our Kinetworks™ Custom Sample Screen KCSS 1.0 immunoblotting service. With the KRLM-1.0 service, the results are provided in a Microsoft Excel spreadsheet. For an additional US \$300, Kinexus can prepare a series of Powerpoint Slides with the results from a single KRLM-1.0 analysis as exemplified here. Individual graphs can be prepared for US \$20 each. For more information, contact our Technical Services representatives toll free in North America at 1-866-KINEXUS or by e-mail at info@kinexus.ca.

KINEXUS

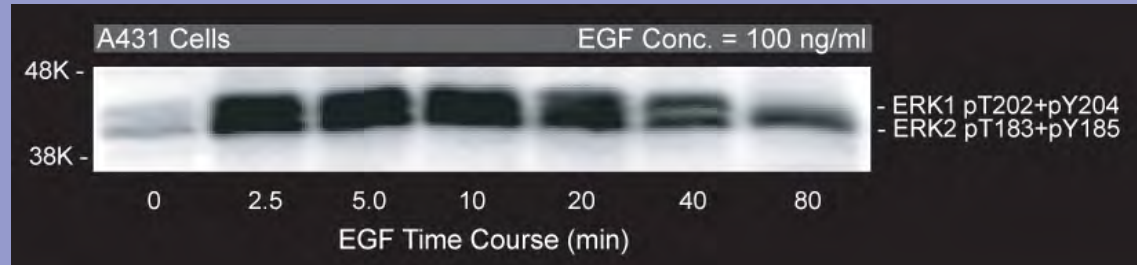
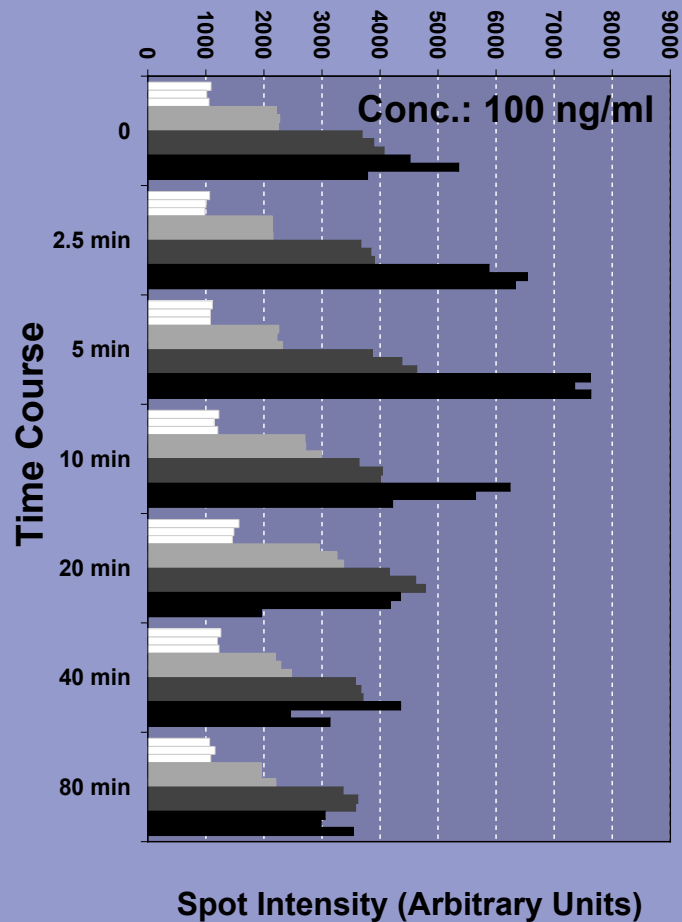


Kinex™ Reverse Lysate Microarray KRLM-1.0

Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

Cell Line: A431 Human Epidermoid Carcinoma

Treatment: Epidermal Growth Factor



Legend

- 2.5 mg/ml
- 1.25 mg/ml
- 0.625 mg/ml
- 0.3125 mg/ml

www.kinexus.ca

KINEXUS

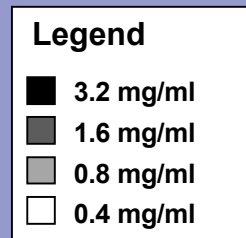
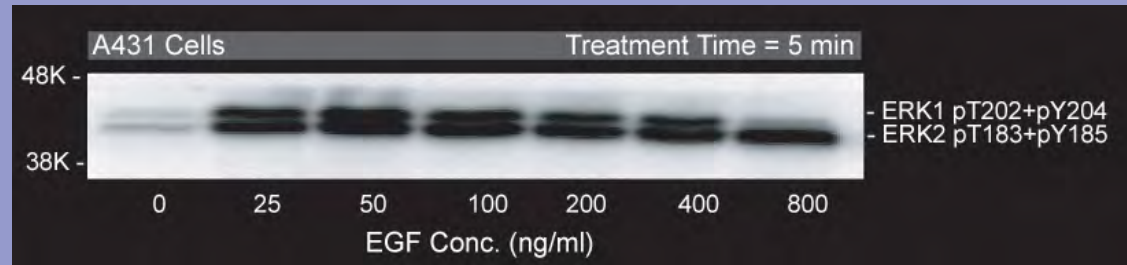
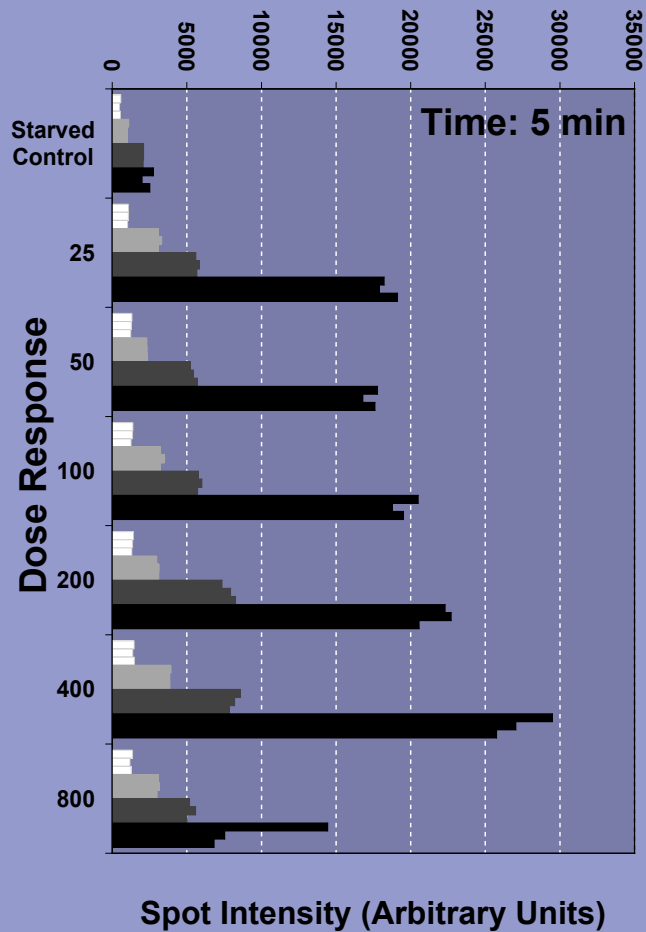


