PREPARATION OF CELL AND TISSUE PELLETS

A sufficient number of cells (>2 x 106 cells) should be provided for each sample to be subjected to KAM-1325 analysis.
If Kinetworks™ multi-immunoblotting is desired for validation of the KAM-1325 results, the number of cells required is ten-fold higher (>2 x 107 cells).

A) Adherent Cells

1. Remove the medium and rinse the cells in dish with ice-cold PBS once;
2. Detach cells with trypsin as one does in passaging cells or scrape the cells with a rubber policeman, followed by the addition of equal volume of medium;
3. Collect cells in a 15-ml conical tube and centrifuge at 500 x g for 2 minutes at 4°C in a swinging bucket benchtop centrifuge;
4. Wash the pellet twice with ice-cold PBS thoroughly, (the presence of serum from medium could skew the protein assay) and remove as much PBS as possible (the presence of liquid residue dilutes the sample and may also result in the damage of cells during freezing process); and
5. Freeze the pellets for shipping. Pellets must be shipped on dry ice.

B) Suspended Cells

Simply follow Steps 3-5 above for "A) Adherent Cells" and freeze the cell pellet immediately. Pellets must be shipped on dry ice.

C) Tissues

Freshly harvested tissues are preferred if possible. When harvesting, the tissues should be cut into small pieces on the surface. Wrap the tissues individually in tinfoil and snap freeze them in liquid nitrogen for 10 minutes before storing them at -80 °C. The tissues should be shipped on dry ice.