Assessing the quality and patulin contamination in infected traditional and commercial apple fruits

Ante Lončarić [1*], Ana-Marija Gotal Skoko [1], Goran Fruk [2], Antun Jozinović [1], Ivana Tomac [1] and Tihomir Kovač [1]

[1]Josip Juraj Strossmayer University of Osijek, Faculty of Food Technology Osijek, Franje Kuhača 18, 31000 Osijek, Croatia; ante.loncaric@ptfos.hr (A.L.); amgotal@ptfos.hr (A-M.G.S.); antun.jozinović@ptfos.hr (A.J.); ivana.tomac@ptfos.hr (I.T.); tihomir.kovac@ptfos.hr (T.K.)
[2]University of Zagreb, Faculty of Agriculture, Svetošimunska cesta 25, 10000 Zagreb, Croatia; gfruk@agr.hr (G.F.)
*Correspondence: ante.loncaric@ptfos.hr (A.L.)

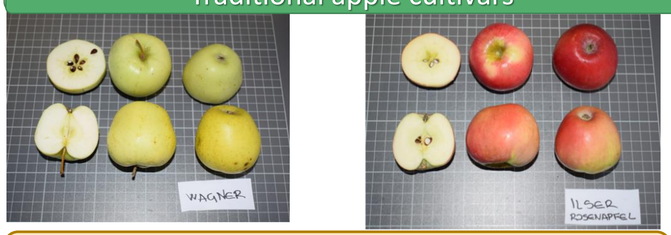
ABSTRACT

This study investigates the significance of various parameters, including water content, total acid content, sugar content, polyphenol content, antioxidant activity, anthocyanin content, and flavonoid content, on the quality of apples and their resistance to *Penicillium expansum* infection and subsequent patulin production. The research was conducted on four apple cultivars, namely, the traditional cultivars 'Wagener' and 'Ilzer Rosenapfel', and the commercial cultivars 'Jonagold' and 'Idared'. The results of the study provide valuable insights into the composition, quality attributes, and potential resistance to *Penicillium expansum* infection among different apple cultivars. These findings have implications for the selection and cultivation of apple cultivars with desirable characteristics, such as taste, antioxidant potential, and reduced mycotoxin contamination.

MATERIALS AND METHODS

Materials

Traditional apple cultivars



Commercial apple cultivars

The old apple cultivars 'Wagener' and 'Ilzer Rosenapfel' were collected at Šašinovec (45°85'00.3"N, 16°17'75.2"E); and commercial apple cultivars "Idared' and 'Jonagold' at Novaki Bistranski, Donja Bistra (45°54'57.0"N 15°52'56.0"E). All studied apple cultivars were authenticated by a pomologist and confirm by 12 SSR markers.

Chemical parameters

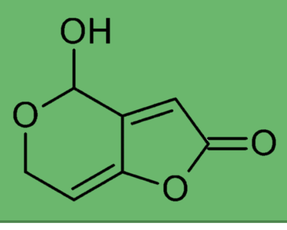
The amount of total acids was determined by the titration method with 0.1 M NaOH and expressed in percentage (as apple acid) according to AOAC 932.14c (AOAC, 1999). The soluble solids were determined from the apple juice from the bottom half of the fruit in which the proportion of soluble solids will be determined by a digital refractometer (Atago Co., Ltd., Tokio, Japan) and expressions in Brix (° Brix). The total dry matter (U.S.T.), i.e. the water content in apples, was determined by lyophilization to constant mass. Dehydration was conducted by Alpha 2-4 LSCplus (Christ, Alpha LSCplus) freeze-dryer. Sublimation was carried out at temperatures from -35 ° C to 0 ° C and a pressure of 0.22 mbar, final drying and isothermal desorption was carried out at temperatures of 0 ° C to 20 ° C and a pressure of 0.065 mbar (Lončarić et al., 2014). Total sugars was determined volumetric by Luff-Schoorl, described in Lončarić et al. (2014).

Determination of bioactive profile

The extraction was carried out from healthy fruits. Fruits were crushed and frozen at -80 °C as soon as possible, after which the samples was lyophilized in the Alpha 2-4 LSCplus freeze-dryer (Christ, Alpha LSCplus,) and milled into a powder. The polyphenols were extracted from the obtained powder with methanol-acidified with hydrochloric acid (1%) in an ultrasonic bath, centrifuged in a centrifuge (Thermo Scientific, SL 8R,) and filtered through syringe filters PTFE 0.45 µm (Lončarić et al., 2014).
The total polyphenol content was determined by the Folin-Ciocalteu method by measuring colour intensity at a wavelength of 765 nm on spectrophotometer (Lambda 365, Perkin Elmer) (Lončarić et al., 2014).
Total flavonoids were determined by a method based on the reaction of flavone and flavonol with aluminum ions and measuring absorbance at 420 nm. Concentrations of total flavonoids was converted from the previously prepared calibration curves with quercetin (Lončarić et al., 2016).
Total anthocyanins was determined using the pH-differential method described by Giusti and Wrolstad [7]. Antioxidant activities was determined by a DPPH method following procedure described by Brand-Williams, 1995 and modified by Lončarić et al., 2016

Patulin determination

Before patulin determination, apple fruit (10 pcs) were sterilized by immersion for 1 min in 2% sodium hypochlorite solution, after which apples were infected with a prepared spore suspension (20 µL) of *P. expansum* with a density of 2.5 x 106 spores / mL. Apples were kept in refrigerator until maximum infection is reached. Upon reaching the maximum diameter of the colony, the infected apple sample were excluded and subjected to the determination of the produced patulin concentration. The produced concentration of patulin was determined according to the multimycotoxin "diluent and shoot" LC-MS / MS method described by Skoko et al., 2022.



