**HT 8-oxo-dG ELISA Kit**

**Product #: 4370-096-K**

**Technical Specifications**

A competitive immunoassay for the detection and quantitation of 8-oxo-dG in urine and serum samples.

**Description**

The Trevigen 8-oxo-dG ELISA (enzyme-linked immunosorbent assay) is a fast and sensitive competitive immunoassay for the detection and quantitation of 8-hydroxy-2’-deoxyguanosine (8-oxo-dG). 8-oxo-dG has become a frequently used biomarker of oxidative DNA damage and oxidative stress. Measurement of urinary 8-oxo-dG may be useful as an indicator of oxidative damage.

**Specificity**

Recognizes 8-hydroxy-2’-deoxyguanosine; also detects 8-hydroxyguanosine (product of oxidative RNA damage) and 8-hydroxyguanine (product of oxidative DNA damage by hydroxyl radical).

**Sample Types**

Urine, Serum

**Scientific Background**

Intracellular and extracellular free radical species can be potentially damaging to the living cell. Intracellular free radical species are produced as a result of normal metabolism and extracellular forms are produced as a result of ultraviolet radiation or ionizing radiation. Reactive oxygen species (ROS) are of particular interest in the research of oxidative damage and disease. The various ROS include the highly reactive hydroxyl radical (OH·), superoxide radical (O2·−), hypochlorite ion (OCl·) and non-radical hydrogen peroxide (H2O2). DNA, lipids, and proteins are cellular targets for oxidative damage by ROS and the order of preference for modification depends on location of ROS production, availability of metal ions, and the relative ability for the target to be oxidized. Cells have acquired a number of defense mechanisms to cope up with oxidative damage by ROS and other free radicals. The simplest defense mechanisms involve Vitamin C and E intercepting free radicals, becoming radicals themselves and protecting cellular biomolecules from damage. Complex defense mechanisms involve enzymes like superoxide dismutase, catalase and glutathione peroxidase that have evolved to reduce ROS levels. Low background levels of damage occur even in normal cells because ROS have a tendency to escape. Indeed, when defense mechanisms cannot prevent the accumulation of ROS, then there is an increase in cellular damage. Modified lipids and proteins are removed via normal lipid- and protein-turnover mechanisms. However, modified DNA cannot be replaced and has to be repaired. Numerous DNA repair mechanisms have evolved in the cell and have become the focus of research in many disease states. Removal of DNA damage and restoration of the continuity of the DNA duplex response, activation of the DNA damage checkpoint, which stops the cell cycle and prevents transmission of damaged chromosomes, changes in the transcriptional response of the cell and apoptosis are some of the important DNA damage response reactions. 8-hydroxy-2’-deoxyguanosine (8-oxo-dG) is the most commonly studied and detected by-product of DNA damage that is excerted in the urine upon DNA repair. Urinary 8-oxo-dG and its analogs 8-oxo-G and 8-hydroxyguanine are linked to many degenerative diseases. The association of ROS and the use of 8-oxo-dG as a biomarker of oxidative stress have been investigated in many diseases, which include bladder and prostate cancer, cystic fibrosis, atopic dermatitis and rheumatoid arthritis. Oxidative stress is associated with the pathogenesis of Parkinson’s disease, Alzheimer’s disease and Huntington’s diseases and elevated levels of DNA damage have been measured in a wide range of neurological conditions (1-9).

**Assay Procedure Summary**

1. Add 8-oxo-dG Standards and samples in duplicate to the pre-coated ready-to-use 8-oxo-dG Immunoassay Plate.
2. Add diluted Anti-8-oxo-dG; incubate then wash.
3. Add diluted Anti-Mouse IgG:HRP Conjugate; incubate then wash.
4. Develop with TMB Substrate.
5. Add Stop Solution 2.
6. Measure absorbance at 450nm.
7. Plot 8-oxo-dG standard curve and determine sample concentrations using the Calculations Worksheet provided on the website.

**Typical Standard Curve**

![Typical Standard Curve Graph]

**Background References**

Please refer to the instruction manual for a listing of the background references.

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**HT 8-oxo-dG ELISA Kit**

**Product #: 4370-096-K**

**Number of Samples:** 1 standard curve, 39 samples in duplicate or 2 standard curves and 30 samples in duplicate

**Sensitivity:** 1.9 ng/mL

**Range:** 1.875–60 ng/mL

**Sample Volume:** 50 µL/well

**Incubation Time:** <3 hours

**Wavelength Setting:** 450nm

**Materials Included:**

- 8-oxo-dG Immunoassay Plate
- 8-oxo-dG Standard
- Sample Diluent
- 20X Wash Buffer
- Anti-8-oxo-dG
- Antibody Diluent
- Anti-Mouse IgG:HRP Conjugate
- HRP Conjugate Diluent
- TMB Substrate
- Stop Solution 2
- Instruction Manual

**Storage of Kit:** All reagents are stable as supplied at 4°C, except the **8-oxo-dG Standard**, which should be stored at -20°C. For optimum storage, the 8-oxo-dG Standard should be aliquoted into smaller portions and stored at -20°C. Avoid repeated freeze thaw cycles.

**Note:** The components in each kit lot # have been quality assured and warranted in this specific combination only; do not mix them with components from other kit lot #'s.
Section 1—Product Identification and Use

Product Name: 8-oxo-dG ELISA Kit
Product#: 4370-096-K

This product is sold only for research use by qualified laboratory personnel, and is not to be used as a drug, medical device, food additive, cosmetic, nor household chemical. It is not to be used in diagnostic, therapeutic, consumer, agricultural, nor pesticidal applications.

