

Quality control of New 8-OHdG Check ELISA

(code: KOG-200S/E)

1) Comparison of standard curves of different production lot.

| Lot # | 8-OHdG standards (absorbance at 450nm) | | | | | |
|----------|--|---------|---------|----------|----------|-----------|
| | 0.5 ng/mL | 2 ng/mL | 8 ng/mL | 20 ng/mL | 80 ng/mL | 200 ng/mL |
| Lot. 032 | 1.621 | 1.483 | 1.099 | 0.678 | 0.263 | 0.138 |
| Lot. 033 | 1.855 | 1.684 | 1.272 | 0.874 | 0.363 | 0.184 |
| Lot. 034 | 1.782 | 1.656 | 1.314 | 0.894 | 0.385 | 0.229 |

2) Preparation of QC samples.

Prepare three normal human urine samples. Add 8-OHdG standard to prepare QC sample with high concentration of 8-OHdG. Please repeat freeze-thaw cycle at least three times and remove insoluble materials by centrifugation. Store the aliquots below -80 degree C. Before use, QC samples should be thawed out by incubation at 37 degree C for 1 hour or 4 degree C for over night.

3) Result of QC samples.

| Lot # | QC samples (ng/mL) | | | | | |
|----------|--------------------|------|-----|------|------|------|
| | S-1 | S-2 | S-3 | U-1 | U-2 | U-3 |
| Lot. 032 | 107.3 | 28.5 | 9.6 | 74.8 | 28.3 | 15.9 |
| Lot. 033 | 121.1 | 29.9 | 9.5 | 90.5 | 29.4 | 14.0 |
| Lot. 034 | 124.2 | 30.6 | 9.5 | 88.7 | 29.2 | 14.5 |

4) Intra-assay variation of purified 8-OHdG (N=6).

| | New 8-OHdG Check, lot.034 (absorbance at 450nm) | | | | | |
|------|---|---------|---------|----------|----------|-----------|
| | 0.5 ng/mL | 2 ng/mL | 8 ng/mL | 20 ng/mL | 80 ng/mL | 200 ng/mL |
| 1 | 1.818 | 1.658 | 1.318 | 0.892 | 0.374 | 0.241 |
| 2 | 1.762 | 1.636 | 1.342 | 0.917 | 0.387 | 0.241 |
| 3 | 1.784 | 1.684 | 1.315 | 0.890 | 0.391 | 0.225 |
| 4 | 1.778 | 1.669 | 1.311 | 0.905 | 0.390 | 0.224 |
| 5 | 1.777 | 1.630 | 1.291 | 0.884 | 0.395 | 0.232 |
| 6 | 1.772 | 1.658 | 1.304 | 0.874 | 0.371 | 0.210 |
| Mean | 1.782 | 1.656 | 1.314 | 0.894 | 0.385 | 0.229 |
| S.D. | 0.019 | 0.020 | 0.017 | 0.015 | 0.010 | 0.012 |
| C.V. | 1.1% | 1.2% | 1.3% | 1.7% | 2.6% | 5.2% |

5) Intra-assay variation of QC samples (N=6).

| | New 8-OHdG Check, lot.034 (ng/mL) | | | | | |
|------|-----------------------------------|------|------|------|------|------|
| | S-1 | S-2 | S-3 | U-1 | U-2 | U-3 |
| 1 | 119.9 | 30.3 | 9.4 | 83.4 | 27.7 | 13.6 |
| 2 | 125.0 | 30.5 | 9.6 | 88.3 | 28.9 | 15.1 |
| 3 | 112.4 | 29.7 | 9.1 | 83.1 | 28.3 | 14.3 |
| 4 | 129.7 | 30.5 | 9.4 | 89.8 | 29.4 | 14.6 |
| 5 | 129.0 | 30.9 | 9.6 | 92.4 | 31.4 | 14.3 |
| 6 | 129.0 | 31.4 | 9.9 | 95.3 | 29.4 | 15.2 |
| Mean | 124.2 | 30.6 | 9.5 | 88.7 | 29.2 | 14.5 |
| S.D. | 6.88 | 0.55 | 0.28 | 4.84 | 1.25 | 0.61 |
| C.V. | 5.5% | 1.8% | 2.9% | 5.5% | 4.3% | 4.2% |

6) Inter-assay variation of QC samples (N=6).

| | QC samples (ng/mL) | | | | | |
|---------|--------------------|------|------|------|------|------|
| | S-1 | S-2 | S-3 | U-1 | U-2 | U-3 |
| Lot.029 | 117.9 | 30.9 | 10.2 | 82.5 | 28.3 | 15.4 |
| Lot.030 | 124.9 | 32.2 | 8.6 | 93.8 | 28.4 | 15.9 |
| Lot.031 | 117.5 | 30.0 | 10.2 | 88.3 | 27.2 | 13.9 |
| Lot.032 | 107.3 | 28.5 | 9.6 | 74.8 | 28.3 | 15.9 |
| Lot.033 | 121.1 | 29.9 | 9.5 | 90.5 | 29.4 | 14.0 |
| Lot.034 | 124.2 | 30.6 | 9.5 | 88.7 | 29.2 | 14.5 |
| Mean | 118.8 | 30.4 | 9.6 | 86.4 | 28.5 | 14.9 |
| S.D. | 6.43 | 1.23 | 0.59 | 6.79 | 0.78 | 0.92 |
| C.V. | 5.4% | 4.0% | 6.1% | 7.9% | 2.8% | 6.1% |

Samples were assayed in N=6 for each days.