COSMO BIO CO., LTD.
Inspiration for Life Science

MONOCLONAL ANTIBODY

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Catalog No. AGE-M01

Anti N$^{\varepsilon}$-(carboxymethyl) lysine (CML)

BACKGROUND

N$^{\varepsilon}$-(carboxymethyl) lysine (CML) is reported as a major antigenic AGE structure. Recent studies demonstrate that CML is generated by the oxidative cleavage of Amadori products by hydroxyl radical, peroxynitrite and hypochlorous acid, thus suggesting CML to be an important biological marker of oxidative stress in vivo.

Product type	Primary antibody
Immunogen	CML-HSA
Host Species	Mouse
Fusion Partner	P3U1
Clone Designation	2G11
Isotype	IgG1
Host	Mouse
Source	Ascites
Purification	Protein G
Form	Liquid
Formulation Buffer	PBS containing 0.1% proclin as a preservative
Concentration	0.2 mg / ml
Volume	100 ul
Label	Unlabeled
Specificity	CML
Cross species reactivity	-
Storage	Store below -20℃ (below -70°C for prolonged storage) Aliquot to avoid cycles of freeze/thaw.

Application notes

Recommended dilutions

- **Western blotting:** 1/200 - 1/400
- **Immunofluorescence:** 1/100 - 1/200
- **ELISA:** 1/200 - 1/400

Other applications have not been tested.
Optimal dilutions/concentrations should be determined by the end user.

References

1) Mera K., Nagai M., Brock JW., Fujiwara Y., Imai H., Murata T., Maruyama T., Baynes JW., Otagiri M., Nagai R. Glutaraldehyde is an Effective Cross-linker for Production of Antibodies Against Advanced Glycation End Products. J. Immunol. Methods 334 (1-2), 82-90 (2008) **PMID:** 18353354

