

FOXP3

CONTACT INFORMATION:	Monoclonal Antibodies Unit. Centro Nacional de Investigaciones Oncológicas
STATUS:	Validated
TYPE:	mouse anti human
CLONE NAME:	221D
PROTEIN:	Human full length FOXP3
ANTIGEN USED:	GST-FOXP3 recombinant protein
FUSION PARTNER:	myeloma p3-NS1/Ag4-1 (NS1) cells
ISOTYPE:	IgG1
SPECIES REACTIVITY:	human, mouse, cat, dog, horse, rabbit, alpaca and pig.
PREPARATION AND STORAGE:	Aliquot and store at 4C. Do not freeze
APP RECOMMENDED:	IHQ-paraffin, IHQ-frozen, WB, Flow cytometry, IF
APP NO TESTED:	IP

DESCRIPTION

FOXP3 is a forkhead-family transcription factor that negatively regulates T cell function. Mice carrying a loss-of-function mutation in FoxP3 present with fatal autoimmune-like disease caused by hyperresponsive CD4(+) T cells. Mice that overexpress scurfin possess fewer mature T cells with reduced functional capabilities compared with normal littermate control mice. FOXP3 is critical for normal CD4+ T cell function and for the successful coordination of a normal response to immunological challenge in vivo.

PUBLICATION DESCRIBING ANTIBODY CHARACTERIZATION/VALIDATION

Roncador G, Brown PJ, Maestre L, Hue S, Martínez-Torrecuadrada JL, Ling KL, Pratap S, Toms C, Fox BC, Cerundolo V, Powrie F, Banham AH. Analysis of FOXP3 protein expression in human CD4(+)CD25(+) regulatory T cells at the single-cell level. Eur J Immunol 2005 35:1681-1691. <http://www.ncbi.nlm.nih.gov/pubmed/15902688>

Banham AH, Lyne L, Scase TJ, Blacklaws BA. Monoclonal antibodies raised to the human FOXP3 protein can be used effectively for detecting Foxp3(+) T cells in other mammalian species. Vet Immunol Immunopathol. 2009 Feb 15;127(3-4):376-81.

<http://www.sciencedirect.com/science/article/pii/S0165242708007083>

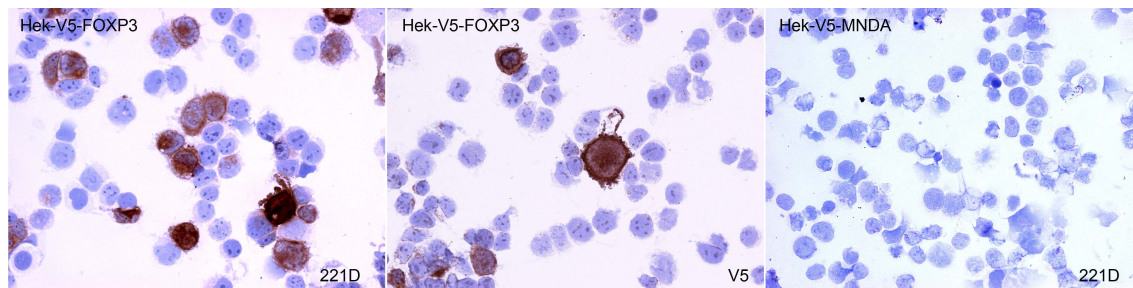
REFERENCES

Roncador G, Brown PJ, Maestre L, Hue S, Martínez-Torrecuadrada JL, Ling KL, Pratap S, Toms C, Fox BC, Cerundolo V, Powrie F, Banham AH. Analysis of FOXP3 protein expression in human CD4(+)CD25(+) regulatory T cells at the single-cell level. Eur J Immunol 2005 35:1681-1691.

Banham AH, Lyne L, Scase TJ, Blacklaws BA. Monoclonal antibodies raised to the human FOXP3 protein can be used effectively for detecting Foxp3(+) T cells in other mammalian species. Vet Immunol Immunopathol. 2009 Feb 15;127(3-4):376-81.

