

DRIED URINE TEST SPECIFICATIONS

6-Sulfatoxymelatonin

Clinical Information

6-Sulfatoxymelatonin (MT6s) is the sulfated conjugate of the primary metabolite of melatonin. It collects in the urine, and is a reliable indicator of total melatonin production during the time since the last urine void. The MT6s level in a 1st morning urine sample therefore represents overnight melatonin production.

Melatonin is a hormone produced by the pineal gland during the dark phase of the light/dark cycle. It regulates the sleep/wake cycle and the biological clock. It has free radical scavenging, neuroprotective, and antioxidant properties and plays a role in the immune system to protect against the growth of abnormal tissues such as breast and prostate cancers. Adequate nocturnal melatonin production counterbalanced by suppression of production by light exposure during the day is vital to optimal health. Shift workers are particularly susceptible to suppression of melatonin production due to light exposure during the night, potentially leading to increased risks for cancer and other diseases.

Urinary MT6s levels are corrected using urinary creatinine to allow for variations in hydration status. Reference ranges were established at 4 diurnal time points: 1st morning void, 2nd morning void (2 h after the first), evening (just before dinner), and bedtime. At these time points, MT6s ranges in women are: 18.0-40.9, 7.3-31.9, 0.7-2.2, and 1.7-11.1 $\mu\text{g/g}$ creatinine, respectively; and in men are: 10.1-26.0, 6.0-17.0, 0.5-3.6, and 1.3-8.4 $\mu\text{g/g}$ creatinine, respectively.

References:

Reiter RJ. Melatonin: clinical relevance. *Best Pract Res Clin Endocrinol Metab.* 2003;17(2): 273-85.
Reiter RJ, Tan DX, Gitto E, et al. Pharmacological utility of melatonin in reducing oxidative cellular and molecular damage. *Pol J Pharmacol.* 2004;56(2):159-70.
Vela-Bueno A, Olavarrieta-Bernardino S, Fernández-Mendoza J, Aguirre-Berrocal A. Melatonin, Sleep, and Sleep Disorders. *Sleep Medicine Clinics* 2007;2(2): 303-12.
Mediavilla MD, Sanchez-Barcelo EJ, Tan DX, et al. Basic mechanisms involved in the anti-cancer effects of melatonin. *Curr Med Chem.* 2010;17(36):4462-81.

Assay Method: LC-MS/MS

Intra-assay Precision

Intra-assay precision was determined by choosing 3 dried urine samples spanning the reference range for MT6s, and analyzing them 8 times within the same run. Results are shown below:

Mean MT6s Concentration (ng/mL)	Standard Deviation	Coefficient of Variation (C.V. %)
8.4	0.60	7.2
28.0	1.79	6.4
151.9	7.90	5.2

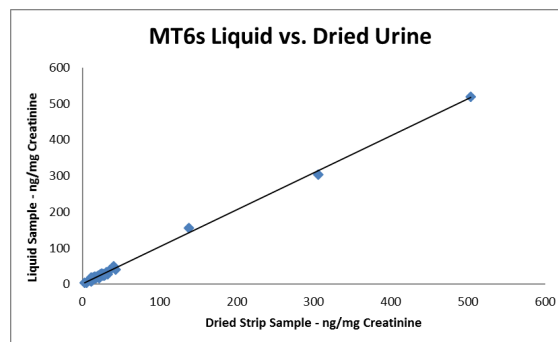
Inter-assay Precision

Inter-assay precision was determined by analyzing a group of method controls in 9 different runs. Results are shown below:

Mean MT6s Concentration (ng/mL)	Standard Deviation	Coefficient of Variation (C.V. %)
9.9	1.06	10.7
18.1	1.56	8.6
111.6	8.04	7.2

Accuracy

To test the accuracy of the dried urine assay for MT6s, dried urine samples collected at the same time as corresponding liquid urine samples were analyzed. Resulting correlation data are shown below ($R^2 = 1.0$):



Linearity

The MT6s assay gives excellent linearity over the reportable range of 0.5-1588 ng/mL.

Analyte Stability

The dried urine MT6s samples are stable for 30 days at room temperature.

Specimen Collection

Kits for dried urine collection contain four filter paper collection strips, easy-to-follow instructions, and a mailer to return the sample for analysis.