SIMPLE WESTERN ASSAY (automatic Western Blot)

ABOUT SIMPLE WESTERN ASSAYS

Simple Western is a capillary-based nano-proteomic immunoassay and an automatic Western Blot with high throughput capability. It is a hands-free, gel-free, mess-free alternative to the labor-intensive traditional Western Blot.

CAPABILITIES

- Absolute or relative quantitation of any protein of interest
- Quick assay development for novel antigens/targets/antibodies
- Clear separation of proteins based on either sizes (MW ≤ 440 KDa) or electric charges (pI)
- Reliable quantitation of multiple target proteins / biomarkers in samples of limited amount, including fine needle aspirates, laser capture microdissection, cell culture, etc, from animals or human patients
- Real-time toxicological and PK/PD monitoring drug candidates in experimental animals
- Monitoring signaling transduction events in limited samples (as low as 25 cells), great for those from stem cells, bone marrow cells, sorted cells, etc.
- Discovery and validation of biomarkers to support developing predictive and companion diagnostics
- Characterization of monoclonal and polyclonal antibody binding affinities
- Characterization of relative specificity of antibody to the target protein of interest
- Quantitation of protein phosphorylation even without phospho-specific antibodies
- Phospho-protein profiling and quantitation of post-translational protein modifications using pan antibodies (after protein separation by charge/pI value) or using isoform specific antibodies (after protein separation by size/MW)
- Screening test compounds for effects on degradation, expression or modifications of any protein or signaling molecule

SAMPLE REQUIREMENTS

- Samples should be BSL- I or II
- Samples (about > 10 µl of 2 - 4 mg/ml protein) should be submitted
- Any extraction procedures are the responsibility of the researchers. Special arrangements can be made in advance
- Submit samples in a minimum-binding microfuge tubes, accompanied by a template that outlines the sample order
- Ship samples on dry ice

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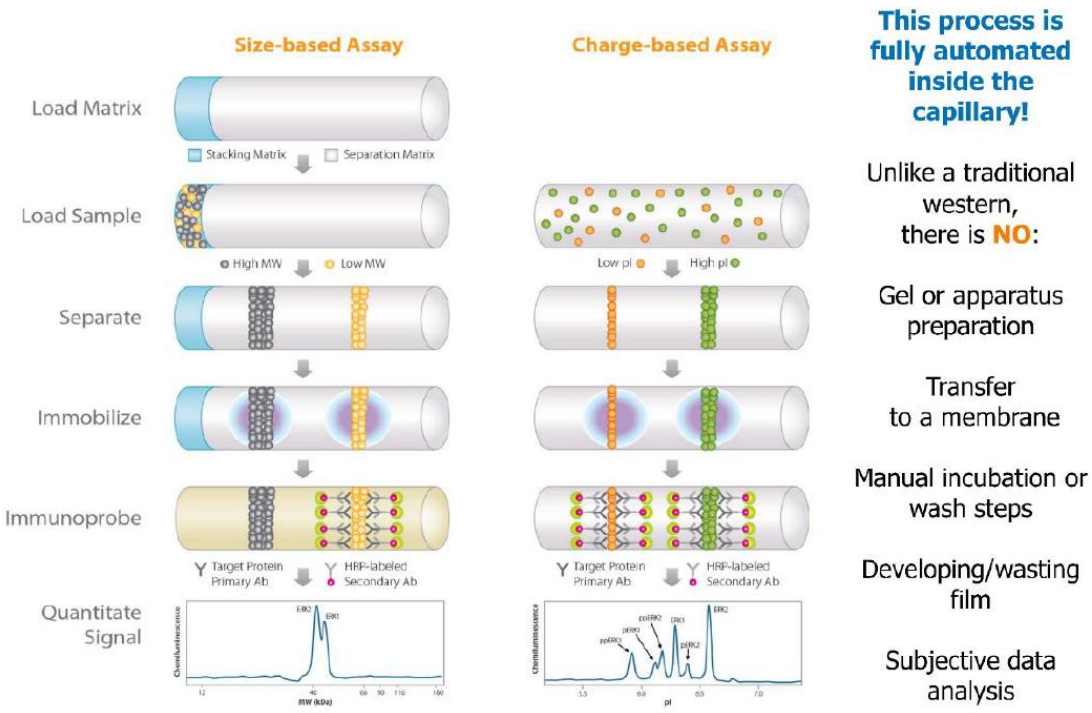
HOW DOES OUR ASSAY WORK?



