

## Anti-phosphorylated Histone H2AX ( $\gamma$ -H2AX) Polyclonal Antibody

Catalog #: 4418-APC-100

Size: 100  $\mu$ l

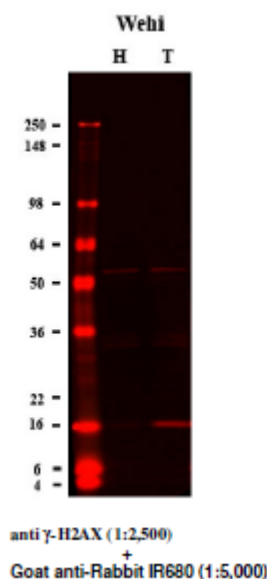
**Background:** Histone H2AX is a 14 kDa ubiquitous member of the H2A histone family that contains an evolutionarily conserved SQ motif at the C-terminus in eukaryotes. Serine 139 within this motif becomes rapidly phosphorylated to yield a form known as  $\gamma$ -H2AX in response to double-strand DNA damage and apoptosis. Phosphorylation reaches half its maximum between 1-3 minutes after DNA damage occurs, and hundreds to several thousand molecules of  $\gamma$ -H2AX are present per double-strand break. This antibody is unique in only detecting double-strand DNA breaks.

**Physical State:** Peptide affinity purified rabbit polyclonal antibody raised against synthetic phosphorylated peptide (CKATQA[pS]QEY). Provided at 0.5  $\mu$ g/ $\mu$ l in phosphate buffered saline buffer with 50% glycerol.

**Specificity:** Recognizes human and mouse  $\gamma$ -H2AX.

**Storage:** -20°C.

**Applications:** Suitable for Western blotting and immunocytochemistry (IC). For Western blot analysis, a starting dilution of 1:1,000 – 1:2,500 is recommended, whereas for IC, a starting dilution of 1:100 is recommended. Empirical determination of antibody concentration is required for optimal results.



**Fig. 1. Western blot analysis of Wehi cells using Trevigen's anti-phosphorylated Histone H2AX antibody.** Wehi cells were treated with (T) and without (H) 25  $\mu$ M etoposide for 4 hours. Cells were lysed in Tris-Glycine SDS sample buffer at the concentration  $1 \times 10^7$  cells/ml and 10  $\mu$ l of each lysate were loaded per well of 4-20% Tris-Glycine gel. Proteins were transferred onto an Immobilon FL membrane and  $\gamma$ -H2AX was detected with anti-phosphorylated Histone H2AX antibody (Cat# 4418-APC-100) followed by IR800-conjugated secondary anti-body (Licor). Membranes were scanned using an Odyssey Infrared Imaging System (Licor).

**References:**

1. Mahadevaiah, S.K., J.M. Turner, F. Baudat, E.P. Rogakou, P. de Boer, J. Blanco-Rodriguez, M. Jasin, S. Keeney, W.M. Bonner, P.S. Burgoyne. 2001. Recombinational DNA double-strand breaks in mice precede synapsis. *Nature Gen* **27**:271-6.
2. Rogakou, E.P., W. Nieves-Neira, C. Boon, Y. Pommier, and W.M. Bonner. 2000. Initiation of DNA fragmentation during apoptosis induces phosphorylation of H2AX histone at serine 139. *J Biol Chem* **275**:9390-5
3. Rogakou, E.P., C. Boon, C. Redon, W.M. Bonner. 1999. Megabase chromatin domains involved in DNA double-strand breaks in vivo. *J Cell Biol* **146**:905-16.
4. Rogakou, E.P., D.R. Pilch, A.H. Orr, V.S. Ivanova, W.M. Bonner. 1998. DNA double-stranded breaks induce histone H2AX phosphorylation on serine 139. *J Biol Chem*. **273**:5858-68.

**Related Products:**

Catalog #	Description	Size
6370-MC-100	Anti-human/murine-Cytochrome C	100 µg
6380-MC-100	Anti-human/murine-Holocytochrome C	100 µg
2291-MC-100	Anti-human-Bcl-2 mAb (clone YTH-8C8)	100 µg
2281-MC-100	Anti-human-Bax mAb (clone YTH-6A7)	100 µg
6361-PC-100	Anti-human/mouse-PBR polyclonal	100 µl
4335-MC-100	Anti-PAR polymer mAb (10HA)	100 µl
4336-BPC-100	Anti- PAR polymer polyclonal	100 µl
4338-MC-50	Anti-human/murine-PARP mAb (clone C2-10)	50 µg

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