

**Gas Chromatography-Mass Spectroscopy conditions to run 6-Chloronicitinoic Acid and 6-Hydroxynicitinoic Acid and sample preparation [1]:**

**Materials Needed:**

GC vials  
GC vial micro-inserts  
GC vial caps  
Nitrogen Gas  
Pyridine  
BSTFA + TMSC 99:1 (Derivatizing Agent)  
Vortex  
GC/MS  
Pipet

**GC/MS Conditions:**

<b>GC Temperature Program</b>			
	°C/min	Temperature (°C)	Minutes
Initial	-	80	2.00
Ramp 1	5.0	100	0.00
Ramp 2	15.00	300	10.00

<b>Oven Configuration:</b>	
Max Temperature (°C)	320
Equilibration time (min)	0.50

<b>Temperatures MSD:</b>	
Set Point Quad:	150
Set Point Source:	230

<b>Inlet Temperature:</b>	
Initial Front (°C)	280
Initial Back (°C)	

<b>Auxiliary Channel #2</b>	
Temperature (°C)	290
Type	MSD

Solvent Delay (min)	4
Injection volume (µL)	1

Protocol:

- 1) Solvent that the sample is resuspended in is evaporated with nitrogen gas.
- 2) The residue is resuspended in 100  $\mu\text{L}$  of Pyridine and transferred to GC vials with micro-inserts.
- 3) 100  $\mu\text{L}$  of the derivatizing agent is added and vortexed for approximately 10 seconds or until the mixture was homogenous.
- 4) After one hour, samples are run on the GC/MS using the above conditions.

References:

[1] Use of High-Performance Liquid Chromatography–UV and Gas Chromatography– Mass Spectrometry for Determination of the Imidacloprid Content of Honeybees, Pollen, Paper Filters, Grass, and Flowers- S-Rossi.