Kinex™ Reverse Lysate Microarray KRLM-1.0

Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

Cell Line: A431 Human Epidermoid Carcinoma

Treatment: Epidermal Growth Factor

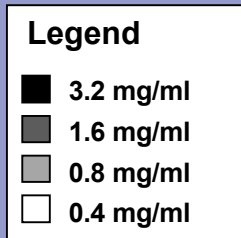
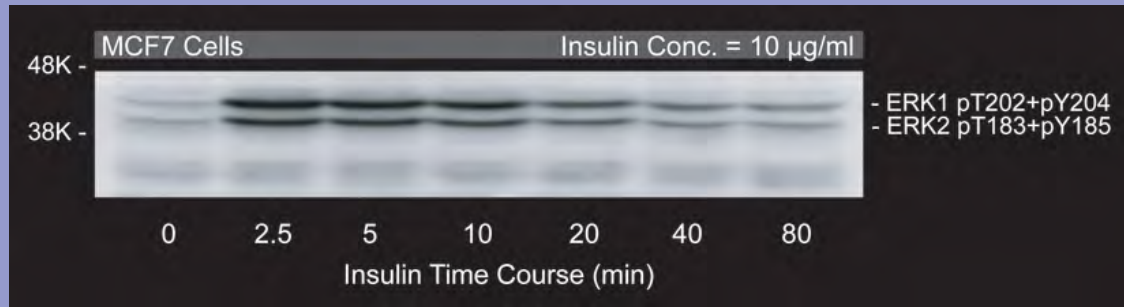
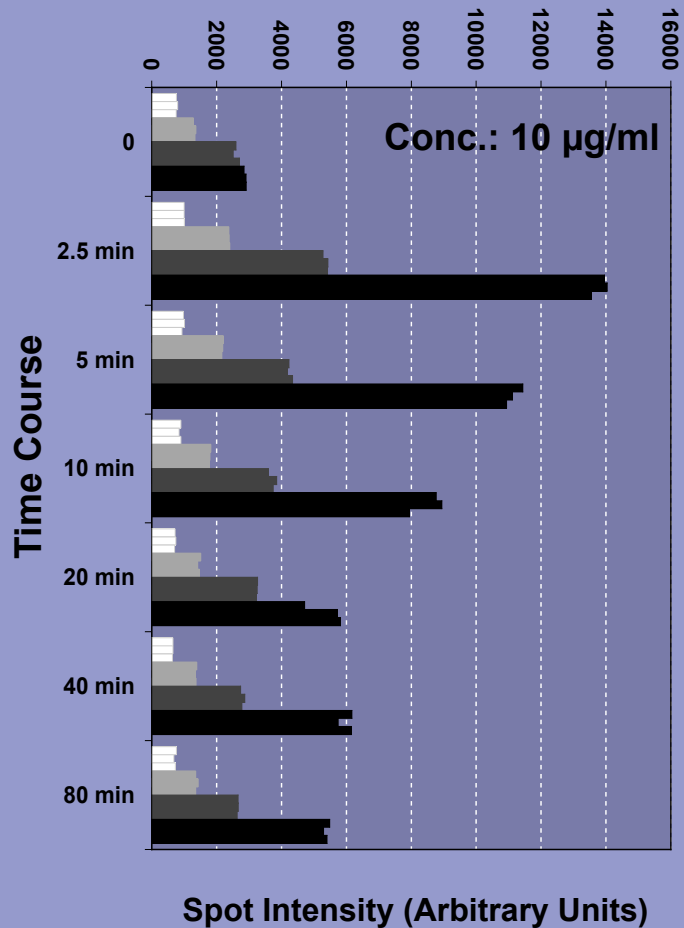


www.kinexus.ca

Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

Cell Line: MCF7 Human Breast Adenocarcinoma

Treatment: Insulin

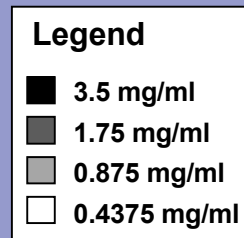
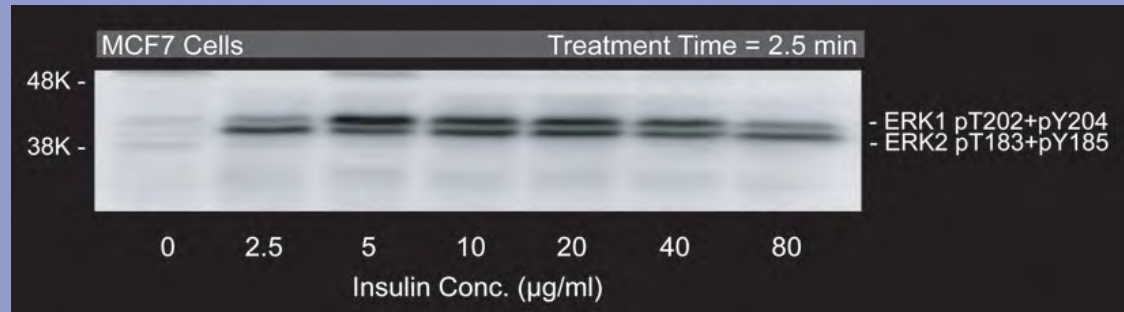
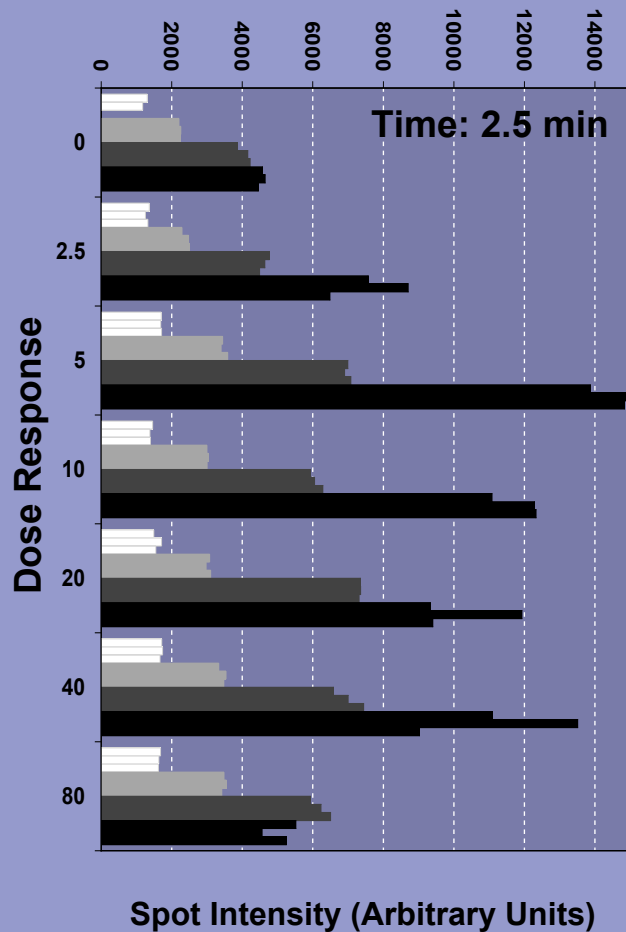