SERVICE FEATURES

- **Highly sensitive and Automatic** Western blot to examine proteins in a minimal amount sample
- **Multiplex**
- **Qualitative and quantitative**
- **Highly reproducible**
- **Flexible**: we can quantify any protein of interest, even of rare or low abundance, in samples of limited amount (as low as 25 cells),
- **Minimal samples**: Samples can be cells from live animals and patients including fine needle aspirates (FNA), laser capture microdissection (LCM), FFPE samples, sorted cells, as well as serum, plasma, saliva, urine, etc.
- **High throughput**: 24 or 96-well format
- **State-of-art platforms**: Wes, Peggy.
- **Timely data delivery**: 1-4 weeks or sooner dependent upon receiving test samples/ primary antibodies
- **20+ years of experience**: Expert data analysis and interpretation, high quality scientific and technical support
- **Optional tests**
  - ✓ We have a library of >**4000 validated antibodies** to examine target proteins
  - ✓ We offer ~100 human cancer cell lines and various primary human cells for testing drugs and biologicals for specific projects
  - ✓ we can do homogenization and preparation of tissue lysates
  - ✓ we can do protein concentration measurement/ normalization using assays such as BCA

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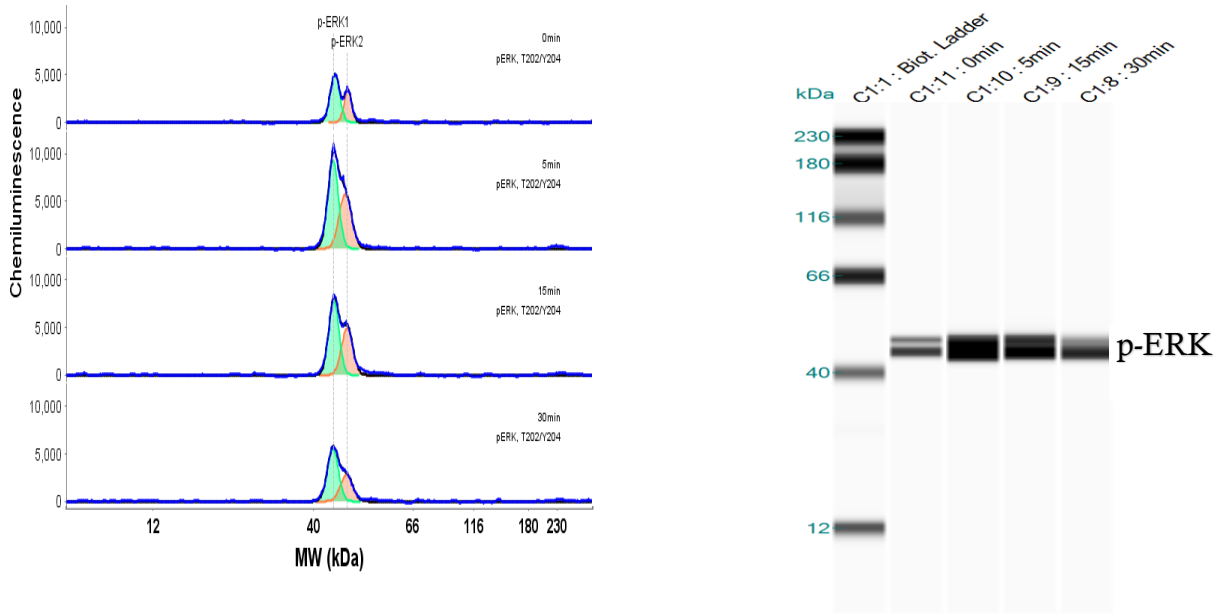
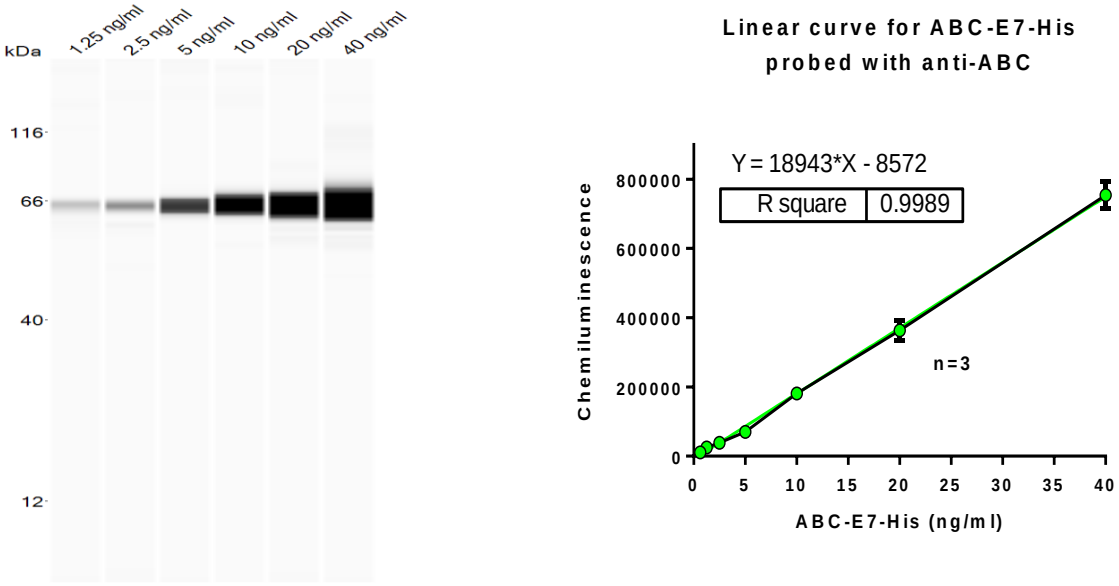
DATA EXAMPLES



