abcam

ab8069

Anti-Vimentin antibody [V9] - Cytoskeleton Marker

Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069) is a mouse monoclonal antibody detecting Vimentin in **Western Blot, Flow Cytometry (Intra), IHC-P, ICC/IF**. Suitable for **Human, Rat**.

- KO validated for confirmed specificity
- Over 170 publications
- Trusted since 2002

Lab Essentials | KO Validated

Key facts

Isotype	IgG1
Host species	Mouse
Storage buffer	pH: 7.4 Preservative: 0.02% Sodium azide Constituents: PBS, 6.97% L-Arginine
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	V9
Purification technique	Affinity purification
Concentration	1 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	0.5-5 µg/mL
Notes	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Expected

Species	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Horse, Chicken, Cow, Cat, Dog, Pig
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Rat
Dilution info	0.1-1 µg for 10^6 Cells
Notes	ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.

Species	Human
Dilution info	0.1-1 µg for 10^6 Cells
Notes	ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.

Predicted

Species	Horse, Chicken, Cow, Cat, Dog, Pig
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	1 µg/mL
Notes	-

Expected

Species	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Horse, Chicken, Cow, Cat, Dog, Pig
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Rat
Dilution info	0.5-5 µg/mL
Notes	-

Species	Human
Dilution info	0.5-5 µg/mL
Notes	-

Predicted

Species	Horse, Chicken, Cow, Cat, Dog, Pig
Dilution info	-

Notes	-

Target data

See full target information VIM

Function	Vimentins are class-III intermediate filaments found in various non-epithelial cells, especially mesenchymal cells. Vimentin is attached to the nucleus, endoplasmic reticulum, and mitochondria, either laterally or terminally. Plays a role in cell directional movement, orientation, cell sheet organization and Golgi complex polarization at the cell migration front (By similarity). Protects SCRIB from proteasomal degradation and facilitates its localization to intermediate filaments in a cell contact-mediated manner (By similarity). May promote axon outgrowth and motor fiber repair via DSP-mediated recruitment to outgrowth tips (By similarity). Involved with LARP6 in the stabilization of type I collagen mRNAs for CO1A1 and CO1A2.

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

What is this antibody validated in?
Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069) is a mouse monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Immunohistochemistry (IHC-P), Immunocytochemistry/immunofluorescence (ICC/IF) in Human, Rat samples.

What is the molecular weight of Vimentin?
Anti-Vimentin [V9] - Cytoskeleton Marker (ab8069) specifically detects a band for Vimentin (UniProt: P02543) at a molecular weight of 54kDa.

Trusted by the scientific community
Anti-Vimentin [V9] - Cytoskeleton Marker (ab8069) was first used in a scientific publication in 2002 and has been cited over 170 times in peer-reviewed journals.

Reviewed by scientists

Anti-Vimentin [V9] - Cytoskeleton Marker (ab8069) has over 25 independent reviews from customers.

Specificity confirmed
The specificity of Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069) has been confirmed by Immunocytochemistry testing in VIM Knockout HAP1 cells.

Other related products
We have a range of other formats of antibody clone [V9] also available for your convenience: ab8069, Alexa Fluor® 488 - ab195877, Alexa Fluor® 647 - ab195878, HRP - ab196602, Carrier free - ab223871, ab283641, Carrier free - ab302471, ab316575, Carrier free - ab316907

Want a custom formulation?
This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or conjugation for your experiments, please contact orders@abcam.com

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

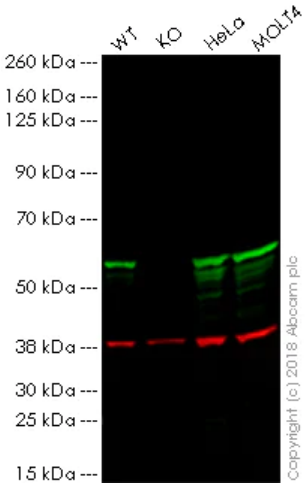
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

14 product images



Western blot - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

Lanes 1 - 4: Merged signal (red and green). Green - ab8069 observed at 57 kDa. Red - loading control, ab181602, observed at 37 kDa.

