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High resolution melting analysis to discriminate Artichoke (*Cynara scolymus*) in plant food supplements

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Artichoke (*Cynara scolymus* L.) is a medicinal plant mainly used for its antioxidant, diuretic, choleric and hepatoprotective properties, being frequently included in weight-loss plant food supplements (PFS) (Lattanzio et al, 2009). PFS are legally considered as foods under EU Directive 2002/46/EC, which means that PFS are not submitted to any safety assessment prior to their commercialisation. This can lead to adulteration issues, such as accidental swap of plants or deliberate substitution of high value plant material by other species of lower cost. In order to ensure consumer's safety, the development of analytical methods for plant species identification in complex matrices has become crucial. So far, DNA-based methods have been reported as the most adequate tools for plant authentication (Kazi et al, 2013). Thus, the main goal of the present study was to discriminate *C. scolymus* from other *Cynara spp.* in PFS by real-time polymerase chain reaction (PCR) coupled to high resolution melting (HRM) analysis. For this purpose, different *Cynara* species (*C. scolymus*, *C. cardunculus*, *C. humilis* and *C. syriaca*) were obtained from Portuguese, Spanish and French germplasm banks. A total of eight PFS samples containing artichoke were acquired at local herbal stores. DNA from plant material and PFS was extracted using the commercial NucleoSpin Plant II kit. The specificity and sensitivity of the designed primers targeting the *C. scolymus* (GenBank EU744973.1) were assayed by qualitative and real-time PCR. Prior to the specific amplification of *C. scolymus*, DNA extracts were positively tested targeting an universal eukaryotic sequence (18S rRNA gene). The application of the specific PCR assay was successful for the detection of the genus *Cynara* in some of the PFS samples. The results of real-time PCR coupled to HRM analysis showed that different *Cynara spp.* were included in three distinct clusters with a level of confidence above 99.4%, thus discriminating artichoke from other *Cynara* species. The proposed HRM analysis allowed confirming the unequivocal presence of *C. scolymus* in the tested PFS with high level of confidence (>98.8%). To our knowledge, this is the first successful attempt for the rapid discrimination of *C. scolymus* in PFS.

Keywords: *Cynara spp.*, Plant Food Supplements, DNA-based methods, authentication

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