Example 1. Time kinetics of anti-IgM induced protein phosphorylation in Raji B lymphoma. Log phase growing Raji cells were stimulated with anti-IgM for 0, 5, 15, and 30 min. Cell lysates were prepared for Simple Western analysis of ERK/MAPR phosphorylation. Left, electropherogram view. Right, Western Blot lane view.



Example 2. Titration of ABC-E7-his and detection with anti-ABC antibody. Left, Western Blot lane view; Right, linear regression of standard curve for absolute quantitation.

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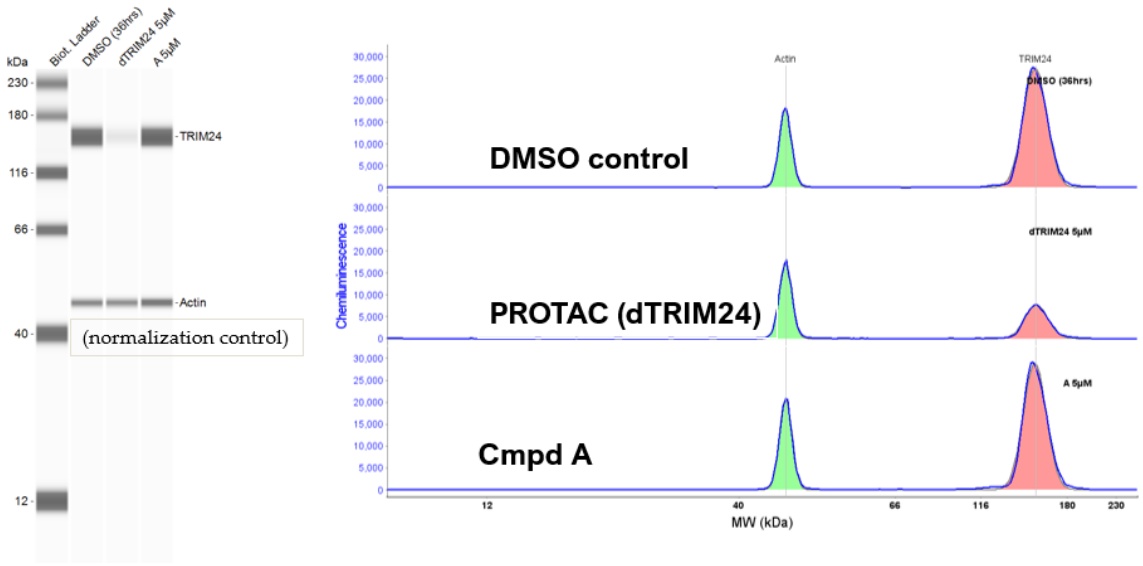
