

**FORENSIC TOXICOLOGY LABORATORY  
OFFICE OF CHIEF MEDICAL EXAMINER  
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**LCMS TUNING  
(AUTOTUNE/CHECKTUNE)**

**PRINCIPLE**

Liquid chromatography/mass spectrometry (LCMS) quadrupole parameters are adjusted to achieve maximum sensitivity, acceptable resolution and accurate mass assignment during the tuning process. As per manufacturer, the LCMS quadrupole is very stable once the autotune has been performed and therefore does not require frequent autotunes. Instead, a checktune is used to determine whether the quadrupole is correctly tuned. A checktune performs a single profile scan of the tune masses and compares the peak widths and mass axes with target values. Autotune can be performed in negative and/or positive ionization. Checktune can be performed in either positive or negative ionization mode.

LCMS single quadrupole mass spectrometers utilize an electrospray calibrant solution called "ESI tuning mix" to tune the instrument, insuring that instrument is operational. The tuning mix is comprised of several known compounds, as listed in the Certificate of Analysis, which is received with each lot of calibrant. For tuning in positive polarity, the autotune and checktune programs use five ions: m/z 118.09, 622.03, 922.01, 1521.97, and 2121.93. In addition, the autotune program uses the abundance of the naturally-occurring C<sup>13</sup> isotope at m/z 119.09, 623.00, 923.00, 1523.00, 2122.90. For tuning in negative polarity, the autotune and checktune programs use five ions: m/z 112.99, 602.00, 1034.00, 1633.90, 2233.90. In addition, the autotune program uses the abundance of the naturally-occurring C<sup>13</sup> isotope at m/z 113.97, 602.90, 1035.00, 1634.90, 2234.90.

A checktune is run each day an analysis is performed. An autotune is run as needed.

**AGILENT G1946 (LCMS1) AND G6130 (LCMS2)**

1. Select View, #6 MSD Tune, from LCMS online session of ChemStation/Method & Run Control.
2. Select Atunes.tun file from the dialog box. Verify polarity. Select OK.
3. For daily tuning purposes: Select Checktune from Tune option on toolbar.
  - a. If Results Summary generates passing results for Mass Axis and Peak Width (allowable difference  $\leq 0.13$ amu and  $<0.1$ amu respectively) for all 5 masses, save tune file as prompted.

- b. If Results Summary generates any failures, exit Checktune by selecting No to peakwidth and mass axis calibration.
  - i. Under Instrument option on toolbar, select Flush Calibrant.
  - ii. Verify correct calibrant. Flush for 2 minutes.
  - iii. Rerun Checktune. If acceptable Checktune is obtained, the instrument is ready for analysis. If Checktune still fails, run Autotune.
4. For Autotune: Select Autotune from Tune option on toolbar.
  - a. A successfully completed autotune is acceptable. Save autotune file as prompted. Run Checktune. Save as prompted.
  - b. If autotune does not complete successfully, consult with a supervisor.

#### **ADDITIONAL CHECKTUNE CONSIDERATIONS**

1. Signal intensity for the masses is evaluated in addition to the Mass Axis and Peak Width criteria in the Checktune. If signal intensity diminishes over a period of several checktunes, an autotune may need to be performed.

#### **REFERENCE**

Agilent 6100 Series Quadrupole LCMS Systems manuals

Agilent 1100 Series LCMS Systems. Installation Guide.

Agilent 1100 Series LCMS Systems. Users Guide.