# **Product Datasheet**

# HSP70/HSPA1A Antibody - BSA Free NBP1-77455

Unit Size: 0.1 ml

Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.

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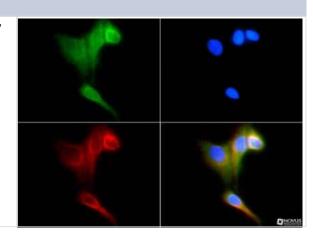
### NBP1-77455

HSP70/HSPA1A Antibody - BSA Free	
Product Information	
Unit Size	0.1 ml
Concentration	1.18 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.05% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	PBS and 30% Glycerol
Product Description	
Host	Rabbit
Gene ID	3303
Gene Symbol	HSPA1A
Species	Human, Mouse
Immunogen	A synthetic peptide made to an internal portion of the human Hsp70 protein (between residues 550-600) [Uniprot: P08107]
Product Application Details	
Applications	Simple Western, Flow Cytometry, Immunocytochemistry/Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin
Recommended Dilutions	Simple Western 1:50, Flow Cytometry 1:200, Immunohistochemistry 1:400, Immunocytochemistry/Immunofluorescence 1:25, Immunohistochemistry-Paraffin 1:400, Immunohistochemistry-Frozen reported in scientific literature (PMID 26755821)

# **Images**

**Application Notes** 

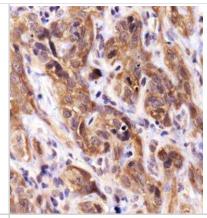
Immunocytochemistry/Immunofluorescence: HSP70/HSPA1A Antibody [NBP1-77455] - HSP70 antibody was tested at 1:25 in HeLa cells with FITC (green). Nuclei were counterstained with DAPI (blue).



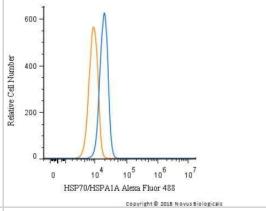
In Simple Western only 10 - 15 uL of the recommended dilution is used per data

point. Separated by Size-Wes, Sally Sue/Peggy Sue.

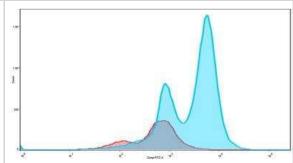
Immunohistochemistry: HSP70/HSPA1A Antibody [NBP1-77455] - Analysis of Hsp70 in human xenograft kidney cancer using DAB with hematoxylin counterstain.



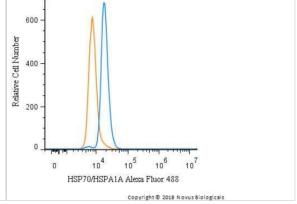
Flow Cytometry: HSP70/HSPA1A Antibody [NBP1-77455] - An intracellular stain was performed on Jurkat cells with HSP70/HSPA1A Antibody NBP1-77455AF488 (blue) and a matched isotype control (orange). Cells were fixed with 4% PFA and then permeablized with 0.1% saponin. Cells were incubated in an antibody dilution of 10 ug/mL for 30 minutes at room temperature. Both antibodies were conjugated to Alexa Fluor 488.



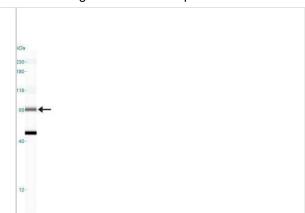
Flow Cytometry: HSP70/HSPA1A Antibody [NBP1-77455] - Single cell suspension generated from B16-F10 tumor excised from C57BL/6 Mouse stained with Hsp70 & FITC secondary (blue) or unstained (red). [Image provided by a varified research professional; see Product Reviews for details on protocol employed].



Flow Cytometry: HSP70/HSPA1A Antibody [NBP1-77455] - An intracellular stain was performed on A549 cells with HSP70/HSPA1A Antibody NBP1-77455AF488 (blue) and a matched isotype control (orange). Cells were fixed with 4% PFA and then permeablized with 0.1% saponin. Cells were incubated in an antibody dilution of 10 ug/mL for 30 minutes at room temperature. Both antibodies were conjugated to Alexa Fluor 488.



Simple Western: HSP70/HSPA1A Antibody [NBP1-77455] - Lane view shows a specific band for Hsp70 in 1.0 mg/ml of HeLa lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.



#### **Publications**

Kines RC, Thompson CD, Spring S et al. Virus-Like Particle-Drug Conjugates Induce Protective, Long-lasting Adaptive Antitumor Immunity in the Absence of Specifically Targeted Tumor Antigens Cancer Immunology Research 2021-06-01 [PMID: 33853825] (In Vivo, FLOW)

Skalina KA, Singh S, Chavez CG et al. Low Intensity Focused Ultrasound (LOFU)-mediated Acoustic Immune Priming and Ablative Radiation Therapy for in situ Tumor Vaccines Sci Rep. 2019-10-29 [PMID: 31664044] (FLOW, Mouse)

#### Details:

Citation used the FITC format of this antibody.