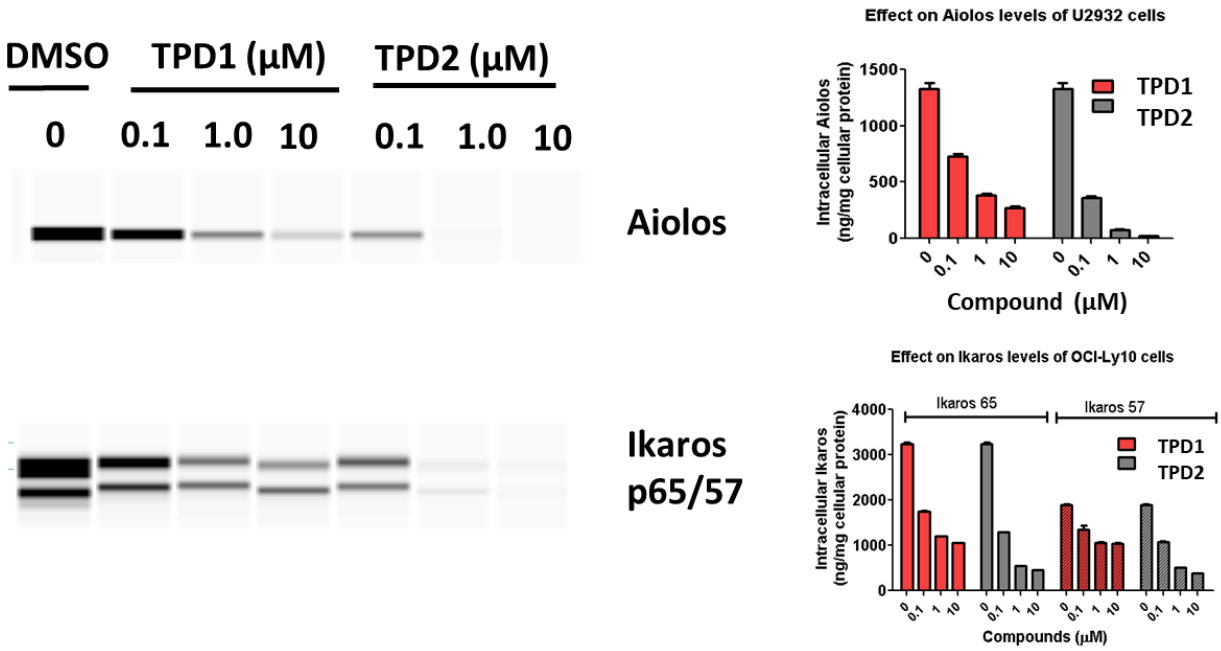


Example 3. Screening of PROTAC candidates. MCF-7 cancer cells were treated with test compounds and then cell lysates were analyzed with Simple Western. Electropherogram to show bands (left) or peaks (right) or of β-actin and TRIM24. β-actin was used as loading control.



Example 4. Quantitation of IKZF1/3 degradation in human lymphoma cells treated with a molecular glue compound. Left, lane view of Aiolos and Ikaros protein expression in U2932 cells; Right, quantification of each target protein.

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