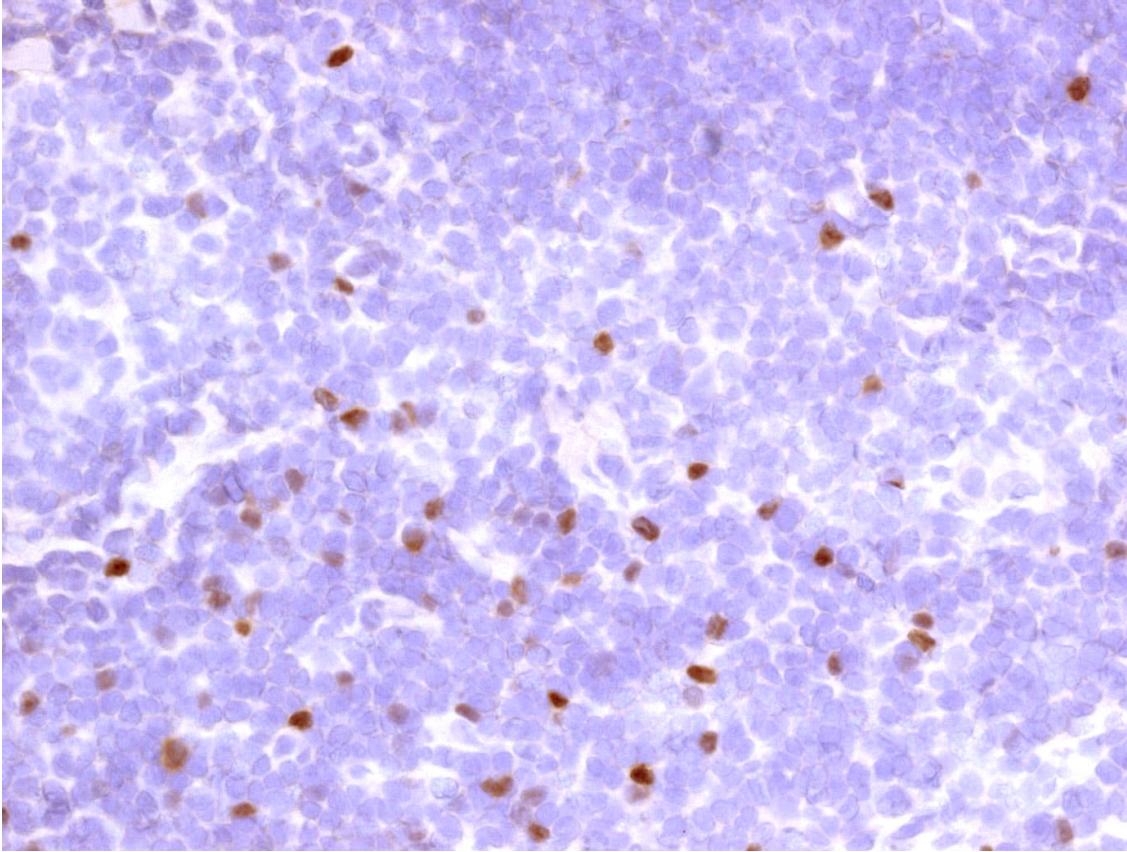
APPLICATIONS

IHC Techniques	Clone	Dilution	Antibody concentration	Antigen retrieval method	Visualization kit	Positive control	Negative control	Protein localization	Positivity in other species
Frozen tissue and cytopins									
Recommended	221D	1:2	supernatant			Tonsil		nuclear	mouse
Paraffin tissue									
Recommended	221D	1:20	supernatant	20 min ER2 (Tris-EDTA)	Novolink kit	Tonsil		nuclear	mouse
Immunofluorescence									