ab8069 was shown to specifically react with Vimentin in wild-type HAP1 cells as signal was lost in VIM (Vimentin) knockout cells. Wild-type and VIM (Vimentin) knockout samples were subjected to SDS-PAGE. ab8069 and ab181602 (Rabbit anti-GAPDH loading control) were incubated overnight at 4°C at 1 µg/ml and 1/10000 dilution respectively. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed ab216772 and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed ab216777 secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.
All lanes:
Western blot - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069) at 1 µg/mL

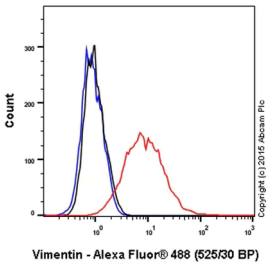
Lane 1:
Wild-type HAP1 whole cell lysate at 20 µg

Lane 2:
VIM (Vimentin) knockout HAP1 whole cell lysate at 20 µg

Lane 3:
HeLa whole cell lysate at 20 µg

Lane 4:
MOLT4 whole cell lysate at 20 µg

Predicted band size: 53 kDa

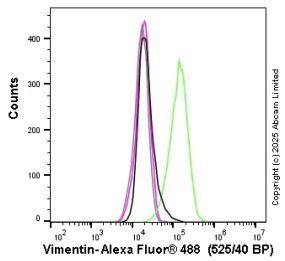


Flow Cytometry (Intracellular) - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

Vimentin Flow Cytometry (Intracellular) staining using mouse Anti-Vimentin antibody

Overlay histogram showing MDA-MB-231 cells stained with ab8069 (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilized with 0.1% PBS-Triton X-100 for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab8069, 1µg/1x10^6 cells) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-mouse IgG (H&L) (ab150113) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (ab91353, 1µg/1x10^6 cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control.
Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.

This antibody gave a positive signal in MDA-MB-231 cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Triton X-100 for 20 min used under the same conditions.



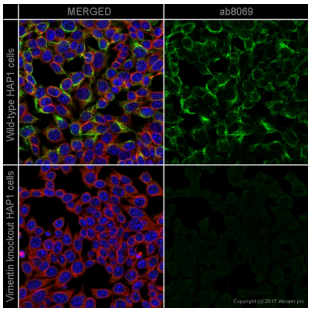
Flow Cytometry (Intracellular) - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

Vimentin Flow Cytometry (Intracellular) staining using mouse Anti-Vimentin antibody

Flow cytometry overlay histogram showing wild-type HAP1 (green line) and VIM knockout HAP1 cells stained with ab8069 (magenta line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS containing 10% normal goat serum to block non-specific protein-protein interaction followed by the antibody (ab8069) ($1x10^6$ in 100 µl at 0.04 µg/ml) for 30 min at 22°C.
The secondary antibody Goat anti-mouse IgG H&L (Alexa Fluor® 488, pre-adsorbed) (ab150117) was used at 1/4000 for 30 min at 22°C.

Isotype control antibody was mouse IgG1κ (ab170190) used at the same concentration and conditions as the primary antibody (wild-type HAP1 - black line, VIM knockout HAP1 - grey line). Unlabelled sample was also used as a control (this line is not shown for the purpose of simplicity).

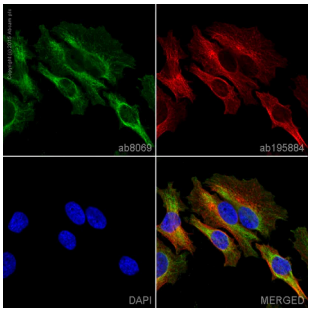
Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.



Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

Vimentin Immunocytochemistry/ Immunofluorescence staining using mouse Anti-Vimentin antibody

ab8069 staining Vimentin (colored green) in wild-type HAP1 cells (top panel) and VIM knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab8069 at 0.5µg/ml and ab202272 (Rabbit monoclonal [EP1332Y] to alpha Tubulin (Alexa Fluor® 594)) at 1/250 dilution overnight at +4°C, followed by a further incubation at room temperature for 1h with ab150117 (Goat secondary antibody to Mouse IgG (Alexa Fluor® 488)) at 2 µg/ml (colored green). Nuclear DNA was labelled in blue with DAPI.
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

Vimentin Immunocytochemistry/ Immunofluorescence staining using mouse Anti-Vimentin antibody

ab8069 staining Vimentin in HeLa cells. The cells were fixed with 4% formaldehyde (10min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab8069 at 1/100 dilution (shown in green) and ab195889, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 594), at 2µg/ml (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