www.kinexus.ca

Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

Cell Line: MCF7 Human Breast Adenocarcinoma

Treatment: Insulin



www.kinexus.ca

KINEXUS

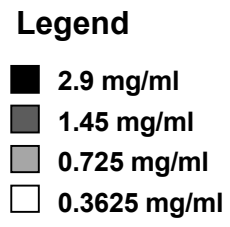
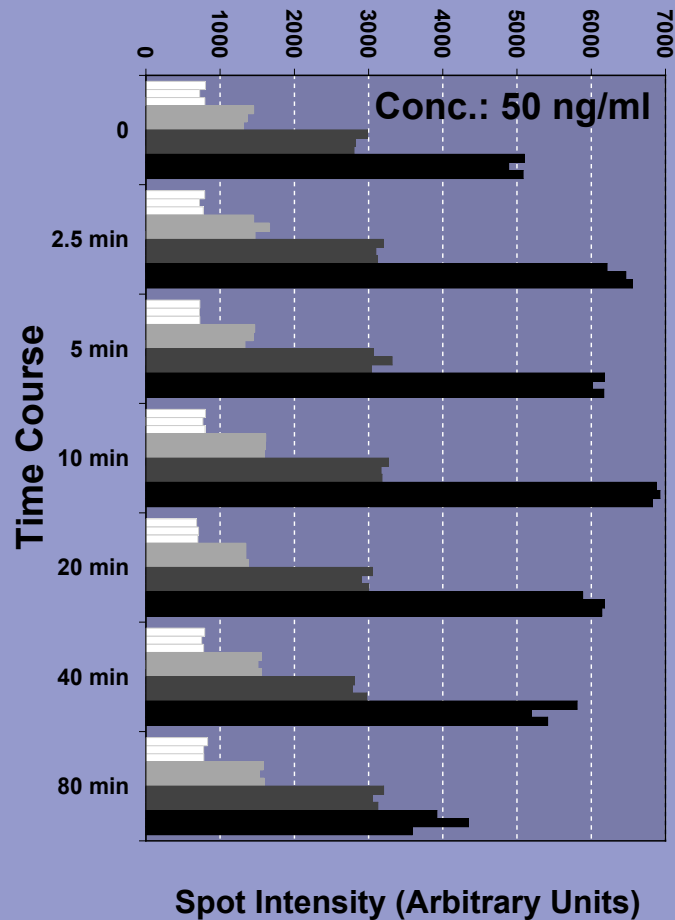


Kinex™ Reverse Lysate Microarray KRLM-1.0

Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

Cell Line: T98G Human Brain Glioblastoma

Treatment: Platelet Derived Growth Factor



www.kinexus.ca

KINEXUS

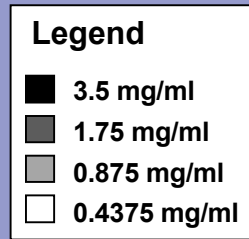
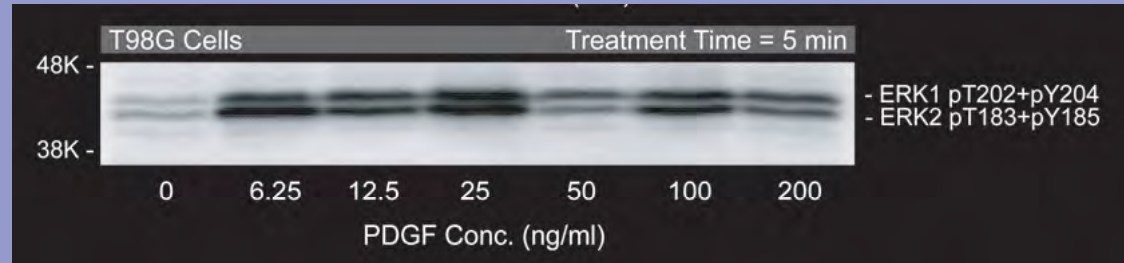
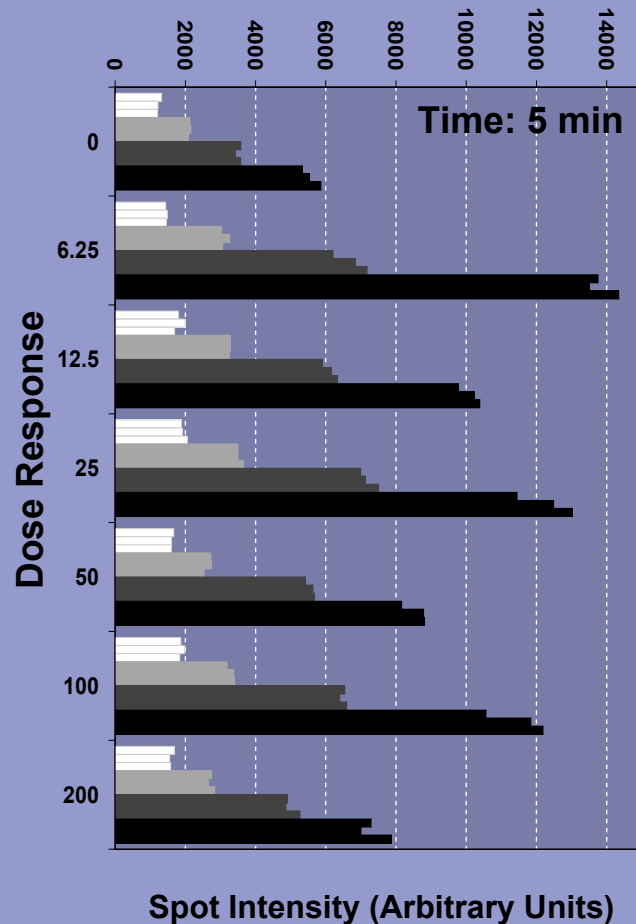