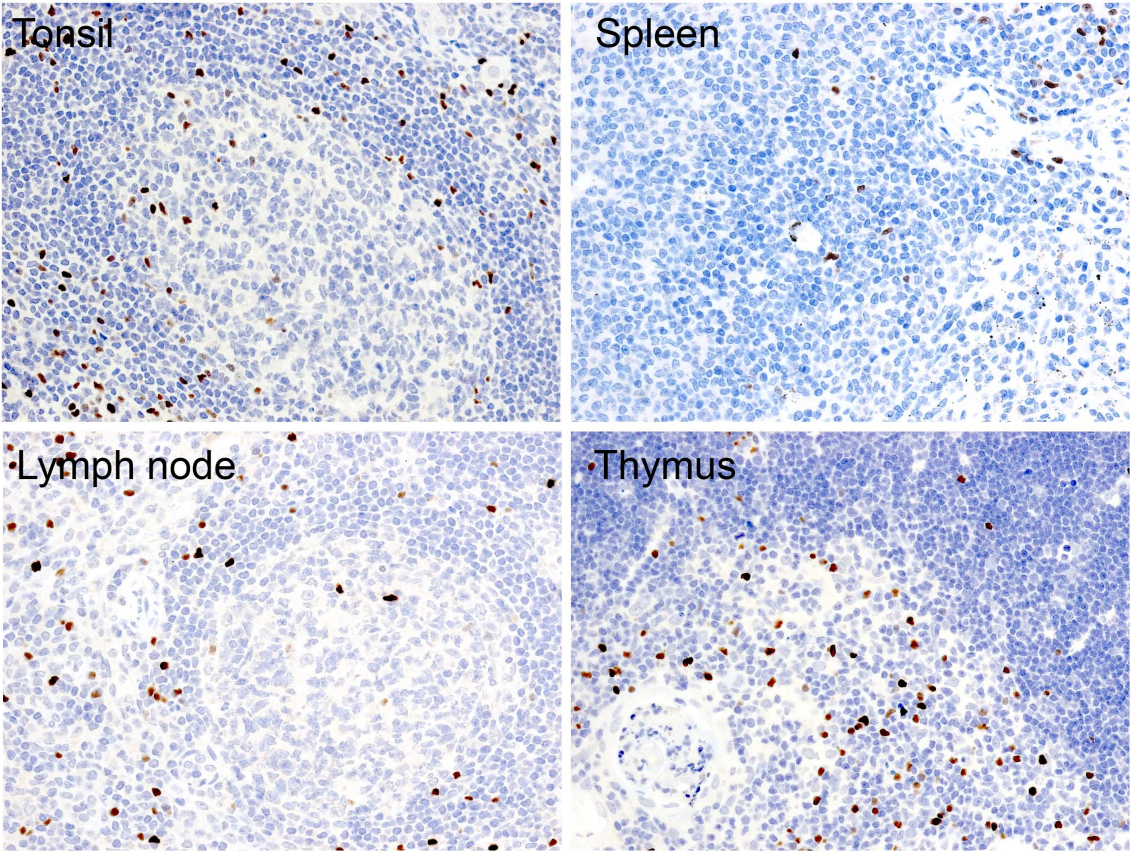


Anti-FOXP3 antibody in transfected cells

Hek-V5-MNDA was used as negative control.