Manufacturer’s Name: Trevigen, Inc.
Street Address: 8405 Helgerman Court
City, Prov. Postal Code: Gaithersburg, MD 20877
USA Fax: (301) 560-4973
EMERGENCY PHONE: (301) 216-2800 Toll Free in North America

Section 2—Hazardous Ingredients

Kit components may contain the following components:

<table>
<thead>
<tr>
<th>CAS Registry Numbers</th>
<th>Chemical Name</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>26172-55-4</td>
<td>5-chloro-2-methyl-isothiazolin-3-one</td>
<td>&lt;&lt;1.15</td>
</tr>
<tr>
<td>2682-20-4</td>
<td>2-methyl-4isothiazolin-3-one</td>
<td>&lt;&lt;0.35</td>
</tr>
<tr>
<td>10031-43-4</td>
<td>Copper (II) nitrate</td>
<td>&lt;&lt;0.35</td>
</tr>
<tr>
<td>7786-30-3</td>
<td>Magnesium Chloride</td>
<td>&lt;&lt;1.0</td>
</tr>
<tr>
<td>1.077-60-3</td>
<td>Magnesium Nitrate</td>
<td>&lt;&lt;1.7</td>
</tr>
<tr>
<td>14933-09-6</td>
<td>n-Tetradecyl-N,N-dimethyl-1-propanesulfonate</td>
<td>0.125</td>
</tr>
<tr>
<td>N/A</td>
<td>Methylisothiazolone</td>
<td>&lt;&lt;0.02</td>
</tr>
<tr>
<td>N/A</td>
<td>Bromonitrodioxide</td>
<td>&lt;&lt;0.02</td>
</tr>
<tr>
<td>54827-17-7</td>
<td>3,3’,5,5’-tetramethylbenzidine (TMB)</td>
<td>N/A</td>
</tr>
<tr>
<td>7647-01-0</td>
<td>Hydrogen chloride</td>
<td>&lt;8.5</td>
</tr>
</tbody>
</table>

Section 3—Physical Data

The physical properties of the kit components have not been investigated thoroughly.

TMB: Clear liquid
Stop Solution: Clear, colorless to slight yellow, liquid

Section 4—Fire and Explosion Hazard

Components of the kit are non-flammable. Fire and explosion data have not been investigated.

Hazardous Combustion Products:
Kit components may emit toxic vapors
TMB: carbon monoxide, carbon dioxide, nitrogen oxide fumes
Stop Solution: Carbon dioxide, carbon monoxide, hydrogen gas fumes

Section 5—Reactivity Data

Kit components are stable. Avoid contact with reducing agents, amines, strong acids, strong bases, and strong oxidizing agents.

TMB: Stable. Protect from direct UV light, avoid elevated temperatures, protect from moisture. Incompatible with strong oxidizing agents, metals.
Stop Solution: Stable. Avoid elevated temperatures. Incompatible with oxidizing agents, reducing agents, and bases.
MATERIAL SAFETY DATA SHEET

Section 6—Toxicological Properties
The toxicological properties of the kit components have not been investigated. Kit components may burn eyes severely, be harmful if absorbed through skin, burn skin severely, irritate respiratory tract, be harmful if inhaled, be harmful if swallowed. May cause an allergic reaction in some individuals.

TMB: Irritant to eyes and skin. May cause irritation of the digestive tract and respiratory tract.
Stop Solution: Corrosive to skin and eyes. Inhalation of vapors may cause irritation of nasal and respiratory tract. Ingestion causes gastrointestinal tract discomfort, nausea and vomiting. IARC: Group 3 Carcinogen

Section 7—Spill and Disposal
Wear chemical safety glasses or goggles and compatible chemical-resistant gloves. Avoid inhalation, contact with eyes, skin or clothing.

*****MULTIPLE COMPONENT SPILL OR LEAK PROCEDURES*****

- Wear protective equipment (respirator, chemical safety goggles, rubber boots, and heavy rubber gloves).
- Ventilate area.
- Absorb on sand or vermiculite and place in closed containers for disposal.
- Dispose or mix the material with a combustible solvent and burn in a chemical incinerator equipped with an afterburner and scrubber.
- Containers of this material may be hazardous when emptied. Emptyed containers retain product residues: handle as if they were full. Observe all federal, state and local environmental regulations.

Section 8—First Aid Measures

- If any of the kit components are ingested, immediately rinse mouth with water and seek medical attention.
- In case of skin contact, flush with copious amounts of water for at least 15 minutes. Remove contaminated clothing and shoes. If a rash or other irritation develops, seek medical attention.
- If inhaled, remove to fresh air. If breathing becomes difficult, seek medical attention.
- If in case of eye contact, flush with copious amounts of water for at least 15 minutes while separating the eyelids with fingers. Seek medical attention.

Section 9—Preparation

Prepared by: Trevigen, Inc.  Phone#: (301) 261-2800  Created: 08/10/05  Modified: 07/12/06
Toll free in North America: 1-800-TREVIGEN

The above information is believed to be correct but does not purport to be all inclusive and shall be used only as a guide. Trevigen shall not be held liable for any damage resulting from handling or from contact with the above product. See the Technical Specification, Packing Slip, Invoice, and Product Catalogue for additional terms and conditions of sale.