



MONOCLONAL ANTIBODY

For research use only. Not for clinical diagnosis

Catalog No. AGE-M01

Anti N^ε-(carboxymethyl) lysine (CML)

BACKGROUND

N^ε-(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 and 2% sorbitol as a preservative |
| Concentration | 0.2 mg / ml |
| Volume | 100 ul |
| Label | Unlabeled |
| Specificity | CML |
| Cross species reactivity | - |
| Storage | Store below -20°C (below -70°C for prolonged storage) Aliquot to avoid cycles of freeze/thaw. |

- Application notes**
- **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

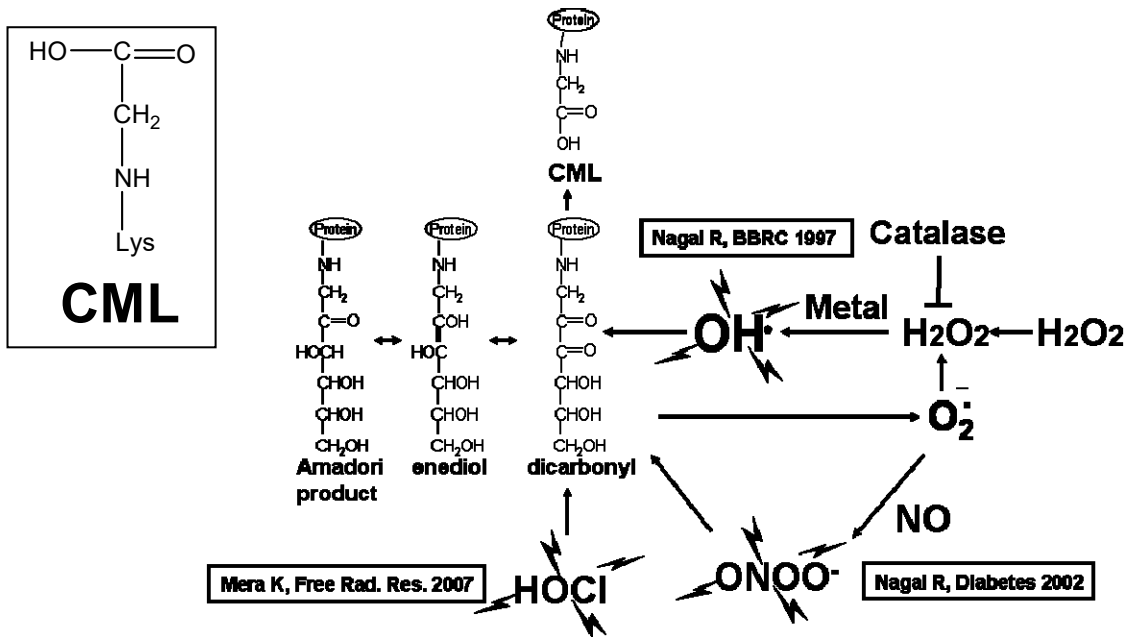


Fig.1 CML production pathway

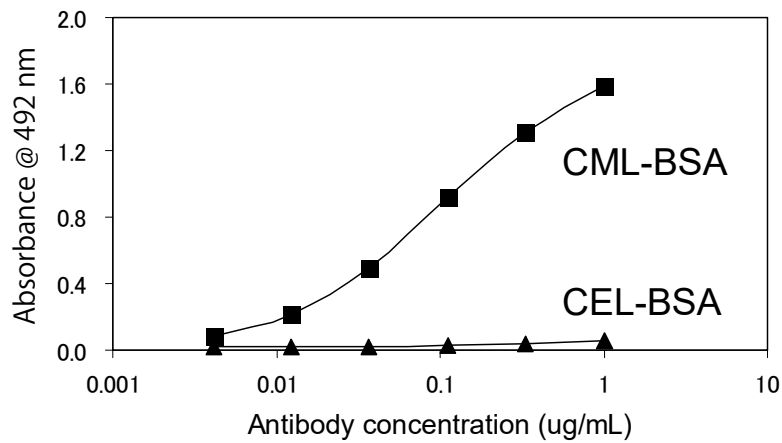


Fig.2 Immunoreactivity of the CML(2G11) monoclonal antibody to CML-BSA and CEL-BSA

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 H₂SO₄ 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/N ^ε -(carboxymethyl) lysine-BSA | 200 ul | CSR | AGE-GP01 |
| CEL-BSA/N ^ε -(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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