Kinex™ Reverse Lysate Microarray KRLM-1.0

Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

Cell Line: T98G Human Brain Glioblastoma

Treatment: Platelet Derived Growth Factor

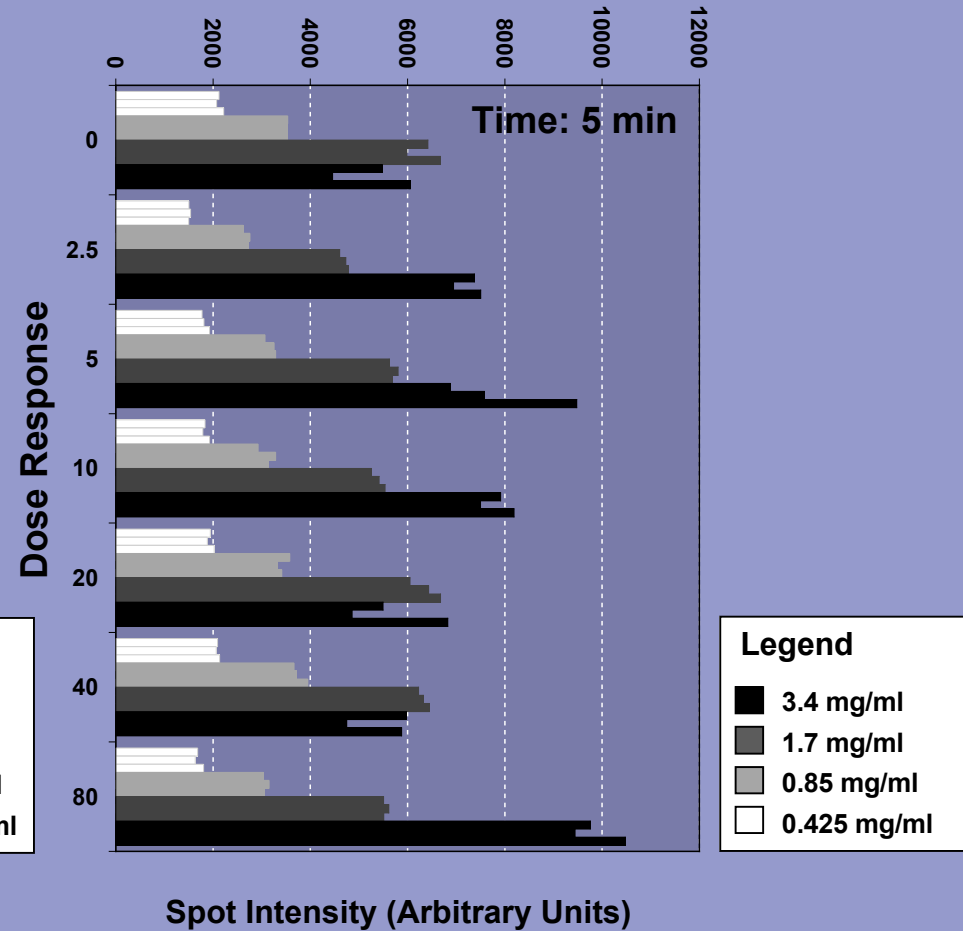
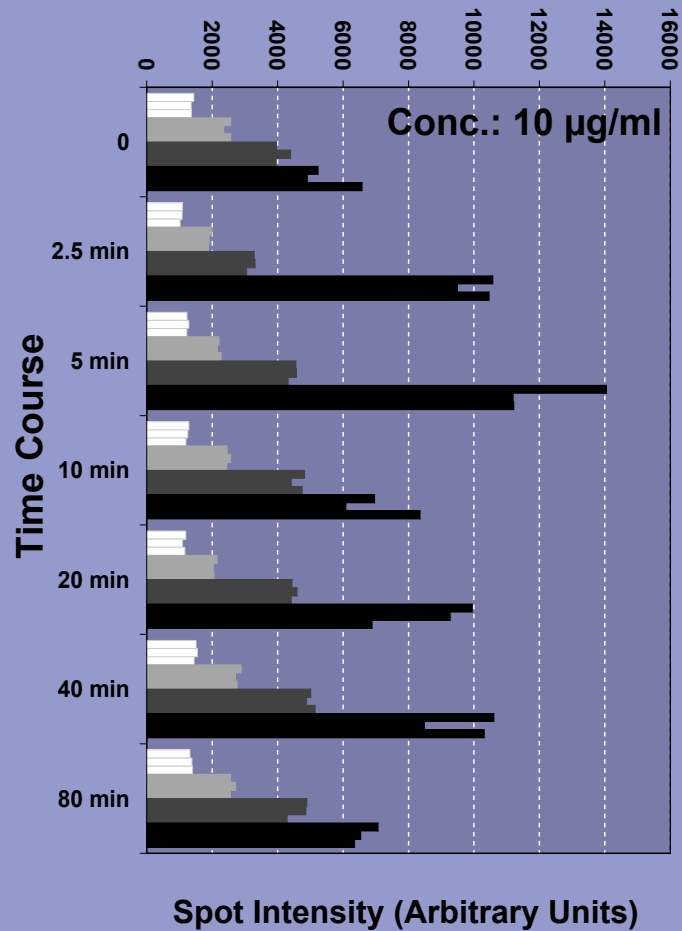


www.kinexus.ca

Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

Cell Line: HepG2 Human Liver Carcinoma

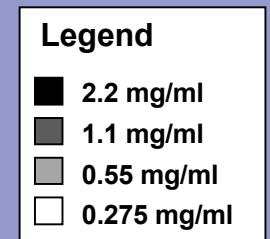
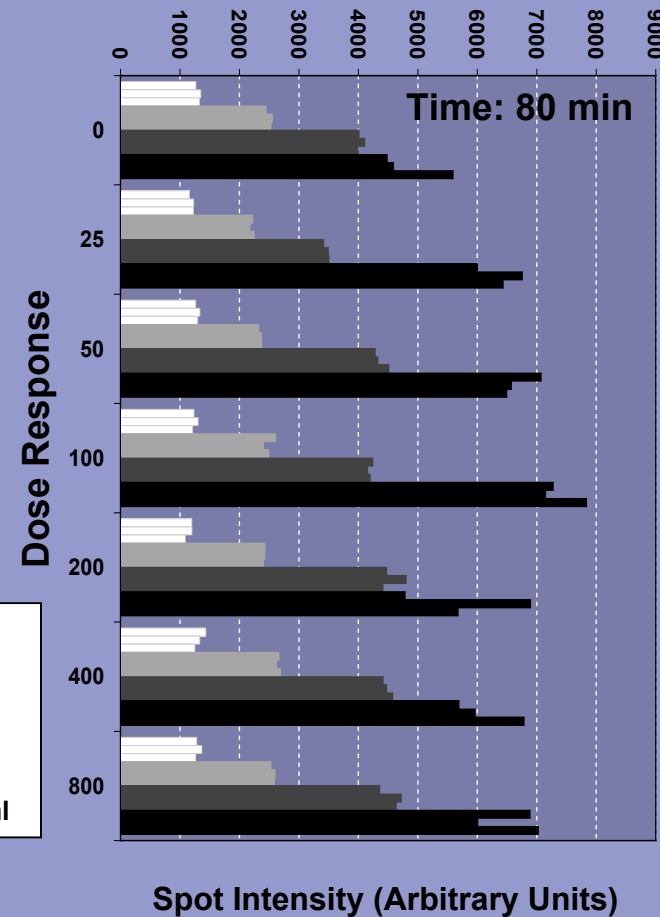
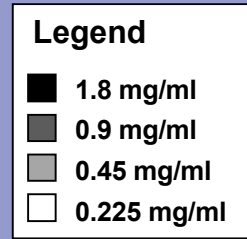
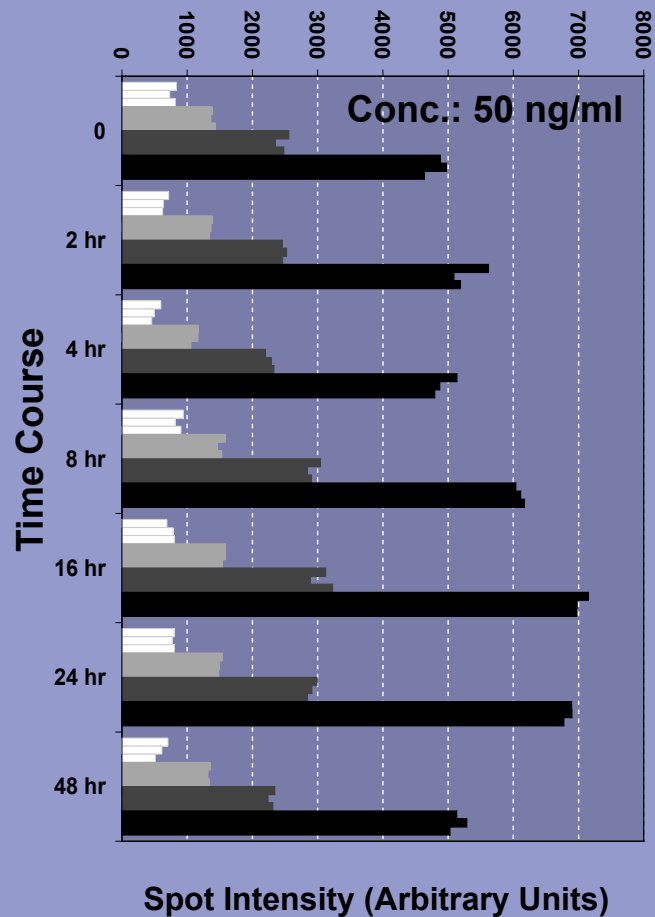
Treatment: Insulin



Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

Cell Line: A549 Human Lung Carcinoma

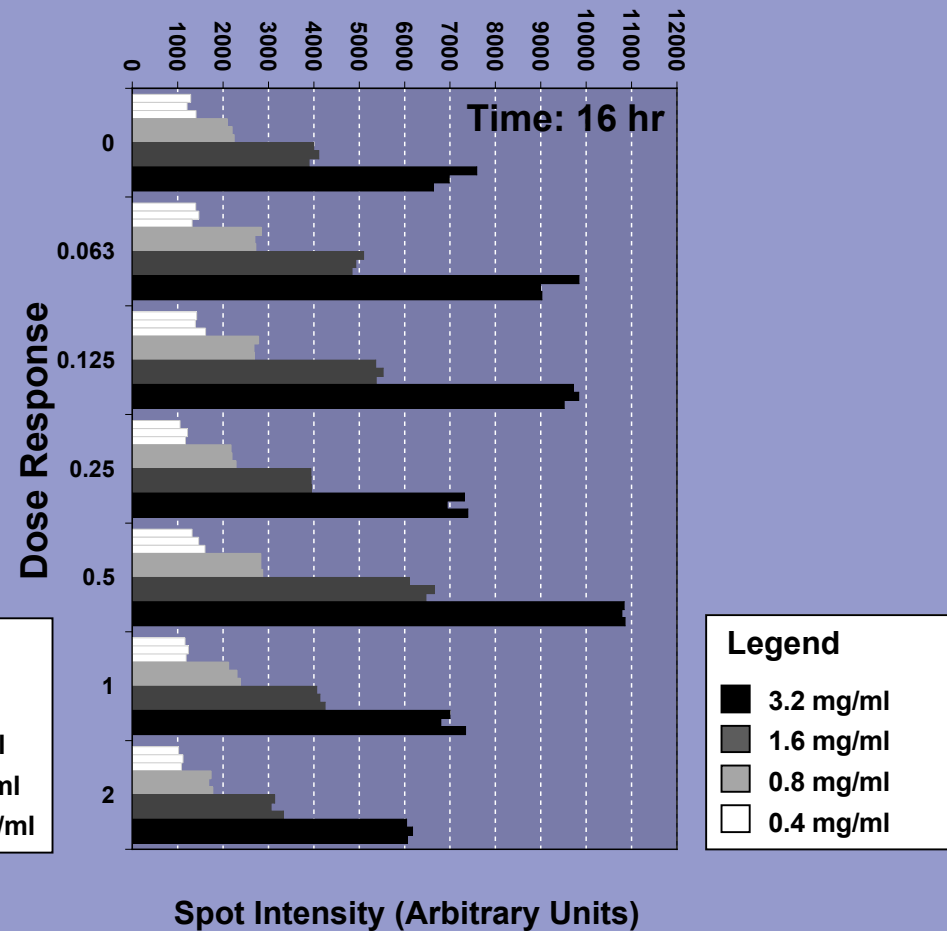
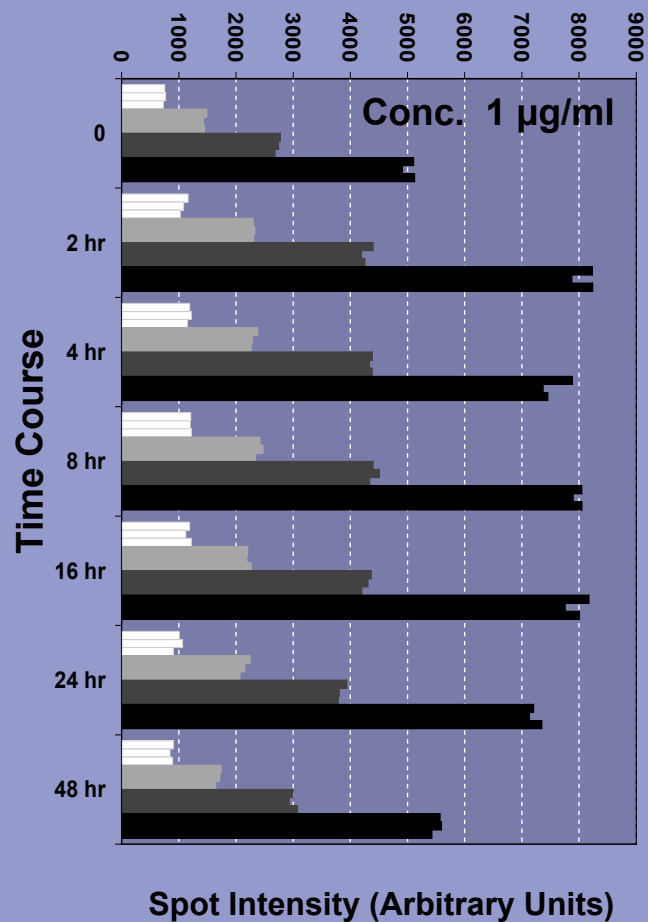
Treatment: Interferon-gamma



Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

Cell Line: HL60 Human Promyelocytic Leukemia

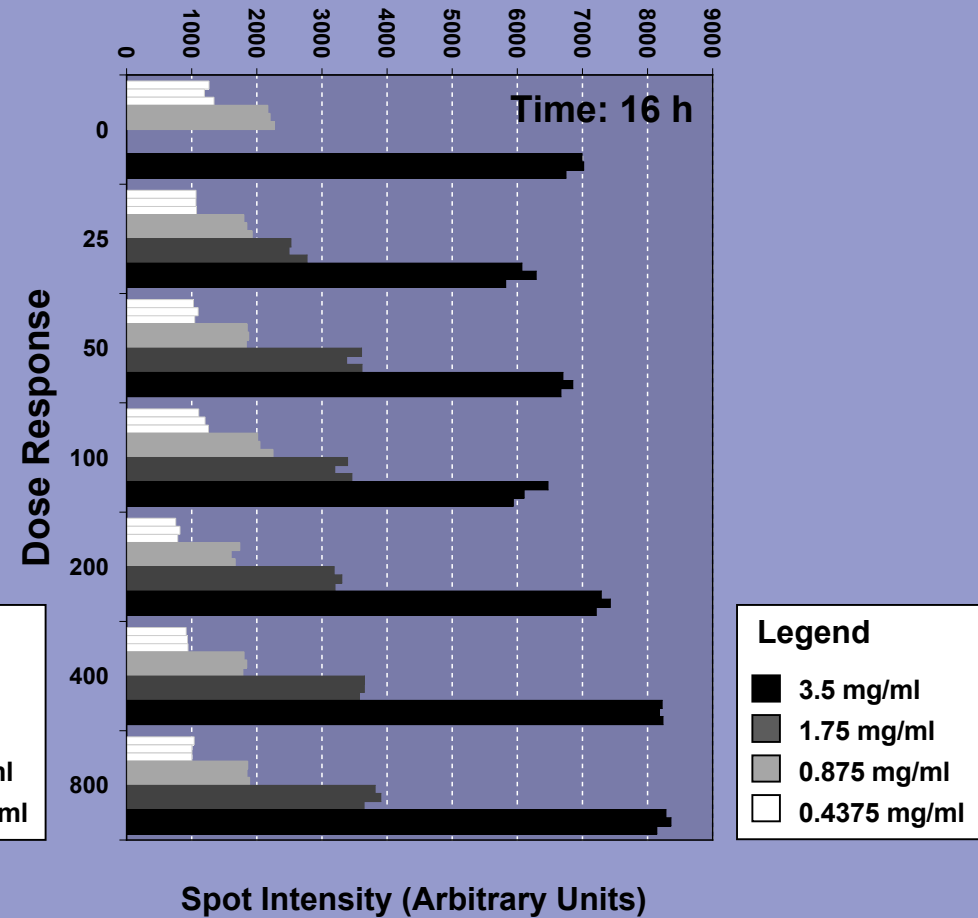
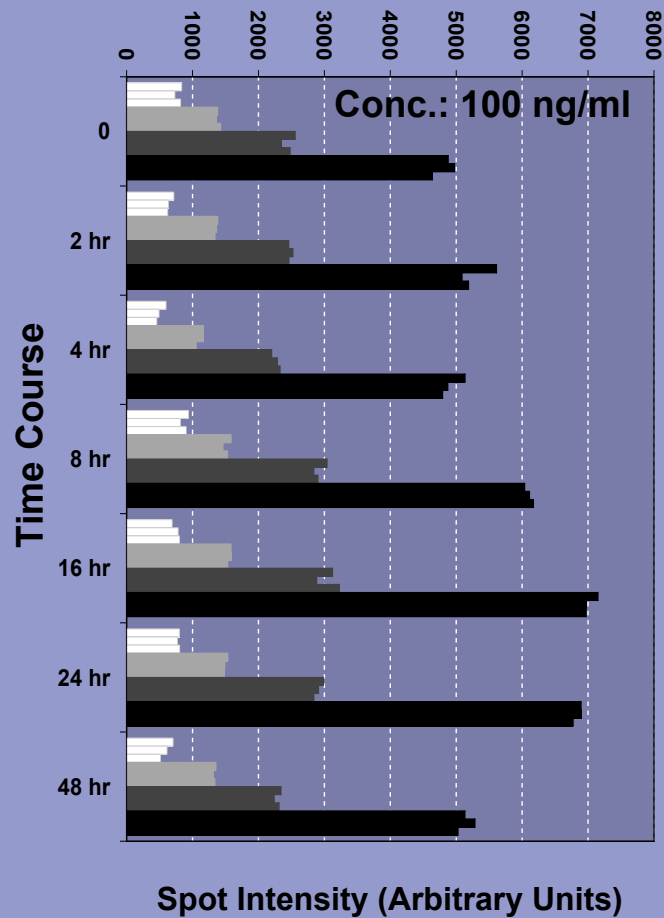
Treatment: Staurosporine



Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

Cell Line: HCT-116 Human Colon Carcinoma

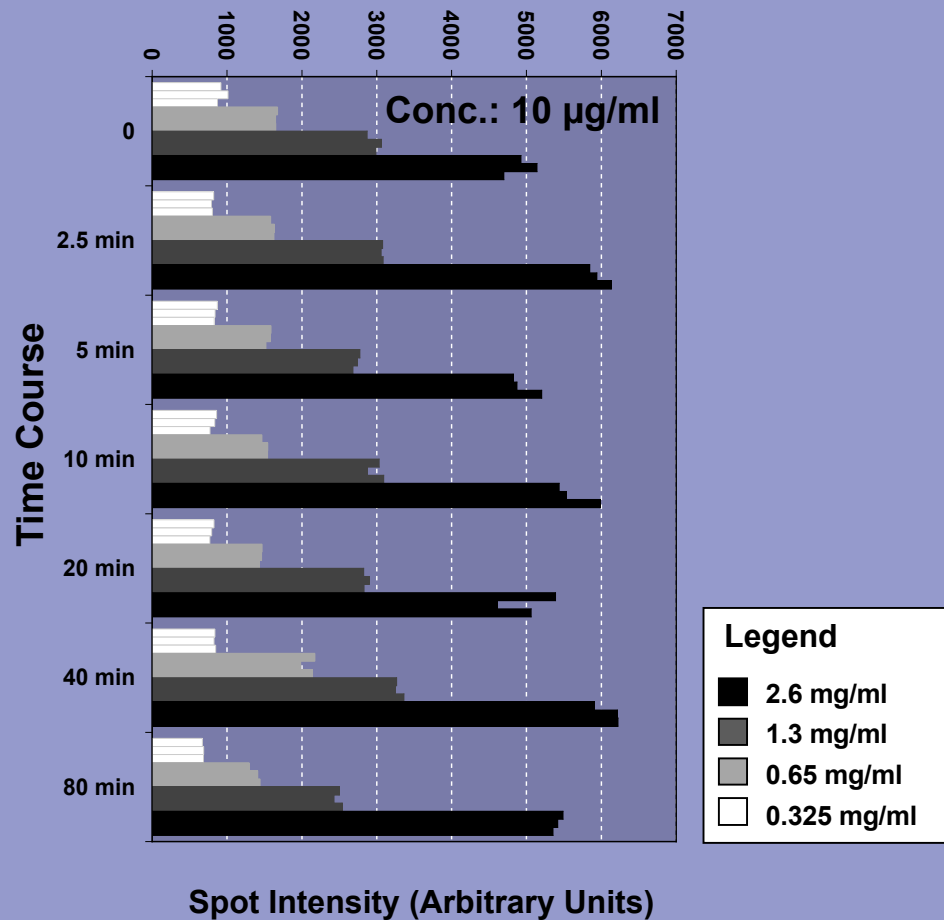
Treatment: Nocodazole



Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

Cell Line: HEK-293 Human Embryonic Kidney

Treatment: Anisomycin



www.kinexus.ca

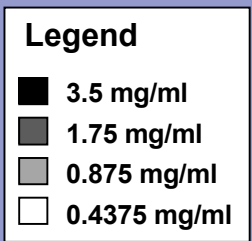
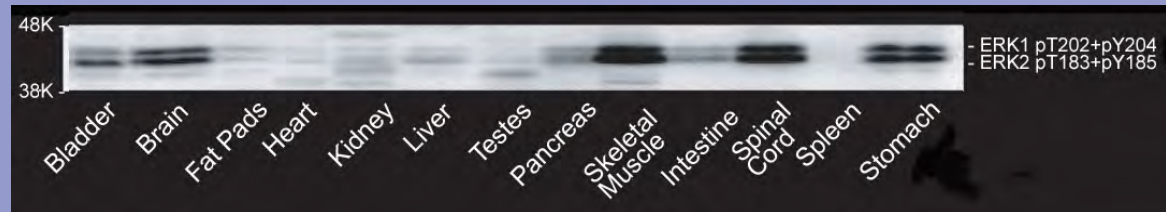
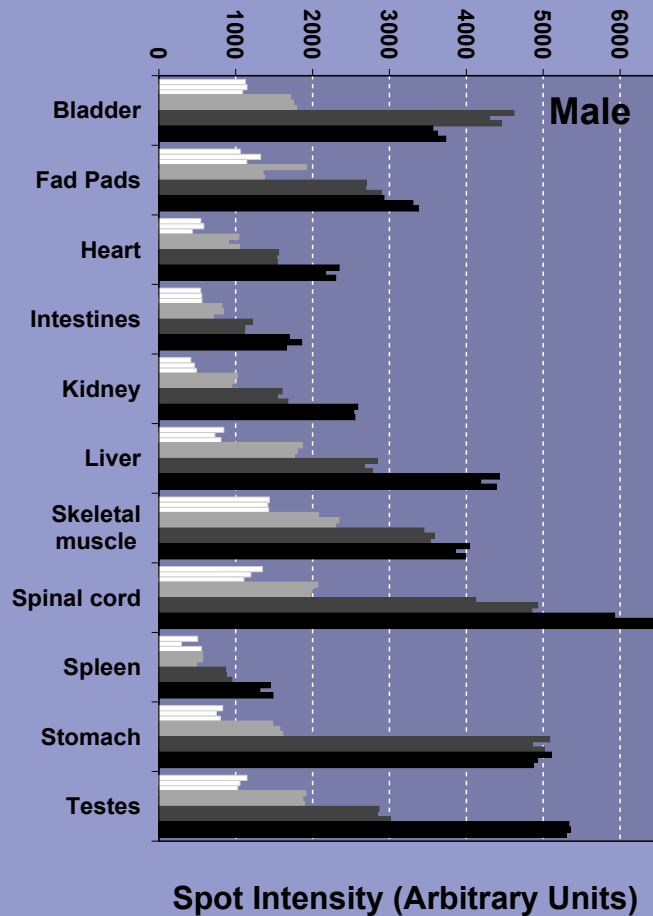
KINEXUS



Kinex™ Reverse Lysate Microarray KRLM-1.0

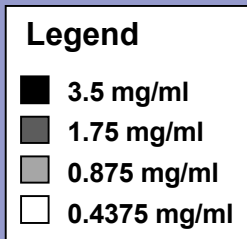
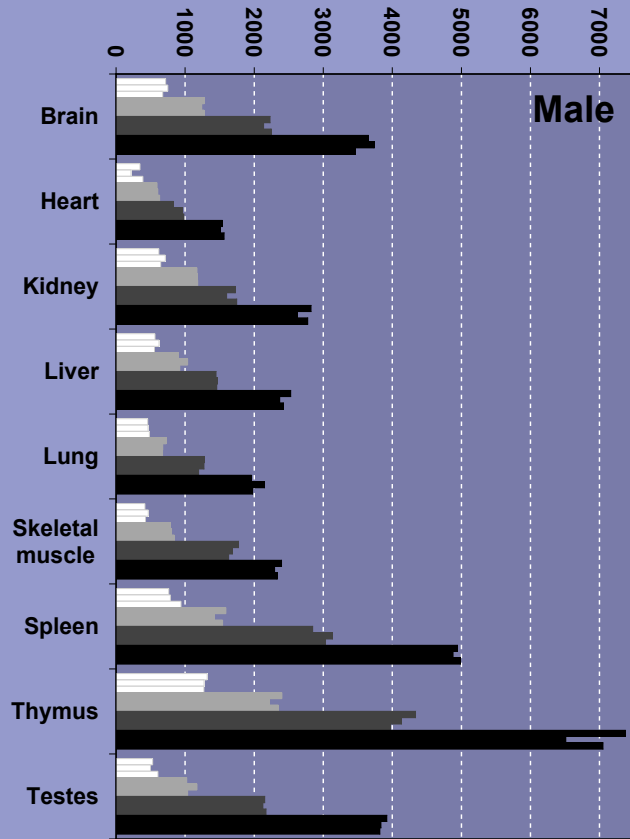
Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

Rhesus Monkey Tissues - Untreated

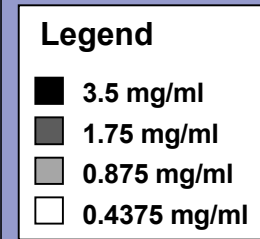
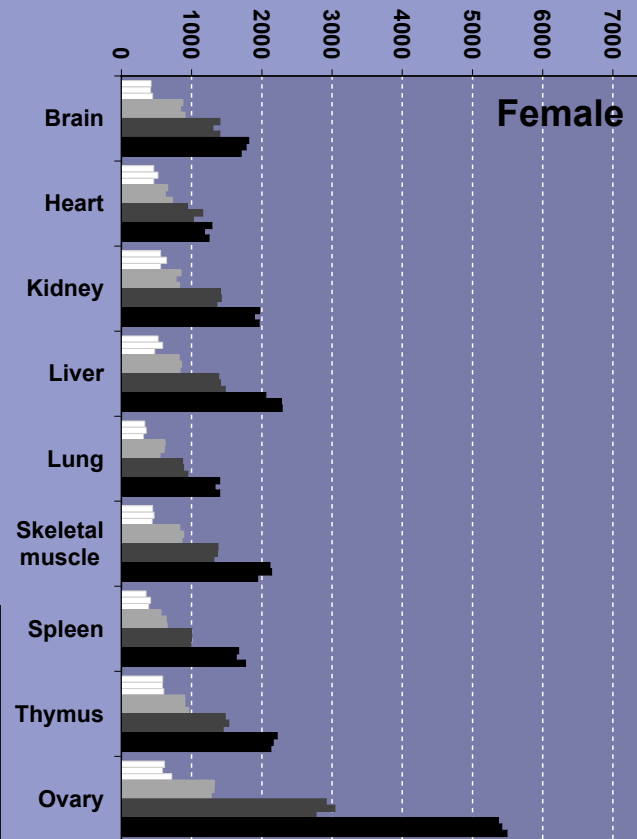


www.kinexus.ca

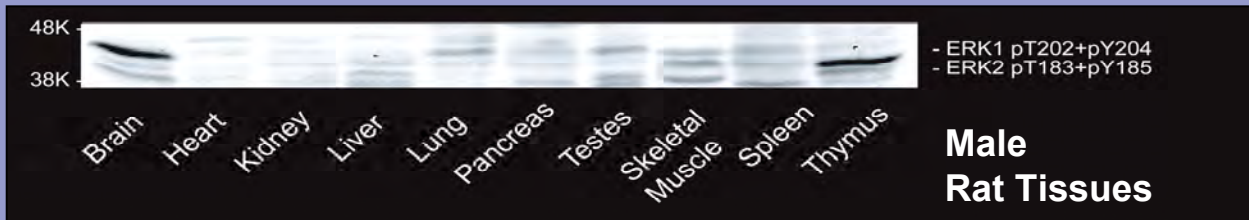
Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation Rat Tissues - Untreated



Spot Intensity (Arbitrary Units)



Spot Intensity (Arbitrary Units)



