MS analysis was conducted on a single-quadrupole mass spectrometer (ACQUITY QDa; Waters, Milford, MA) equipped with an electrospray interface and coupled via a TLC-MS Interface 2 (oval elution head) to a Dionex HPLC pump (Sunnyvale, CA). Waters Empower software was used for data acquisition. The chromatographic bands were eluted from the plate into the mass spectrometer using methanol with 0.1% ammonium hydroxide at a flow rate of 0.5 mL/min. Electrospray ionization MS spectra were acquired in negative ion mode. The capillary voltage was adjusted to 0.8 kV and the desolvation temperature adjusted to 600°C. Cone voltage was set to 30 V. Masses were scanned between m/z 50 and 900 at a sampling rate of 10 points/s.

The MS spectra of the zones corresponding to avicularin in the sample and standard were compared and the following Precursor ion/Ion fragment were found:

|  |  |  |  |
| --- | --- | --- | --- |
| Standard  | *R*F | Precursor Ion (ESI -) | Fragment (ESI -) |
| Avicularin | 0.48 | 434\* | 303 (301)\* |

\*information regarding the precursor ion and fragments of avicularin were obtained from the following articles:

1. Quantitative and qualitative investigations of pharmacopoeial plant material polygoni avicularis herba by UHPLC-CAD and UHPLC-ESI-MS methods. Phytochemical analysis, vol 26, issue 5, p374-382.

2. Identification of phenolic compounds in artichoke waste by high-performance liquid chromatography–tandem mass spectrometry. [Journal of Chromatography A](http://www.sciencedirect.com.sire.ub.edu/science/journal/00219673). [Volume 1008, Issue 1](http://www.sciencedirect.com.sire.ub.edu/science/journal/00219673/1008/1), 1 August 2003, Pages 57–72

3. Qualitative analysis of phenolic compounds in apple pomace using liquid chromatography coupled to mass spectrometry in tandem mode. Rapid Communications in Mass Spectrometry. Vol 18, issue 5, 2004, pages 553-563

4. Characterization and quantification of flavonoid glycosides in the Prunus genus by UPLC-DAD-QTOF/MS. [Saudi Journal of Biological Sciences](http://www.sciencedirect.com/science/journal/1319562X). 2016



