www.tag-tools.de

tag-tools GmbH
Oberdorfstr. 16B
78465 Konstanz
Germany
E-Mail: mail@tag-tools.de
www.tag-tools.de

HA-tag control-lysate

Description

The HA-tag control-lysate was generated from transfected HEK293 cells, expressing a HA-tagged protein. The HA-tag corresponds to a peptide of the hemaglutinin protein of the influenza virus. The peptide sequence of the HA-tag (YPYDVPDYA) is not present in any mammalian protein.
To generate the HA-tag control-lysate, HEK293 cells were transiently transfected with a mammalian expression vector coding for a murine cytoplasmic protein harbouring a double HA-tag at the carboxy-terminus. A whole cell lysate was prepared from these transiently transfected cells. Besides all other cellular proteins, this whole cell lysate contains the HA-tag protein, which shows an apparent molecular weight of ~45 kDa upon SDS-PAGE.

In addition to the HA-tag control-lysate, a mock-lysate was prepared from HEK293 cells, which were transfected with the empty expression vector.

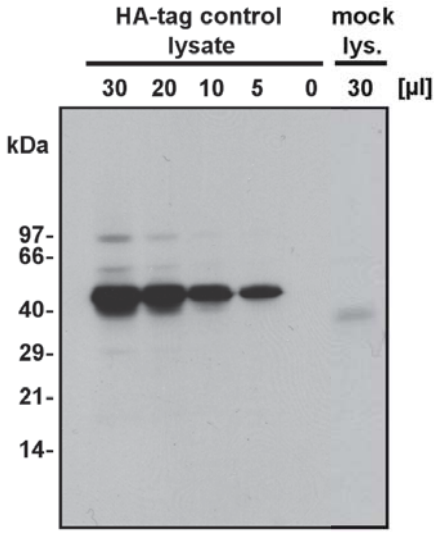
Product

Whole cell lysate:
50 µl
Cell line:
HEK293 cells
Tagged protein:
Murine cytoplasmic protein with a carboxy-terminal double HA-tag.
MW:
~45 kDa
Lysis buffer:
60 mM Tris/HCL pH 6.8, 2% SDS,
5% mercaptoethanol,
50 µg/ml bromophenolblue, 10% glycerol
Application:
For SDS-PAGE/Western Blotting
20 µl of the lysates should be applied per lane of a 10% polyacrylamide-gel.

Shipping and storage

Shipping:
The lysate is shipped in cold case.
Storage:
Lysate is stable for 1 week at 4°C.
For prolonged storage, the lysate should be stored at -80°C. Aliquot to avoid repeated freezing and thawing. At -80°C, the product is stable for at least 1 year from shipment.

Application in quality control



Control cell lysates
anti-HA-tag blot

Western Blotting using the HA-tag control-lysate and the mock-lysate.
HEK293 cells were transiently transfected either with an expression vector encompassing the cDNA of a HA-tag protein (HA-tag control-lysate) or with the empty expression vector (mock-lysate; mock lys.).
The indicated amounts of the HA-tag control-lysate (30, 20, 10, 5, or 0 µl) or 30 µl of the mock-lysate were separated by SDS-PAGE using a 10% polyacrylamide-gel and then transfered onto a PVDF membrane. The membrane was probed with the monoclonal anti-HA-tag antibody from *tag*-tools (clone TT7; dilution 1:2,000).
The HA-tag bound primary antibody was detected by incubation with HRP-coupled goat-anti-mouse antibody (dilution 1:10,000) and visualized using chemiluminescence.
The X-ray film was exposed for 10 seconds.
Whereas a specific ~45 kDa band is detected in the HA-tag control-lysate at all applied volumes (5 - 30 µl), no binding of the anti-HA tag antibody to cellular proteins can be detected in the lane, where 30 µl of the mock-lysate was applied.

Use

The HA-tag control-lysate and the mock-lysate are for quality control in research applications only. Not for diagnostic or therapeutic purpose.