2.6. *Chemometrics*

Chemometrics of hierarchical cluster analysis (HCA) and principal component analysis (PCA) was applied to the FTIR and Raman data of *R. damascena* essential oils and commercial samples using the OPUS software version 7.2 (Bruker, Germany). Dissimilarities and similarities were clearly observed by HCA dendrogram. The HCA dendrogram was built using second derivative spectra (9 smoothing points), Euclidian distance and Ward's algorithm. Three dimensional PCA plots were obtained for displaying the scattering pattern of authentic and commercial samples. Second derivative spectra were used through the factorization algorithm for the PCA analyses. The spectral range of 1800-500 cm^{-1} and 2000-200 cm^{-1} was used in the FTIR and Raman analyses, respectively.

SIMCA 15 (Umetrics, Umea, Sweden), as a supervised chemometrics technique, was used to perform HCA and PCA analyses of GC-MS data, which were obtained from *R. damascena* essential oil samples (n=12) and commercial (n=20) samples.

3. Results

3.1. *Analysis of FTIR spectra of* R. damascena *essential oil*

The typical FTIR spectrum of *R. damascena* essential oil and overlaid FTIR spectra of *R. damascene* essential oil samples and commercial samples are presented in **Figure 1(A)** and **Figure 1(B)**, respectively. The FTIR spectrum of *R. damascene* essential oil had significant vibrational bands at 3345, 2960, 2922, 2853, 1668, 1515, 1451, 1377, 1260, 1235, 1053, 1004 and 829 cm^{-1}. The spectral band at 3345 cm^{-1} could be assigned to the stretching vibrations of the OH functional group of alcohols (Sandasi et, 2011; Tankeu, 2014). The band with a peak point at 2960 cm^{-1} may be attributed to the C=C-C ring vibrations of volatile compounds (Tankeu et al., 2014). Two bands at 2922 and 2853 cm^{-1} were assigned to the methylene C-H asymmetric and symmetric stretching vibrations, respectively (Berechet et al., 2015).

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Deleted: The band with a peak point at 1666 cm^{-1} corresponded to the C=C stretching vibrations (Yang et., Irudayaraj, & Paradkar, 2005). The peak around 1515 cm^{-1} originated from the aromatic ring C=C skeleton vibrations of aromatic substances (Li et al., 2013). The strong bands at 1451 and 1377 cm^{-1} were assigned to the C-OH bending vibrations and C-H asymmetric+ symmetric bending vibrations, respectively (Berechet et al., 2015; Li et al., 2013); Berechet et al., 2015). Bands at 1260 and 1235 cm^{-1} resulted from the C-C-O stretching vibrations and C-O stretching vibrations of phenolics (Bardakçı & Seçilmiş, 2006). The significant bands at 1053 and 1004 cm^{-1} could be attributed to the hydroxyl group vibrations and methylene vibrations, respectively (Berechet et al., 2015; Sandasi et al., 2011). The band at 829 cm^{-1} could be assigned to the C-H stretching vibrations (Tavares & Noreña, 2020). Additionally, the overlapped spectra of all samples are presented in **Figure 1(B).** As can been seen, significant spectral diversities were observed between samples.¶
3.2. Determination of the authenticity of R. damascena essential oil based on FTIR data¶
R. damascena essential oils and commercial samples were classified based on their FTIR spectra by using HCA and PCA. HCA was used to observe the relationship between authentic samples and commercial samples. What is striking about the HCA is it presents a dendrogram, in which all samples clustered related to their similarities and differences. In other words, HCA groups the elements into clusters in terms of their nearness in the multidimensional space (Li et al., 2012). In the present study, HCA was implemented in the spectral range of 1800-500 cm^{-1} by using 2nd derivative FTIR spectra through Ward's algorithm. The superiority of Ward's algorithm is that it provides well-separated clusters by minimizing the variance between groups (Taylan et al., Cebi, Tahsin Yilmaz, Sagdic, & Bakhsh, 2020). Calculations were performed by using the Euclidian distance and dendrograms were built by Ward's algorithm (OPUS Version 7.2, Bruker, Germany). **Figure 2(A)** presents the HCA dendrogram, revealing the similarity and disparity patterns of all samples with a high heterogeneity value of 10000. All samples were clustered as two main groups, authentic rose essential oil samples were clustered on the left arm (numbered as 1) of the dendrogram, and fraudulent samples were clustered on the right arm of the dendrogram (numbered as 2). In this way, authentic rose essential …