Apavaloaei A, Brochu S, Dong M et al. PSMB11 Orchestrates the Development of CD4 and CD8 Thymocytes via Regulation of Gene Expression in Cortical Thymic Epithelial Cells J. Immunol. 2018-12-19 [PMID: 30567730] (ICC/IF, Mouse)

Bandyopadhyay S, Quinn TJ, Scandiuzzi L et al. Low-Intensity Focused Ultrasound Induces Reversal of Tumor-Induced T Cell Tolerance and Prevents Immune Escape. J. Immunol. 2016-01-11 [PMID: 26755821] (FLOW, IHC-Fr, Mouse)

#### Details:

HSP70 antibody was used for immunofluorescence staining of cryosections from B16-F1 tumor tissues isolated from untreated mice or from mice treated with Low-Intensity Focused Ultrasound/LOFU (FIGURE 4D). The antibody was also used for FLOW on CD452TRP1+ B16 cells from tumors of LOFU-treated and untreated mice (FIGURE 4E).



#### **Procedures**

### Immunohistochemistry-Paraffin protocol for Hsp70 Antibody (NBP1-77455)

HSP70/HSPA1A Antibody: https://www.novusbio.com/products/hsp70-hspa1a-antibody\_nbp1-77455 Immunohistochemistry-Paraffin Embedded Sections

#### Antigen Unmasking:

Bring slides to a boil in 10 mM sodium citrate buffer (pH 6.0) then maintain at a sub-boiling temperature for 10 minutes. Cool slides on bench-top for 30 minutes.

#### Staining:

- 1. Wash sections in deionized water three times for 5 minutes each.
- 2. Wash sections in wash buffer for 5 minutes.
- 3. Block each section with 100-400 ul blocking solution for 1 hour at room temperature.
- 4. Remove blocking solution and add 100-400 ul diluted primary antibody. Incubate overnight at 4C.
- 5. Remove antibody solution and wash sections in wash buffer three times for 5 minutes each.
- 6. Add 100-400 ul biotinylated diluted secondary antibody. Incubate 30 minutes at room temperature.
- 7. Remove secondary antibody solution and wash sections three times with wash buffer for 5 minutes each.
- 8. Add 100-400 ul Streptavidin-HRP reagent to each section and incubate for 30 minutes at room temperature.
- 9. Wash sections three times in wash buffer for 5 minutes each.
- 10. Add 100-400 ul DAB substrate to each section and monitor staining closely.
- 11. As soon as the sections develop, immerse slides in deionized water.
- 12. Counterstain sections in hematoxylin.
- 13. Wash sections in deionized water two times for 5 minutes each.
- 14. Dehydrate sections.
- 15. Mount coverslips.

## Immunocytochemistry/Immunofluorescence Protocol for Hsp70 Antibody (NBP1-77455)

HSP70/HSPA1A Antibody: https://www.novusbio.com/products/hsp70-hspa1a-antibody\_nbp1-77455 Immunocytochemistry Protocol

Culture cells to appropriate density in 35 mm culture dishes or 6-well plates.

- 1. Remove culture medium and add 10% formalin to the dish. Fix at room temperature for 30 minutes.
- 2. Remove the formalin and add ice cold methanol. Incubate for 5-10 minutes.
- 3. Remove methanol and add washing solution (i.e. PBS). Be sure to not let the specimen dry out. Wash three times for 10 minutes.
- 4. To block nonspecific antibody binding incubate in 10% normal goat serum from 1 hour to overnight at room temperature.
- 5. Add primary antibody at appropriate dilution and incubate at room temperature from 2 hours to overnight at room temperature.
- 6. Remove primary antibody and replace with washing solution. Wash three times for 10 minutes.
- 7. Add secondary antibody at appropriate dilution. Incubate for 1 hour at room temperature.
- 8. Remove antibody and replace with wash solution, then wash for 10 minutes. Add Hoechst 33258 to wash solution at 1:25,0000 and incubate for 10 minutes. Wash a third time for 10 minutes.
- 9. Cells can be viewed directly after washing. The plates can also be stored in PBS containing Azide covered in Parafilm (TM). Cells can also be cover-slipped using Fluoromount, with appropriate sealing.

\*The above information is only intended as a guide. The researcher should determine what protocol best meets their needs. Please follow safe laboratory procedures.





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## **Products Related to NBP1-77455**

HAF008 Goat anti-Rabbit IgG Secondary Antibody [HRP]

NB7160 Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]

NBP2-24891 Rabbit IgG Isotype Control

NBP1-77455AF488 HSP70/HSPA1A Antibody [Alexa Fluor® 488]

#### Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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