

Mass Spectrometry

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Rapid Screening of Adulteration in Pomegranate Juice with Grape Juice Using DSA/TOF with No Sample Preparation

Introduction

Pomegranate juice is in high demand due to its well documented health benefits. Studies based on a significant body of scientific research conducted on authentic pomegranate juice have established that pomegranate juice has superior antioxidant activity compared to other popular fruit juices such as grape, orange and apple^{1,2}. These superior health benefits have led to a meteoric rise in the popularity of pomegranate juice over the last 10 years. This has resulted in pomegranate juice having a significantly higher cost than other juices, especially in comparison to apple, orange and grape juice. This high cost has provided an economic incentive for the adulteration of pomegranate juice³. There are a number of other reasons for adulteration including: customers demanding their suppliers reduce costs; there comes a point when a supplier may adulterate the product to lower the cost and maintain a workable margin. Another reason for pomegranate juice adulteration is that it is in high demand and short supply, leading to incentives to extend limited supplies by the addition of other cheaper fruit juices.

One of the common adulterants of pomegranate juice is grape juice. Grape juice is added as a sweetener and coloring agent substitute for natural pomegranate color. Organic acid analysis plays a fundamental role in testing authenticity of pomegranate juice. One of the organic acids, tartaric acid, is present in large amounts in grape juice (1 g/L) but it is absent in pomegranate juice^{4,5}. Therefore, the presence of tartaric acid in pomegranate juice can be used as an indicator of grape juice adulteration. Analytical methods used routinely for organic acid analysis in fruit juices are based on liquid chromatography (reverse phase or ion exchange) coupled to UV detection⁶ or mass spectrometry^{7,8}. These measurement techniques are either expensive or time consuming, or both and require extensive method development and sample

preparation. In this work, we demonstrated that the AxION® Direct Sample Analysis™ (DSA™) system integrated with the AxION 2 Time-of-Flight (TOF) mass spectrometer can be used for rapid screening of adulteration of pomegranate juice with grape juice with no sample preparation.

Experimental

Pomegranate and grape juices were purchased from a local supermarket. Both juices were mixed in different proportions to simulate the adulteration of pomegranate juice with grape juice at different levels of 1, 2, 5, 10 and 20 percent. All fruit juices and their mixtures were measured with an AxION 2 DSA/TOF system with no sample preparation. 10 µl of each sample was pipetted directly onto the stainless mesh of the AxION DSA system and the position of this was optimized, prior to ionization and analysis. The DSA/TOF experimental parameters were as follows: corona current of 5 µA, heater temperature of 300 °C, auxiliary gas (N₂) pressure of 80 psi, drying gas (N₂) flow of 3 l/min and drying gas (N₂) temperature of 25 °C. The AxION 2 TOF MS was run in negative ionization mode with flight voltage of 8000 V. The capillary exit voltage was set to -80 V for the analysis. Mass spectra were acquired in a range of m/z 50-700 at an acquisition rate of 5 spectra/s. All samples were analyzed within 15 sec. To obtain higher mass accuracy, the AxION 2 TOF instrument was calibrated before each analysis by infusing a calibrant solution into a DSA source at 10 µl/min. The data obtained with DSA/TOF was further analyzed with AxION Solo™ software.

Results

All fruit juice samples of pomegranate and grape were directly analyzed by DSA/TOF, with no sample preparation. Figure 1 and Figure 2 show the mass spectra for pomegranate and grape juice in negative ion mode using DSA/TOF, respectively. The mass spectra shows that the organic acids, citric and malic acid, are present in both juices, whereas, tartaric acid, is only present in grape juice. The data shows that tartaric acid can be used as a marker compound to determine the adulteration of pomegranate juice with grape juice using DSA/TOF. This is supported further by data in Figure. 3, which shows the presence of tartaric acid in a pomegranate fruit juice sample adulterated with 1% grape juice. Figure 4 shows that the response for tartaric acid increase with the increase in adulteration of pomegranate juice with grape juice from 1 to 20%. This confirmed that the adulteration of pomegranate juice with grape juice can be detected by the presence of tartaric acid. All mass measurements showed good mass accuracy with an error of less than 5 ppm.

DSA/TOF data obtained with authentic pomegranate and adulterated pomegranate samples with grape juice was centroided and further analyzed with AxION Solo™ software to demonstrate that data can be analyzed and visualized quickly. For this, adulterated pomegranate samples were

spotted on mesh spots number 1,6,9 and authentic pomegranate samples were spotted on rest of mesh spots. AxION Solo software provides fast visualization to determine if your target compound is present or not in sample analyzed. Therefore, the data was analyzed using tartaric acid as a target compound. Figure 5 shows analysis of data with AxION Solo software in plate format and provides a visual image that tartaric acid is present in adulterated pomegranate juice samples spotted on mesh spots 1,6,9 and it was absent in authentic pomegranate juice samples spotted on other mesh spots.