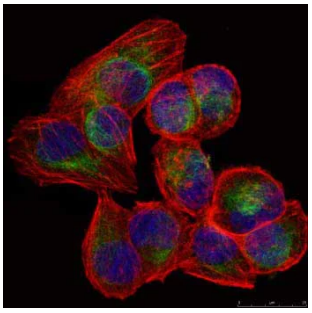
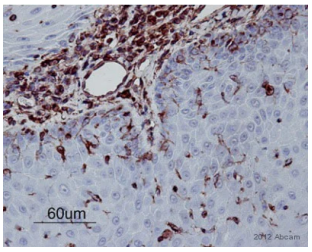


Image from Loessner D et al, Biomaterials. 2010 Nov;31(32):8494-506. Epub 2010 Aug 14. doi:10.1016/j.biomaterials.2010.07.064

Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

ab8069 staining Vimentin in human epithelial ovarian serous adenocarcinoma cell line SKOV-3 by Immunocytochemistry/ Immunofluorescence.

Samples were fixed with 4% PFA in PBS pH 7.4 and then permeabilised using 0.2% saponin for 30 minutes. A blocking step was performed using 1% BSA/PBS for 1 hour. Samples were then incubated with ab8069 at a 1/200 dilution in 1% BSA/PBS for 1 hour. The secondary antibody was a goat anti-mouse Alexa 488 (green) diluted 1/1000, 1% BSA/PBS for 1 hour. Samples were then incubated with phalloidin (red for actin staining) in 1% BSA/PBS for 45 minutes and counterstained with DAPI (blue for nuclei staining) in PBS for 45 minutes.

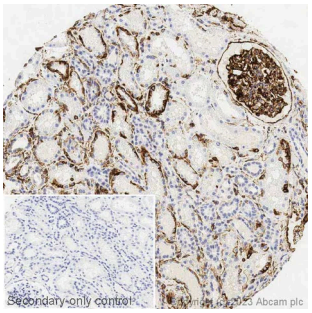


This image is courtesy of an anonymous customer review

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

ab8069 staining Vimentin in Human oral cavity tissue sections by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections).

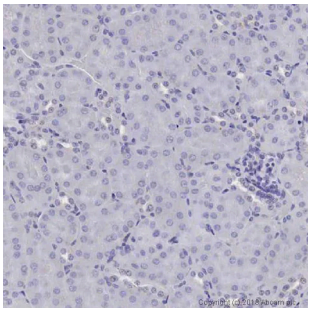
Tissue was fixed with paraformaldehyde and blocked with ab64226 Protein Block for 5 minutes at 25°C; antigen retrieval was by heat mediation in pH 6 buffer . Samples were incubated with primary antibody (1/1000 in 10% NGS) for 16 hours at 4°C. An undiluted biotinylated goat anti-rabbit polyclonal IgG was used as the secondary antibody.



Lab

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

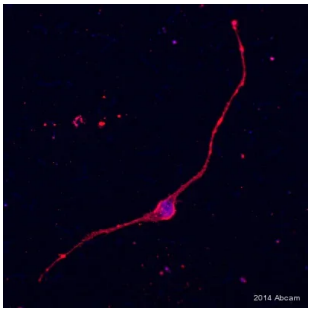
IHC image of Vimentin staining in a section of formalin-fixed paraffin-embedded human normal kidney* performed on a Leica Biosystems BOND® RX instrument. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20mins. The section was then incubated with ab8069, 0.5ug/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. The inset secondary-only control image is taken from an identical assay without primary antibody. For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times. *Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

Vimentin Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-Vimentin antibody

IHC image showing no staining when ab8069 was used on **mouse kidney** formalin fixed paraffin embedded tissue, using MOM detection kit, ab127055. The section was pre-treated using pressure cooker heat mediated antigen retrieval with sodium citrate buffer (pH6) for 30 mins. The section was incubated with ab8069 at 5 µg/ml for 15 mins at room temperature. DAB was used as the chromogen. The section was counterstained with haematoxylin and mounted with DPX.
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

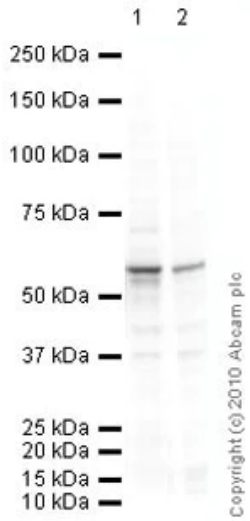


This image is courtesy of an anonymous customer review

Immunocytochemistry/ Immunofluorescence - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

