

# IFNG [IFNG/3996R]

Concentrated and Prediluted Rabbit Monoclonal Antibody  
901-3266-060223

**BIOCARE**  
M E D I C A L

Available Product Formats				
Format	Catalog Number	Description	Dilution	Diluent
Concentrate	ACI 3266 A, C	0.1, 1.0 mL	1:100	Renoir Red
Predilute	API 3266 AA	6.0 mL	Ready-to-use	N/A

## Intended Use:

For In Vitro Diagnostic Use

IFNG [IFNG/3996R] is a rabbit monoclonal antibody that is intended for laboratory use in the qualitative identification of IFNG protein by immunohistochemistry (IHC) in formalin-fixed paraffin-embedded (FFPE) human tissues. The clinical interpretation of any staining or its absence should be complemented by morphological studies using proper controls and should be evaluated within the context of the patient's clinical history and other diagnostic tests by a qualified pathologist.

## Summary and Explanation:

Interferon-gamma (IFNG) is a cytokine secreted predominantly by activated lymphocytes such as CD4 T helper type 1 (Th1) cells and CD8 cytotoxic T cells, natural killer cells, B cells, and antigen-presenting cells. IFNG expression is induced by mitogens and cytokines (1). The downstream target genes of IFNG signaling pathway regulate several biological functions, including cell cycle, apoptosis, and inflammation (2). In adaptive immunity, IFNG directly regulates the differentiation, activation, and homeostasis of Th1 cells; inhibits Th2 cell development; promotes regulatory T cell development and natural killer cell activity; and induces class I MHC expression (3).

## Principle of Procedure:

Antigen detection in tissues and cells is a multi-step immunohistochemical process. The initial step binds the primary antibody to its specific epitope. After labeling the antigen with a primary antibody, a one-step or two-step detection procedure can be applied. A one-step procedure will feature an enzyme labeled polymer that binds the primary antibody. A two-step procedure will feature a linker antibody added to bind to the primary antibody. An enzyme-labeled polymer is then added to bind the linker antibody. These detections of the bound antibodies are evidenced by a colorimetric reaction.

**Source:** Rabbit monoclonal

**Species Reactivity:** Human, others not tested

**Clone:** IFNG/3996R

**Isotype:** IgG

**Protein Concentration:** Call for lot specific Ig concentration.

**Epitope/Antigen:** IFNG

**Cellular Localization:** Cytoplasmic and cell surface

**Positive Tissue Control:** Appendix, thyroid

**Known Applications:**

Immunohistochemistry (formalin-fixed paraffin-embedded tissues)

**Supplied As:** Buffer with protein carrier and preservative

**Storage and Stability:**

Store at 2°C to 8°C. The product is stable to the expiration date printed on the label, when stored under these conditions. Do not use after expiration date. Diluted reagents should be used promptly; any remaining reagent should be stored at 2°C to 8°C.

## Protocol Recommendations (intelliPATH FLX® and manual use):

**Peroxide Block:** Block for 5 minutes with Peroxidized 1.

**Pretreatment:** Perform heat retrieval using Diva Decloaker. Refer to the Diva Decloaker data sheet for specific instructions.

**Protein Block (Optional):** Incubate for 5-10 minutes at RT with Background Punisher.

**Primary Antibody:** Incubate for 30 minutes at RT.

**Probe:** N/A

**Polymer:** Incubate for 30 minutes at RT with a tertiary polymer.

**Chromogen:** Incubate for 5 minutes at RT with Biocare's DAB – OR – Incubate for 5-7 minutes at RT with Warp Red.

**Counterstain:** Counterstain with hematoxylin. Rinse with deionized water. Apply Tacha's Bluing Solution for 1 minute. Rinse with deionized water.

## Protocol Recommendations (intelliPATH FLX and manual use)

### Cont'd:

### Technical Note:

This antibody, for intelliPATH FLX and manual use, has been standardized with MACH4 detection system. Use TBS for washing steps.

### Performance Characteristics:

Sensitivity, specificity, and cross-reactivity are summarized in Tables 1 and 2, respectively.

### Limitations:

The optimum antibody dilution and protocols for a specific application can vary. These include, but are not limited to fixation, heat-retrieval method, incubation times, tissue section thickness and detection kit used. Due to the superior sensitivity of these unique reagents, the recommended incubation times and titers listed are not applicable to other detection systems, as results may vary. The data sheet recommendations and protocols are based on exclusive use of Biocare products. Ultimately, it is the responsibility of the investigator to determine optimal conditions.