ANTIBODY CHARACTERIZATION

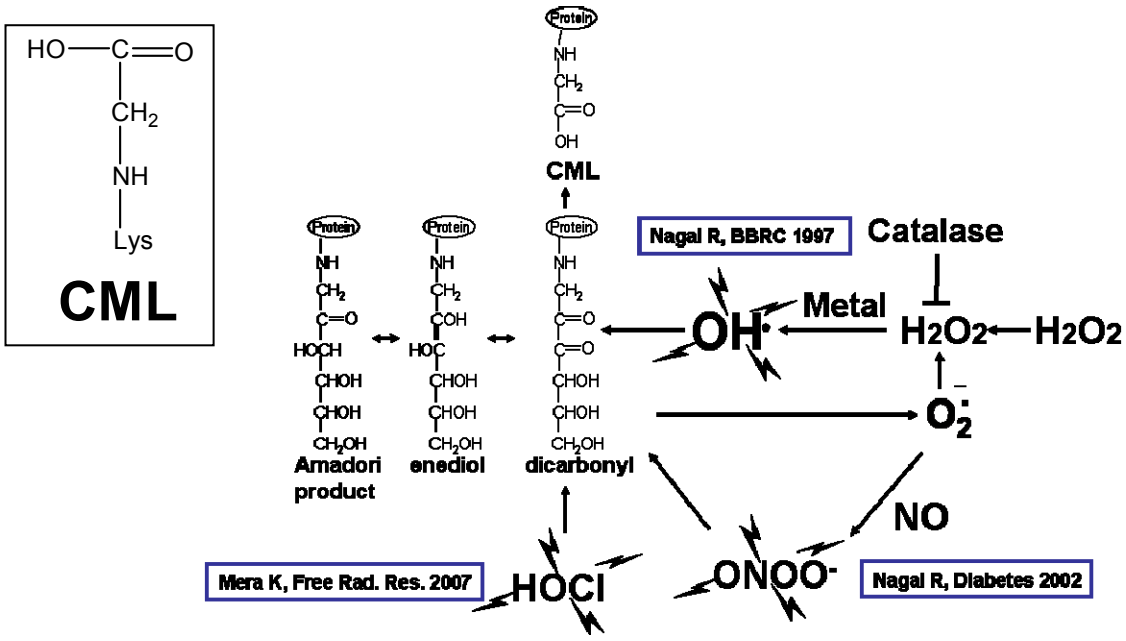

Fig.1 CML production pathway

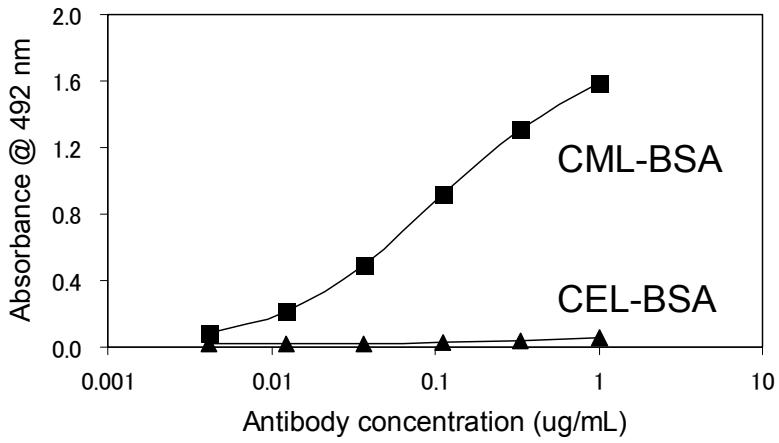

Fig.2 Immunoreactivity of the CML(2G11) monoclonal antibody to CML-BSA and CEL-BSA

ELISA protocol

Coating
1) Distribute 100 ul / well of the sample solution (1 ug/mL in PBS) to 96 well microtiter plates (Thermo, MaxiSorp).
2) Incubate the plates 2h at RT or overnight at 4 degrees.
3) Discard the supernatant of sample solution.
4) Wash the plates three times with washing.buf.(PBS/0.05%Tween 20)

Blocking
1) Distribute 200 ul / well of 0.5% gelatin-PBS to 96 well microtiter plates
2) Incubate the plates 1h at RT.
3) Discard the the supernatant of 0.5% gelatin-PBS
4) Wash the plates three times with washing.buf.(PBS/0.05%Tween 20)

Primary antibody
1) Distribute 100 ul / well of Primary antibodies diluted with washing buf. as suggested in the APPLICATIONS to each well.
2) Incubate the plates 1h at RT.
3) Discard the supernatant of Primary antibody solution.
4) Wash the plates three times with washing.buf.(PBS/0.05%Tween 20)

Secondary antibody
1) Distribute 100 ul / well of secondary antibodies (HRP-anti mouse IgG) diluted with washing buf. as suggested in the APPLICATIONS to each well.
2) Incubate the plates 1h at RT.
3) Discard the supernatant of secondary antibody.
4) Wash the plates three times with washing.buf.(PBS/0.05%Tween 20)

OPD color reaction
1) Reaction for 2-10 minutes at RT.
2) Distribute 100 uL / well of 2M H_2SO_4 to each well and stop enzyme reaction.
3) After gentle mixing, determine the absorbance at 492 nm of each well by a spectrophotometer.

RELATED PRODUCTS:

Product Name	Quantity	Maker	Cat#
Anti N^{ε}-(carboxymethyl) lysine [CML] (2G11) Monoclonal Antibody	100 ul	CAC	AGE-M01
Anti N^{ε}-(carboxyethyl) lysine [CEL] (CEL-SP) Monoclonal Antibody	100 ul	CAC	AGE-M02
Anti GA-pyridine (2A2) Monoclonal Antibody	100 ul	CAC	AGE-M03
Anti N^{ω}-(carboxymethyl) arginine [CMA] (3F5) Monoclonal Antibody	100 ul	CAC	AGE-M04
CML-BSA/Nε-(carboxymethyl) lysine-BSA	200 ul	CSR	AGE-GP01
CEL-BSA/Nε-(carboxyethyl) lysine-BSA	200 ul	CSR	AGE-GP02
GA-BSA/Glycolaldehyde-BSA	200 ul	CSR	AGE-GP03
Ribose-gelatin	500 ul	CSR	AGE-GP04
Mild-AGE-BSA	200 ul	CSR	AGE-GP05

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