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St. Paul's Hospital and Mount St. Joseph Hospital

Laboratory Medicine Technical Bulletin

Testosterone and Bioavailable Testosterone by Liquid Chromatography and Tandem Mass Spectrometry

In late February of 2014, Testosterone analyses at St. Paul's Hospital will transition from immunoassay to liquid chromatography and tandem mass spectrometry (LC-MS/MS) on the ABSCIEX API5000 and 5500 QTRAP Systems. The new assay will be capable of accurate reporting as low as 0.05 nmol/L. This will allow low-level testosterone measurement in: adult males whose testosterone has been medically suppressed for the treatment of prostate cancer, women and children. With the improved analytical performance, new and more comprehensive reference intervals are necessary. The new reference intervals shown in Table 1 have been adapted from Kushnir et al (1) and validated internally.

Males			Females	
Age (d,m,y)	Range (nmol/L)		Age (d,m,y)	Range (nmol/L)
0 d – 30 d	2.6 – 13.89		0 – 30 d	0.69 – 2.22
1 m – <6 m	0.49 – 12.59		1 m – <6 m	< 0.69
6 m – <2 y	<1.28		6 m – <2 y	< 0.31
2 y – <4 y	<0.52		2 y – <4 y	< 0.69
4 y – <6 y	<0.66		4 y – <6 y	< 1.04
6 y – <8 y	<0.45		6 y – <8 y	< 0.24
8 y – <10 y	0.07 – 0.28		8 y – <10 y	0.03 – 0.38
10 y – <12 y	0.07 – 5.72		10 y – <12 y	0.10 – 1.11
12 y – <14 y	0.10 – 21.46		12 y – <14 y	0.21 – 1.73
14 y – <16 y	1.07 – 25.42		14 y – <16 y	0.21 – 1.8
16 – <18 y	5.48 – 28.60		16 – <18 y	0.31 – 2.01
18 – <40 y	8.14 – 38.28		18 – <40 y	0.31 – 1.91
40 – <60 y	6.30 – 35.98		40 – <60 y	0.31 – 1.91
≥60 y	6.30 – 28.10		≥60 y	0.17 – 1.11

Table 1: Reference intervals for total testosterone, adapted from (1)

The change in testosterone will also affect the the bioavailable testosterone (BioT) assay which will use the LC-MS/MS total testosterone and immunoassay-based sex hormone binding globulin (SHBG) concentrations in a calculation for BioT (2). The new reference intervals for BioT are provided in Table 2.

Males		Females	
Age (y)	Range (nmol/L)	Age (y)	Range (nmol/L)
19-<30	3.39-16.89	19-<50y	0.04-0.38
30-<40	3.14-14.84	≥50y	0.03-0.32
40-<50	2.87-12.86		
50-<60	2.67-11.1		
60-<70	2.52-9.56		
70-<80	2.39-8.14		
80-<90	2.33-6.94		
≥90	>2.33		

Table 2: New reference intervals for BioT.

Please note that the specimen type has changed. Red top serum is **mandatory** because gel containing tubes have a well-known interference with testosterone analysis by LC-MS/MS (3). Labs referring to SPH for testosterone and/or bioavailable testosterone should migrate to Red Top Serum immediately. It is not necessary to send a separate gold-top serum for SHBG analysis. A single red-top tube is sufficient for testosterone, SHBG and BioT reporting.

Specimen Type:	**Red Top Serum Only** **No Gel-Containing Tubes**
Minimum Sample Volume:	**250 µL for Testosterone** **500 µL for BioT**
Testosterone Reportable Range :	**0.05 – 45.00 nmol/L**

If you have any questions, please do not hesitate to contact me.

Daniel T. Holmes, MD FRCPC, Medical Biochemist, St. Paul's Hospital.
Clinical Associate Professor of Pathology and Laboratory Medicine, University of British Columbia
tel: 604 806 8919
email: dtholmes@providencehealth.bc.ca

References:

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2. Vermeulen, Alex, Lieve Verdonck, and Jean M. Kaufman. "A critical evaluation of simple methods for the estimation of free testosterone in serum." J Clin Endocrinol Metab. 1999 Oct;84(10):3666-72.
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