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### Product no AS15 2962

## RBOHD | Respiratory burst oxidase homolog protein D

#### **Product information**

Immunogen KLH-conjugated peptide chosen from Arabidopsis thaliana RBOHD sequence, UniProt: Q9FIJ0, TAIR: AT5G47910

Host Rabbit

Clonality Polyclonal

**Purity** Immunogen affinity purified serum in PBS pH 7.4.

Format Lyophilized

Quantity 50 μg

**Reconstitution** For reconstitution add 50 μl of sterile water

Store lyophilized/reconstituted at -20°C; once reconstituted make aliquots to avoid repeated freeze-thaw cycles. Please

remember to spin the tubes briefly prior to opening them to avoid any losses that might occur from material adhering to

the cap or sides of the tube.

## Application information

Recommended dilution 1:1000 (WB)

Expected | apparent

104 kDa

**Confirmed reactivity** Arabidopsis thaliana, Solanum sp.

Predicted reactivity

Brassica napus, Citrus sinensis, Citrullus colocynthis, Cucumis melo, Fragaria ananassa, Gossypium arboreum, Hordeum vulgare, Manihot esculenta, Morus notabilis, Nicotiana tabacum, Oryza sativa, Picea abies, Ricinus communis, Sinensis osbeck, Solanum tuberosum, Theobroma cacao, Triticum aestivum, Zea mays, Zostera marina Species of your interest not listed? Contact us

Not reactive in Marchantia polymorpha, Medicago sativa

Additional information For solubilization we recommend 65°C for 5 min, Sample boiling is not recommended

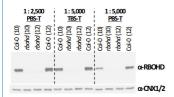
Selected references

Wang et al. (2021) Arabidopsis PUB2 and PUB4 connect signaling components of pattern-triggered immunity. New Phytol. 2021 Dec 17. doi: 10.1111/nph.17922. Epub ahead of print. PMID: 34918346 Lee et al. (2020). Regulation of reactive oxygen species during plant immunity through phosphorylation and ubiquitination of RBOHD. Nat Commun. 2020 Apr 15;11(1):1838. doi: 10.1038/s41467-020-15601-5. Jedelská et al. (2019). Tomato Root Growth Inhibition by Salinity and Cadmium Is Mediated By S-Nitrosative Modifications of ROS Metabolic Enzymes Controlled by S-Nitrosoglutathione Reductase. Biomolecules. 2019 Aug 21;9(9). pii: E393. doi: 10.3390/biom9090393.

Otulak-Kozie? et al. (2019). The Respiratory Burst Oxidase Homolog D (RbohD) Cell and Tissue Distribution in Potato-Potato Virus Y (PVYNTN) Hypersensitive and Susceptible Reactions. Int. J. Mol. Sci. 2019, 20(11), 2741.

(innunofluorescence)

# **Application example**



Total proteins were isolated from 10 or 12 seedlings of 7-day-old Arabidopsis thaliana seedlings of Col-0 (wild-type) and rbohd null mutant (Nühse et al., 2007). 16 µl of total proteins were denatured at 65°C for 5 min, separated on an 8 % SDS-PAGE. and transferred for 70 min at 55V using a tank transfer system to nitrocellulose membrane. Blots were blocked with 1xPBS+0.1% Tween 20 + 5 % milk (PBS-T) or TBS + 0.1 % Tween 20 (TBS-T) + 5 % milk for 2 h at room temperature (RT) with agitation. Primary antibodies were diluted as indicated to 1: 2,500 and 1: 5,000 respectively and incubated ON at 4ºC with agitation in 1xPBS-T pr TBS-T + 5 % milk. The primary antibody solutions were decanted, and the blots



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were washed 3 times (7 min. each) in 1xPBS-T or TBS-T at RT with agitation. Blot was incubated with secondary antibody goat anti-rabbit HRP conjugated (AS09 602, Agrisera) diluted to 1: 20, 000 and developed with chemiluminescent detection reagent, according to manufacture's recommendations. Exposure time was 2 min. for X-ray film. As a control part of the same membrane was incubated with anti-CNX1/2 antibodies (AS12 2365, Agrisera).

#### References:

<u>Nühse</u> et al. Quantitative phosphoproteomic analysis of plasma membrane proteins reveals regulatory mechanisms of plant innate immune responses. Plant J. 2007 Sep; 51(5): 931–940.

Courtesy of Nga Nguyen, Kelly Mason and Antje Heese (University of Missouri- Columbia, MO, USA