Conclusion

This study shows the first work for rapid screening of the adulteration of pomegranate juice with grape juice using DSA/TOF. The data showed that the presence of tartaric acid in pomegranate juice can be used to detect its adulteration

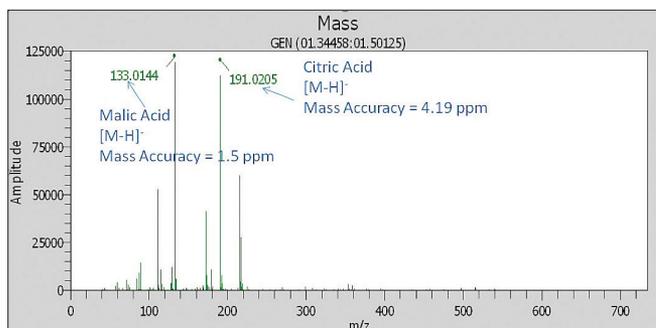


Figure 1. Mass spectra of pomegranate juice in negative ion mode using AxION DSA/TOF.

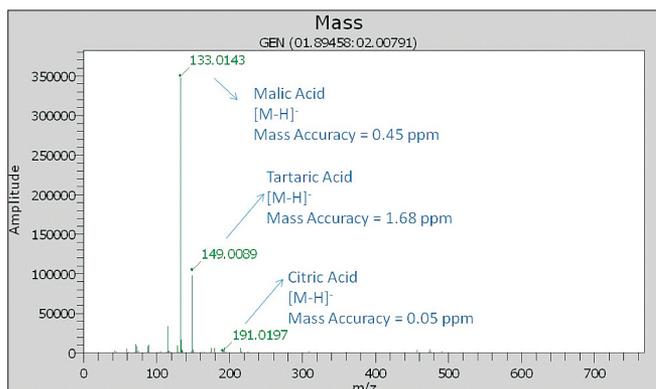


Figure 2. Mass spectra of grape juice in negative ion mode using AxION DSA/TOF.

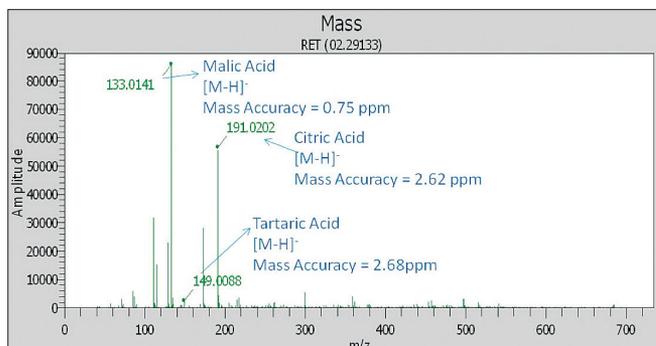


Figure 3. Mass Spectra of pomegranate juice adulterated with 1% grape juice in negative ion mode using AxION DSA/TOF.

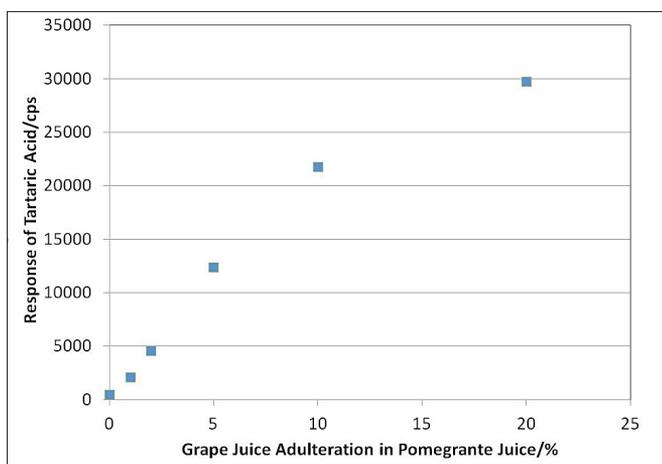


Figure 4. Tartaric acid response with increase in grape juice adulteration in pomegranate juice using AxION DSA/TOF.

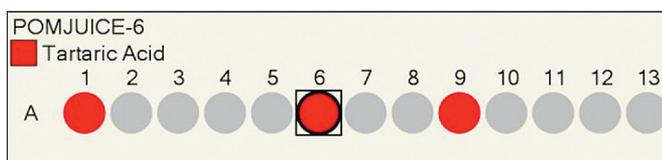


Figure 5. Analysis of authentic pomegranate juice and adulterated pomegranate juice samples with grape juice using AxION Solo software in plate format.

with grape juice. The mass accuracy of all measurements was less than 5 ppm with external calibration. All samples were screened, with no sample preparation, in 15 sec. per sample. We demonstrated that data obtained with DSA/TOF to detect pomegranate juice adulteration with grape juice can be analyzed and visualized quickly with AxION Solo™ software. In comparison to other established techniques such as LC/MS, DSA/TOF will improve laboratory productivity and decrease costs and analysis time.

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