**Mild-AGE-BSA**

| **Product Description** | Fatty acid-free bovine serum albumin (BSA) (0.05 g/ml) was incubated with 50 mM of glucose in a 0.05 M sodium phosphate buffer (pH 7.4) at 37°C for 24 weeks, followed by dialysis against PBS. The CML content (0.4 mol CML/mol BSA) was determined by amino acid analysis. This method is prepared mildly-modified-AGE-BSA (mild-AGE-BSA). The CML content for the mild-AGE-BSA is 24.4 mmol CML/mol Lys. However, the CML contents for the diabetic (DM)- and non-diabetic human lens samples were about 17.4 mmol/mol Lys and about 8.6 mmol/mol Lys, respectively, thus indicating that the CML content of mild-AGE-BSA was similar to physiological samples. |
| **Volume** | 200 ul |
| **Concentration** | 1 mg/ml |
| **Storage** | Store below -20°C (below -70°C for prolonged storage). Aliquot to avoid cycles of freeze/thaw. |

**References**


**Characterization**

![Comparison of CML content in the samples by HPLC](https://www.cosmobio.co.jp)

www.cosmobio.co.jp
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 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:

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<th>Product Name</th>
<th>Quantity</th>
<th>Maker</th>
<th>Cat#</th>
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<td>AGE-M01</td>
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