### Quality Control:

Refer to CLSI Quality Standards for Design and Implementation of Immunohistochemistry Assays; Approved Guideline-Second edition (I/LA28-A2) CLSI Wayne, PA USA ([www.clsi.org](http://www.clsi.org)). 2011

### Precautions:

1. This antibody contains less than 0.1% sodium azide. Concentrations less than 0.1% are not reportable hazardous materials according to U.S. 29 CFR 1910.1200, OSHA Hazard communication and EC Directive 91/155/EC. Sodium azide (NaN<sub>3</sub>) used as a preservative is toxic if ingested. Sodium azide may react with lead and copper plumbing to form highly explosive metal azides. Upon disposal, flush with large volumes of water to prevent azide build-up in plumbing. (Center for Disease Control, 1976, National Institute of Occupational Safety and Health, 1976) (4)
2. Specimens, before and after fixation, and all materials exposed to them should be handled as if capable of transmitting infection and disposed of with proper precautions. Never pipette reagents by mouth and avoid contacting the skin and mucous membranes with reagents and specimens. If reagents or specimens come into contact with sensitive areas, wash with copious amounts of water. (5)
3. Microbial contamination of reagents may result in an increase in nonspecific staining.
4. Incubation times or temperatures other than those specified may give erroneous results. The user must validate any such change.
5. Do not use reagent after the expiration date printed on the vial.
6. The SDS is available upon request and is located at <http://biocare.net>.

### Troubleshooting:

Follow the antibody specific protocol recommendations according to data sheet provided. If atypical results occur, contact Biocare's Technical Support at 1-800-542-2002.

### References:

1. Castro F, Cardoso AP, Gonçalves RM, et al. Interferon-gamma at the crossroads of tumor immune surveillance or evasion. *Front Immunol.* 2018; 9:847.
2. Zaidi MR. The interferon-gamma paradox in cancer. *J Interferon Cytokine Res.* 2019;39(1):30-38.
3. Benci JL, Johnson LR, Choa R, et al. Opposing functions of interferon coordinate adaptive and innate immune responses to cancer immune checkpoint blockade. *Cell.* 2019;178(4):933-948.
4. Center for Disease Control Manual. Guide: Safety Management, NO. CDC-22, Atlanta, GA. April 30, 1976 "Decontamination of Laboratory Sink Drains to Remove Azide Salts."



60 Berry Drive  
Pacheco, CA 94553  
USA



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Tel: 800-799-9499 | [www.biocare.net](http://www.biocare.net) | Fax: 925-603-8080



Westervoortsedijk 60  
6827 AT Arnhem  
The Netherlands

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## References Cont'd:

5. Clinical and Laboratory Standards Institute (CLSI). Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline-Fourth Edition CLSI document M29-A4 Wayne, PA 2014.

**Table 1:** Sensitivity and specificity were determined by testing formalin-fixed, paraffin-embedded diseased tissues.

Tissue	Positive Cases	Total Cases
Breast Adenocarcinoma (IDC)	23	24
Colon Adenocarcinoma	36	40
Lung Adenocarcinoma	16	24
Lung Squamous Cell Carcinoma	12	24
Prostate Adenocarcinoma	23	40

**Table 2:** Tissue cross-reactivity was determined by testing formalin-fixed, paraffin-embedded normal tissues.

Tissue	Positive Cases	Total Cases
Cerebrum	0	6
Cerebellum	0	3
Adrenal	1	3
Ovary	0	3
Pancreas	3	3
Lymph Node	0	3
Tracheal	3	3
Pituitary	0	0
Testis	0	3
Thyroid	0	3
Breast	2	3
Spleen	0	3
Tonsil	2	3
Thymus	0	3
Bone Marrow	0	3
Lung	3	3
Heart	0	3
Esophagus	0	3
Stomach	3	3
Small Intestine	3	3
Colon	9	11
Liver	0	3
Salivary Gland	3	3
Kidney	3	3
Prostate	8	10
Uterus	1	3
Cervix	1	3
Skeletal Muscle	0	3
Skin	0	3
Peripheral Nerve	0	3
Meshothelium	0	3
Eye	0	3
Laryngopharynx	2	2