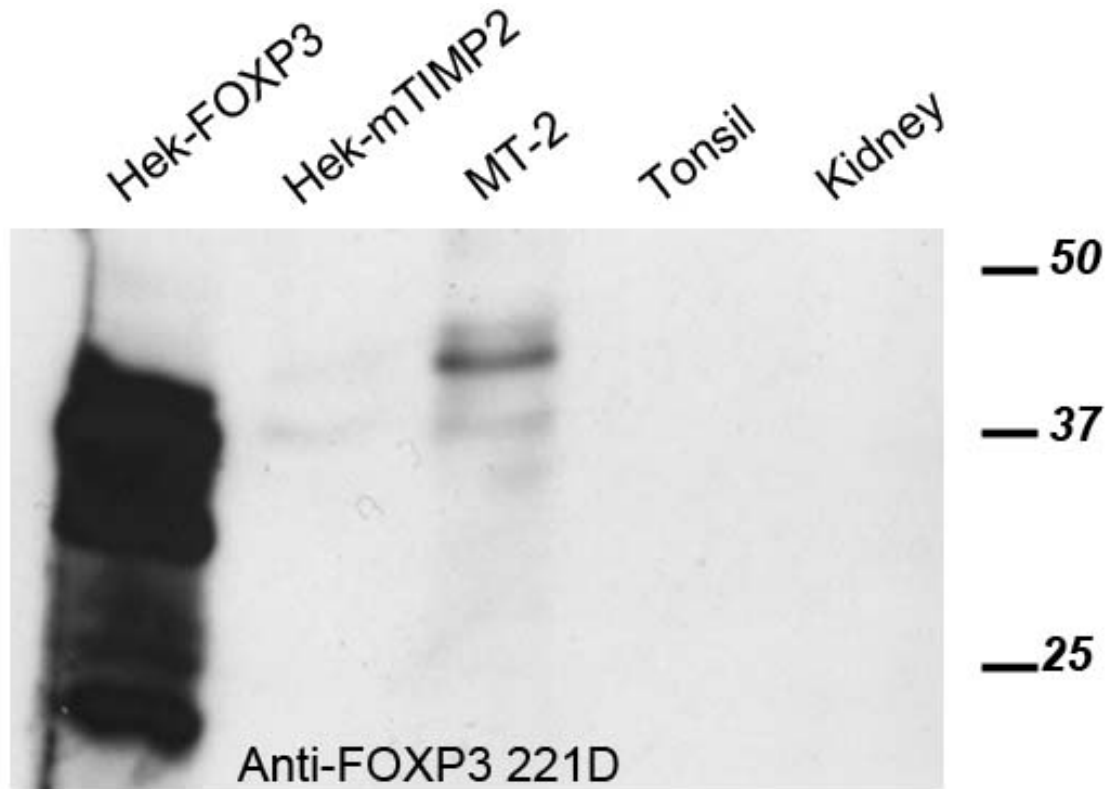


FOXP3 (221D) expression on human frozen tonsil.



Anti-FOXP3 antibody on human paraffin sections.

WB Techniques	Clone	Dilution	Antibody concentration	Positive control	Negative control	Expected MW	Observed Mw	Positivity in other species
Western Blotting								
Recommended	221D	supernatant	neat	MT2 cell line	Kidney	47kDa	47kDa	
Immunoprecipitation								



Western Blotting characterization of 221D monoclonal antibody

Western Blotting characterisation of 221D monoclonal antibody

Antibody 221D recognises HEK-FOXP3 transfected cells and MT-2 cell line (human T cell leukemia virus carrier cell line) by WB.

Lane1 HEK-FOXP3 transfected cells (20ug) (+)

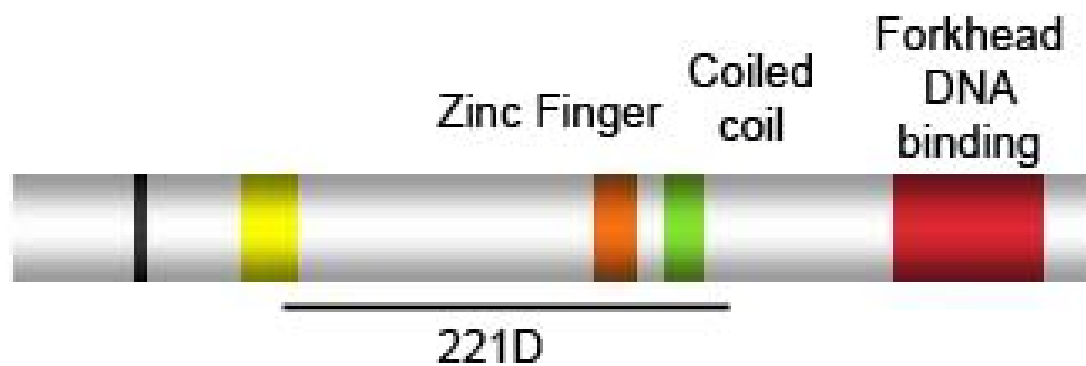
Lane 2 HEK-mTIMP2 transfected cells (20ug) (-)

Lane 3 MT2 cell line (150ug) (+)

Lane 4 Human tonsil (150ug) (-)

Lane 5 Human kidney (150ug) (-)

OTHERS	Title	Description
Recommended	Epitope map of anti-FOXP3 (221D) antibody	

**Epitope map of anti-FOXP3 (221D) antibody**

The figure shows Foxp3 protein functional domains and the location of epitope recognized by 221D antibody (aa105-236). Foxp3 contains a Zn finger/Leucine zipper and a conserved Forkhead Domain.