**Axima Resonance MALDI Calibration Procedure**

1. Apply matrix and calibration sample to target plate, dry, and insert plate

 0.4uL DHB (40mg/mL) + 0.4uL peptide stds mixture

2. Set up data acquisition parameters. Typical conditions are:

 Exp. Tech. tab:

 Tuning Mode: Positive

 Mode: Operate

Mass Range: 100 – 4000

 Max Laser Rep Rate: 5

 Firing tab:

 Auto Quality: unchecked

Power: 65

 Profiles: 500

 Shots: Off

 MSn Off

 Mass Range: 850 +

 Storage tab:

 Store profile: Never (You will store manually)

 Processing / Peak Processing: Confirm parameters set as shown on desktop file

 Use these same Peak Processing parameters for unknown samples.

3. Click Fire button and collect data

 Manually move laser position with arrows to maximize mV in upper window

 Typical data collection for 100 – 200 Profiles

4. Click Abort and Save data file to project folder for today

5. Open Processing / Calibration window

6. If the correct Calibrant References are listed, click Calibrate

 Click Calibrate button a second time if necessary to assign a time to

 all references. If all are still not detected, change current calibration

 file, and/or manually enter calibrant references.

 Parameters: Monoisotopic, check Correct box, Tolerance 400 mDa

 Adjust Calibrant list as necessary for experiment.

 Recalibrate for negative mode; Can use same peptide mixture

7. Open Display / Calibration window to confirm quality of calibration curve:

 +/- 10 mDa or less is acceptable for PMF identifications

8. In Calibration window, enter new Calibration file name and click Save.

 Confirm that this new Cal file appears in upper Spectrum / Profile

 Window.