

BIOTEM  
885, rue Alphonse Gourju  
38140 Apprieu  
FRANCE

Tel: +33 (0)4 76 65 10 91

Fax: +33 (0)4 76 67 48 86

[info@biotem.fr](mailto:info@biotem.fr)

[www.biotem-antibody.com](http://www.biotem-antibody.com)

## TECHNICAL DATASHEET

## Monoclonal Anti-Nitrotyrosine

Clone 2E11

Ref # BIO.000.3

## Product description

Description	Monoclonal antibody to nitrotyrosine protein
Host species	Mouse
Tested applications	ELISA, IP, WB
Cross reactivity	Determined by competitive ELISA Nitrotyrosine: 93 % of inhibition Tyrosine: 0 % of inhibition Nitrotryptophan: 1 % of inhibition Aminotyrosine: 0 % of inhibition Phosphotyrosine: 5 % of inhibition Chlorotyrosine: 13 % of inhibition
Specificity	Antibody specific of nitrotyrosine either free or incorporated in proteins such as human serum nitro albumin, nitro haemoglobin and nitro insulin Reactivity against all species
Epitope	Protein-bound nitrotyrosine

## Target exploration

### Overview:

A large number of physiological and pathological events lead to so-called stress conditions to which the organism can adapt within certain limits by regulating the activity of a series of enzymatic cascades. Most stress responses are characterized by an increased generation of "free radicals" which encompass both reactive oxygen (ROS) and nitrogen species (RNS). Most of these species react with macromolecules of the organism, i.e. aminoacids, proteins, lipids and DNA, leading to functional alterations which can either participate to adaptation or lead to cell death. Oxidative stress however, has different functional consequences depending not only upon its intensity and duration, but also upon the nature of the free radicals, ROS or RNS, generated. Interestingly, the nature of these reactive species depends on the ratio between the initially produced ROS, superoxide anion (O<sub>2</sub><sup>-</sup>), and nitric oxide (NO). Indeed, O<sub>2</sub><sup>-</sup> very rapidly reacts with NO and therefore, as long as the NO/ O<sub>2</sub><sup>-</sup> ratio is  $\geq 1$ , O<sub>2</sub><sup>-</sup> will therefore preferentially react with NO rather than with macromolecules, thus generating reactive nitrogen species (RNS): nitrosonium (NO<sup>+</sup>), N<sub>2</sub>O<sub>3</sub> and peroxynitrite (ONOO<sup>-</sup>).

These RNS induce posttranslational modifications: for NO<sup>+</sup> and N<sub>2</sub>O<sub>3</sub>, S-nitrosation (Cys-SNO) and for ONOO<sup>-</sup>, tyrosine nitration (Tyr-NO<sub>2</sub>), methionine sulfoxidation (Met-SOH) and thiol oxidation (RS-S-R). When the O<sub>2</sub><sup>-</sup>/NO ratio becomes  $> 1$ , the O<sub>2</sub><sup>-</sup> and NO<sub>2</sub> ions and thereafter the OH<sup>•</sup> radicals, cause irreversible oxidations and peroxidations of macromolecules which generally lead to cell death(1-4). The monoclonal antibodies have been selected for their high affinity and specificity towards Tyr-NO<sub>2</sub> residues. They do not cross-react with Tyr or Tyr derivatives such as

aminotyrosine, chlorotyrosine or phosphotyrosine , neither with nitroTrp which can also be generated in response to peroxynitrite. They also recognize Tyr-NO<sub>2</sub> residues in various sequences as shown by their ability to recognize nitrated proteins including albumin, ovalbumin, insulin, hemoglobin, KLH and various cytoplasmic and mitochondrial proteins.

## Properties

Format	Liquid – 1mg/ml
Storage instructions	Store at +4°C short term (1-2 weeks). Aliquot and store at -20°C or -80°C. Avoid repeated freeze / thaw cycles
Storage buffer	Phosphate Buffered Saline 10 mM – NaCl 0.15 M – pH 7.4 – Thimerosal 0,01% may be used as preservative
Purity	Purified by Affinity chromatography on Protein A
Clonality	Monoclonal
Isotype	IgG2b
Restrictions	For research use only.

## Applications

### Recommended dilutions

WB	1/10 000
IP	1/1 000
ELISA	1/10 000 to 1/500 000

Optimal dilutions/concentrations should be determined by the end user

## Publications

- García-Heredia JM et al. - Nitration of tyrosine 74 prevents human cytochrome c to play a key role in apoptosis signaling by blocking caspase-9 activation – *Biochimica et Biophysica Acta*, 1797: 981-993 (2010)
- Csibi A et al. - Angiotensin II Inhibits Insulin-Stimulated GLUT4 Translocation and Akt Activation through Tyrosine Nitration-Dependent Mechanisms – *PLoS One*. 2010 Apr 7;5(4):e10070