

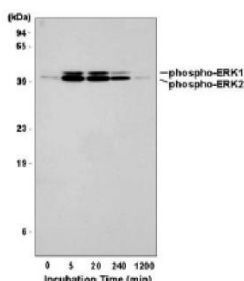
| | |
|--------------------------|-----------------------------------------------------------------------------------------------------------------------|
| Cat. No: | ABP-83922 |
| Conjugate: | Unconjugated |
| Size: | 100 ug |
| Clone: | Poly |
| Concentration: | 1mg/ml |
| Host: | Rabbit |
| Isotype: | IgG |
| Immunogen: | Peptide derived from the protein area including conserved pT-E-pY motif of activated Erk 1,2 |
| Reactivity: | Hu, Ms, Rt |
| Applications: | Western blotting:1:1000 Immunohistochemistry:1:100-500 Immunofluorescence: 1:100-500 Immunocytochemistry: 1:100 |
| Molecular Weight: | 42,44kDa |

Background: The protein encoded by this gene is a member of the MAP kinase family. MAP kinases, also known as extracellular signal-regulated kinases (ERKs), act in a signaling cascade that regulates various cellular processes such as proliferation, differentiation, and cell cycle progression in response to a variety of extracellular signals. This kinase is activated by upstream kinases, resulting in its translocation to the nucleus where it phosphorylates nuclear targets. Alternatively spliced transcript variants encoding different protein isoforms have been described.

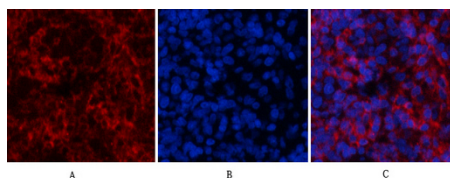
Form: liquid

Buffer: 20 mM Tris Hcl, pH 8 10 mg/mL BSA - 0,05% Sodium Azide

Storage: The antibody can be stored at 2° - 8° C for 1 month without detectable loss of activity. Antibody can also be aliquotted and stored frozen at -20° C to -70° C in a manual defrost freezer for six months without detectable loss of activity. Avoid repeated freeze-thaw cycles.

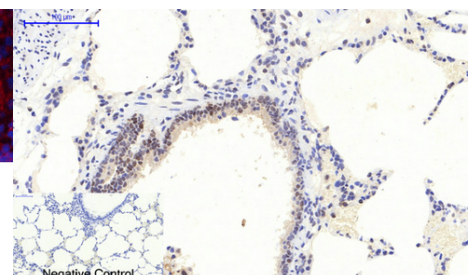


Detection of ERK1 and ERK2 phosphorylated at T202/Y204 and T185/Y187, respectively. Human HeLa cells were incubated with 200 nM PMA for the indicated times. Total cell lysates in gel sample buffer were resolved by SDS-PAGE.



Immunofluorescence analysis of rat-lung tissue.

- 1, ERK 1/2 (phospho Thr202/Y204) Polyclonal Antibody (red) was diluted at 1:200 (4°C, overnight).
 - 2, Cy3 labeled Secondary antibody was diluted at 1:300 (room temperature, 50min).
 - 3, Picture B: DAPI (blue) 10min.
- Picture A: Target.
Picture B: DAPI.
Picture C: merge of A+B



Immunohistochemical analysis of paraffin-embedded Rat-lung tissue.

- 1, ERK 1/2 (phospho Thr202/Y204) Polyclonal Antibody was diluted at 1:200 (4°C, overnight).
- 2, Sodium citrate pH 6.0 was used for antibody retrieval (>98°C, 20min).
- 3, Secondary antibody was diluted at 1:200 (room temperature, 30min). Negative control was used by secondary antibody only

WESTERN BLOT (WB) PROTOCOL

Western immunoblotting solutions:

Wash buffer: 1x Tris Buffered Saline (TBS); 0.1% Triton X-100 -

Blocking buffer: 1xTBS; 0.1% Triton X-100; 5% BSA (used with the primary antibody)

For western blots, incubate the membrane with antibody diluted in blocking buffer 2 hours at room temperature

IMMUNOCYTOCHEMISTRY (ICC) PROTOCOL - INSTRUCTION MANUAL

1. Coat coverslips with 1% gelatin-coating solution for 2 hours at room temperature (RT); rinse with distilled water, and let to dry overnight. Before plating the cells, wash the coated coverslips briefly with PBS. 2. Fix the cells with 4% paraformaldehyde solution (in PBS, pH 7.2), for 15 min at RT. 3. Wash 2 x 3 min with PBS. 4. Permeabilize the cells with 0.1% Triton X-100 solution (in PBS, pH 7.2) for 5 min on ice. 5. Wash 2 x 3 min with PBS. 6. Incubate the cells in blocking buffer (0.3M glycine in PBS, 2% BSA) for 30 min at RT. 7. Incubate the cells with primary antibody: anti-phospho Erk 1,2 clonal antibody at the dilution of 1:100 - in antibody dilution buffer (PBS, 1% BSA) for 1 hour at RT in humid chamber. 8. Wash 2 x 3 min with PBS. 9. Apply the secondary antibody used at 1:300 in antibody dilution buffer, and cells were incubated for 1 hour at RT in dark). 10. Wash 3 x 3 min with PBS. 11. Rinse once with distilled water. 12. Mount the slide for observation, with a drop of anti-fade mounting medium.

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