Vimentin Immunocytochemistry/ Immunofluorescence staining using mouse Anti-Vimentin antibody

ab8069 staining Vimentin in rat glial cells by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with formaldehyde and blocked with 1% BSA for 10 minutes at 25°C. Samples were incubated with primary antibody (1/200) for 10 hours at 4°C. An Alexa Fluor® 555-conjugated anti-mouse IgG polyclonal (1/300) was used as the secondary antibody.



Western blot - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

Vimentin Western blot staining using mouse Anti-Vimentin antibody

All lanes:
Western blot - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069) at 1 µg/mL

Lane 1:
HeLa (Human epithelial cell line from cervix adenocarcinoma) Whole Cell Lysate at 10 µg

Lane 2:
MOLT4 (Human lymphoblastic leukemia cell line) Whole Cell Lysate at 10 µg

Secondary

All lanes:
Western blot - Goat Anti-Mouse IgG H&L (HRP) preadsorbed (ab97040) at 1/5000 dilution

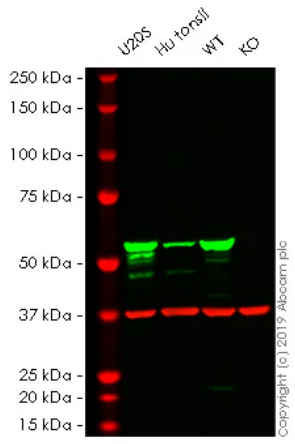
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 53 kDa

Observed band size: 57 kDa

Exposure time: 20min



Western blot - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

Vimentin Western blot staining using mouse Anti-Vimentin antibody

Lanes 1 - 4: Merged signal (red and green). Green - ab8069 observed at 53 kDa. Red - loading control, ab181602 observed at 37 kDa.
ab8069 was shown to react with Vimentin in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab255446 (knockout cell lysate ab263775) was used. Wild-type and Vimentin knockout samples were subjected to SDS-PAGE. ab8069 and Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) were incubated overnight at 4°C at 1 µg/ml and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (ab216772) and Goat Anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (ab216777) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069) at 1 µg/mL

Lane 1:
U-2 OS (Human bone osteosarcoma epithelial cell line) cell lysate at 20 µg

Lane 2:
Human tonsil cell lysate at 20 µg

Lane 3:
Wild-type HeLa cell lysate at 20 µg

Lane 4:
Western blot - Human VIM (Vimentin) knockout HeLa cell lysate (ab263775) at 20 µg

Secondary

All lanes:
Western blot - Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (ab216772) at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 53 kDa

Observed band size: 50 kDa, 53 kDa

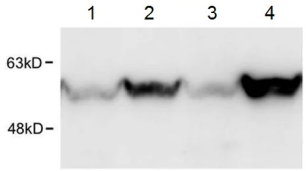


Image from Tange S et al., PLoS One. 2014;9(12):e115684. Fig 9(H).; doi: 10.1371/journal.pone.0115684. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Western blot - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

All lanes:
Western blot - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

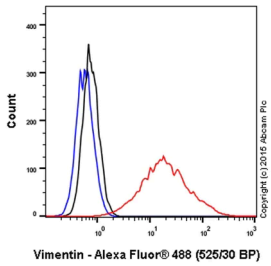
Lane 1:
Whole cell lysate of A549 cells

Lane 2:
Whole cell lysate of A549 cells treated with TGF-beta

Lane 3:
Whole cell lysate of A549 cells overexpressing JARID2

Lane 4:
Whole cell lysate of A549 cells overexpressing JARID2 treated with TGF-beta

Predicted band size: 53 kDa



Flow Cytometry (Intracellular) - Anti-Vimentin antibody [V9] - Cytoskeleton Marker (ab8069)

Vimentin Flow Cytometry (Intracellular) staining using mouse Anti-Vimentin antibody

Overlay histogram showing SV40LT-SMC cells stained with ab8069 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Triton X-100 for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab8069, 0.1µg/1x10^6 cells) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-mouse IgG (H&L) (ab150113) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (ab91353, 0.1µg/1x10^6 cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control.
Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.