RESULTS

Table 1. Physicochemical parameters of traditional and commercial apple fruits

Apple cultivar	Total soluble matter (%)	Water content (%)	Total sugar content (%)	Total acid content (g/100g)
´Wagener´	14.4	83.5 ± 0.06	13.62 ± 0.03	0.08
´Ilzer Rosenapfel´	15.9	80.46 ± 0.11	15.22 ± 0.03	0.08
´Jonagold´	15.1	82.99 ± 0.28	14.95 ± 0.03	0.05
´Idared´	14.5	83.02 ± 0.60	13.87 ± 0.00	0.13

Table 2. Bioactive profile of traditional and commercial apple fruits

Apple cultivar	Total polyphenol content (mg/kg)	DPPH (mmol TE/kg)	Total antocyanin content (mg/kg)	Total flavonoid content (g CE/kg)
´Wagener´	421.38 ± 9.44	0.17 ± 0.00	0.00	67.30 ± 3.11
´Ilzer Rosenapfel´	707.63 ± 22.81	0.29 ± 0.00	5.01	187.97 ± 3.73
´Jonagold´	552.63 ± 29.29	0.29 ± 0.01	5.76	104.74 ± 3.11
´Idared´	550.13 ± 25.23	0.27 ± 0.02	1.24	139.77 ± 0.23

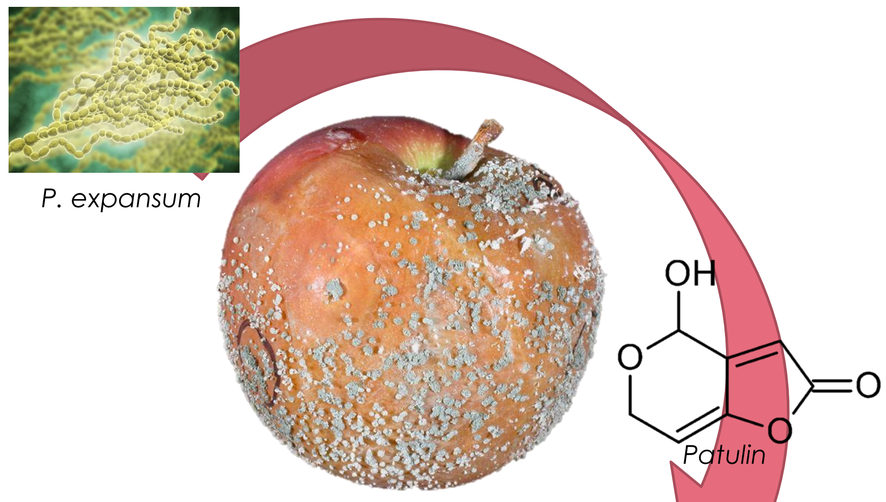
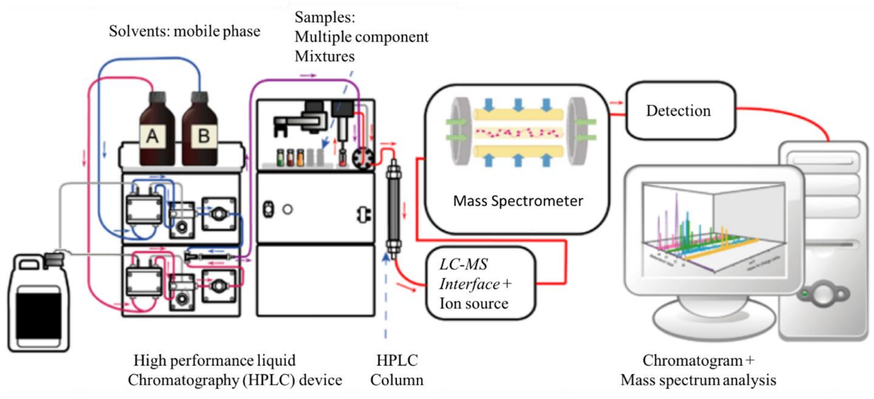


Table 3. The patulin content in infected traditional and commercial apple fruits

Apple cultivar	Patulin µg/kg
´Wagener´	18592 ± 101.82
´Ilzer Rosenapfel´	130.92 ± 0.06
´Jonagold´	292.56 ± 20.93
´Idared´	4732.4 ± 57.10



CONCLUSIONS

In conclusion, our comparative analysis elucidates differences in the phytochemical composition of traditional ('Wagener' and 'Ilzer Rosenapfel') and commercial ('Jonagold' and 'Idared') apple cultivars and their resistance to *P. expansum* infection and subsequent production of patulin. The study showed that cultivars with lower total soluble matter, sugar content, total polyphenol content, antioxidant activity and anthocyanin content exhibit higher patulin levels.

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