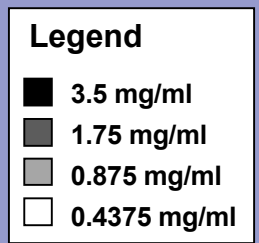
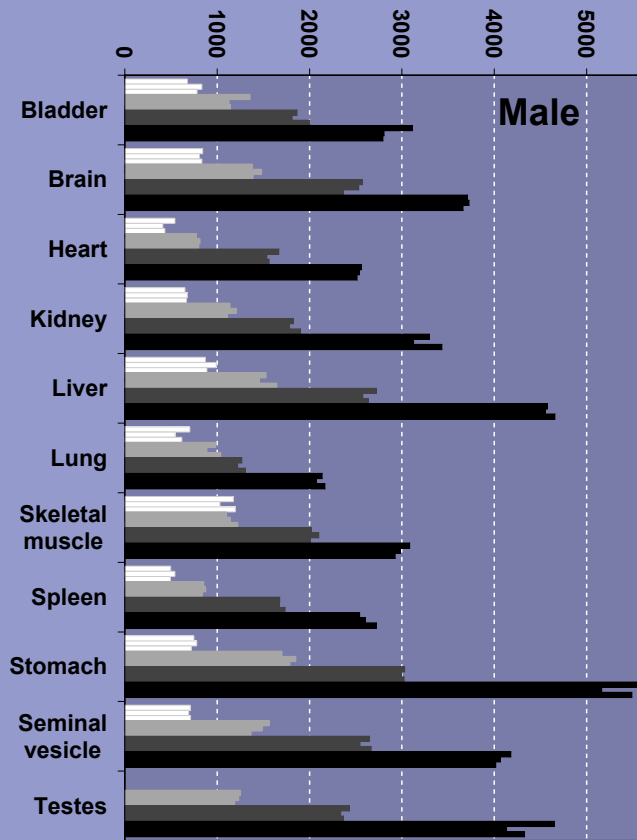
Male Rat Tissues

www.kinexus.ca

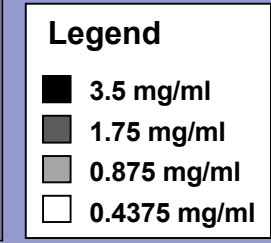
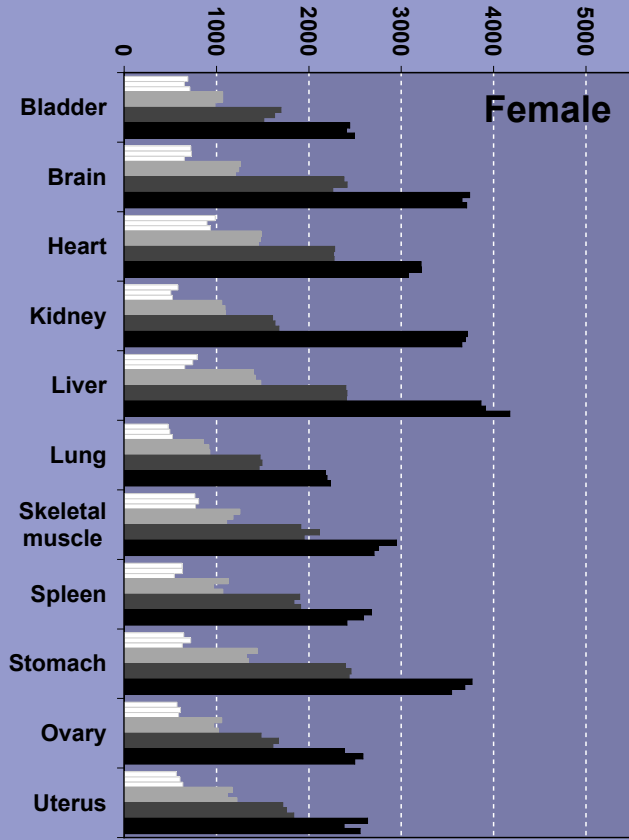


Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

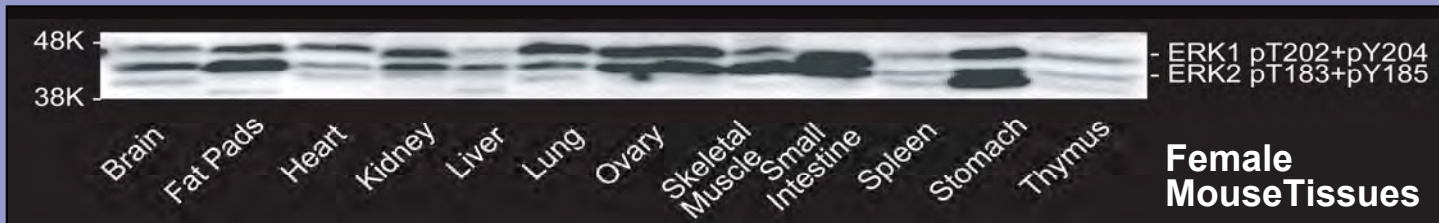
Mouse Tissues - Untreated



Spot Intensity (Arbitrary Units)



Spot Intensity (Arbitrary Units)

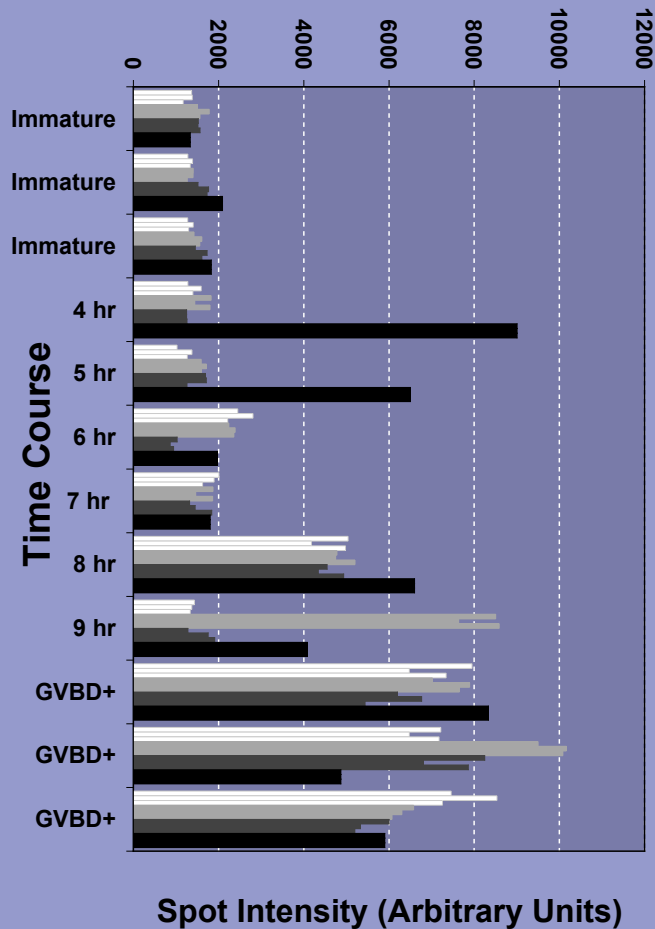


Female Mouse Tissues

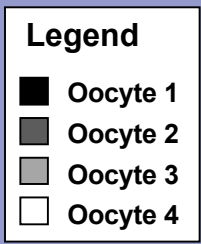
Extracellular Reg. Kinases 1/2 (ERK1/2) T202+Y204/T183+Y185 Phosphorylation

Primary Cell: *Xenopus laevis* Frog Oocyte - Stage VI

Treatment: Progesterone



The results from the microarray analysis of 48 individual frog oocytes are shown at only one dilution in triplicate. Note the high degree of variation in Erk2 phosphorylation despite the fact that all of the oocytes were from the same frog.



www.kinexus.ca