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Reduction of patulin in apple juice after treatment with SO_2 and heat

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Abstract

The aim of this study was to investigate the patulin concentration in apple juice commercialized produced in Mashhad city of Iran after treatment with So_2 and heat. Apple Juice samples were analyzed for patulin using reversed-phase high performance liquidated chromatography. The results of this study showed that temperature (90, 107.5 and 125°C for 30 & 268 min) and SO_2 (0 & 350 ppm) have significant effect on reducing patulin amount. The most effective condition in decreasing patulin was observed at 125°C and 350 ppm which resulted in about 86% reduction of patulin. Results showed that heat and SO_2 are the most effortless methods that be recommended for fruit juice industries in preventing infection which has got a major role in the product of patulin in apple juice.

Keywords: Patulin, apple, juice industry, SO_2, heat, mycotoxin treatment.

Introduction

Patulin (4-hydroxy-4H-furo 1[3, 2-c] pyran -2 (6H)-one] is a secondary metabolite produced by some species of *Aspergillus, Byssochlamys* and *Penicillium* (AOAC, 2000). Apple and apple products are excellent substrates for *Penicillium expansum,* the causal agent of blue mold, to produce the mycotoxin (Gilbert & Pobluad, 2003). The patulin can be used as an indicator of quality of processed apple juice and appreciable concentration of the myctoxin remains in the product after processing (WHO, 1995; Sylos & Amaya, 1999; Kadakal & Nas, 2002; Drusch & Ragab, 2003; Ritieni, 2003).

Acute symptoms of patulin consumption can include agitation, convulsions, edema, ulceration, intestinal inflammation and vomiting (Speijers, 2004). Chronic health effects of patulin include genetoxicity, immune toxicity, and neurotoxicity in rodents while its effects on humans are not clear yet (Wouters & Speijers, 1996).

Table 1. The comparison of mean of patulin conc. after treatment with various parameters (Duncken test)

Sample treatment			Patulin conc. 0/5 (µg/l) in control sample	% reduction
Temp. (°C)	SO_2 (ppm)	Time (min)		
125	350	268	0.071	85.5
107.5	350	268	0.113	77.4
125	0	268	0.146	70.8
107/5	0	268	0.173	65.4
125	350	30	0.192	61.6
107.5	350	30	0.213	57.4
125	0	30	0.278	44.4
90	0	268	0.289	42.2
90	350	30	0.389	22.2
107.5	0	30	0.424	15.2
90	0	30	0.50	0

The maximum permitted level of patulin in fruit juices and nectars and then particularly in apple juice ingredients of other beverages marketed in Europe is 50 µg kg^{-1} (EC, 2003). The joint FAO/WHO expert committee on food additives (JEFCA) established a provisional maximum tolerable daily intake (PMTDI) of 0.4 µg/Kg^1 body wt. d^{-1} based on a un-observable effect level (NOGL) of 43 µg Kg^{-1} body wt. d^{-1} and a safety factor of 100 (WHO, 1995). Several analytical methods, based on high-performance liquid chromatography (HPLC), have been developed for detecting and determining of patulin in apple products. The aim of this study was to investigate the decrease of patulin content in apple juice commercialized in Mashhad fruit juice factories after treatment with heat and Sulfur-dioxide in different times.

Materials and methods

Samples of apple juice (Brix 12, pH=4) were prepared from Shahd-e-Iran factory in Mashhad.

Chemical reagents

Patulin standard (Sigma comp.) acetic acid and ethyl acetate reagent, HPLC grade, sodium bicarbonate, Natrium sulfite (Merck comp.) Tetrahydrofuran and natrium acid, water milli-Q water, as mobile phase.

Methods

HPLC used with model LC-4A Shimadzu, LC column shim-pack CLC-ODS and guard column ZAS- GLC-ODS (spectrophotometric detector SPD and 254 nm bond pack 18). Flow rate was set 1ml/1min and patulin elutes is about 9-10 min. Analytical procedure and treatment standard solution of patulin was prepared form the pure crystal of patulin and with UV apparatus, next to a sample of apple juice containing patulin (0/5 µg as control and temperature levels treatments were 90°C as pasteurization of apple juice for 30 min in factories 125°C for 268 min because 90% of patulin concentration decline in this temperature (Lovett *et al.*, 1975) and 107.5°C as

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the moderate temperature were applied. Also, SO_2 has been used 350 ppm because maximum amount allowed in apple juice, the amount of patulin in the injected samples was determined by using a calibration curve concentration of patulin, initially the standards and then samples are put respectively in auto sample in HPLC, samples are taken absolutely at completely randomly statistical analysis with three replication and factorial design then compared with Dunkan's test.

Results

Results showed that heat by itself and with interaction with SO_2 had significant effect on reduction of patulin and with more increase temperature patulin more reduce, also using SO_2 (350 ppm, in comparison with lack on reduction of patulin. Fig. 1 and 2 showed interactions of temperature and SO_2 the results illustrated that presence of so_2 and higher temperature was gotten the most effect on reduce of patulin concentration in comparison with the model sample (Table 1).

To sum it up, heat as pasteurization is not considered as an eradication factor on patulin concentration in apple juice (Table 1). Besides, high temperature as used in study 125°C decrease of patulin concentration, about 85.5%, but caused low quality of apple juice and deforms the color and clarity of it.

Discussion

Overall the results indicated that pasteurization of apple juice at 90°C cannot be considered enough as an eradication factor to reduce the patulin concentration in apple juice. These results are in line with the earlier finding, which reported that thermal processing causes only moderate reductions and patulin in apple juice will survive the pasteurization process (Harrison, 1989). Treatment at higher temperature (125°C) for 268 min alone and in combination with SO_2 treatment reduced the patulin content in apple juice by 70.8% and 85.5% respectively. The use of SO_2 has been reported to inhibit the biological activity of patulin (Steglich *et al.,* 2000). However, in this study the treatment of apple juice at higher temperature together with SO_2 led to deformation of its color and reduced its clarity and overall caused a reduction in the quality of apple juice. The benefits of using SO_2 together with heat treatment in reducing the patulin concentration in apple juice while preserving its quality and appearance needs further research before any recommendations can be made to the fruit juice industry for its commercial application.

Fig. 2. Effect of interaction of heat and SO_2 on reduction of putulin conc.

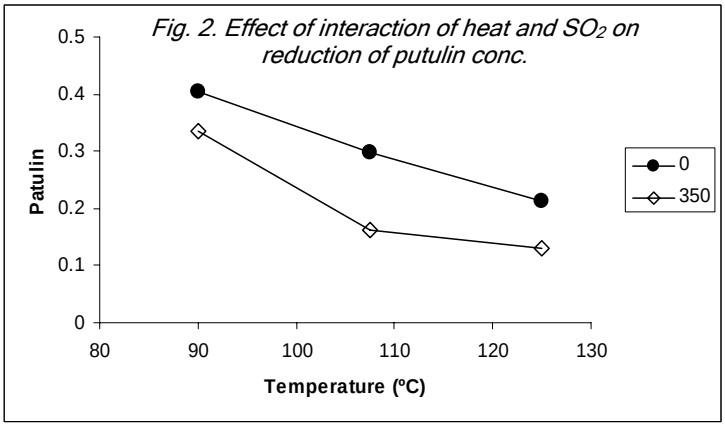
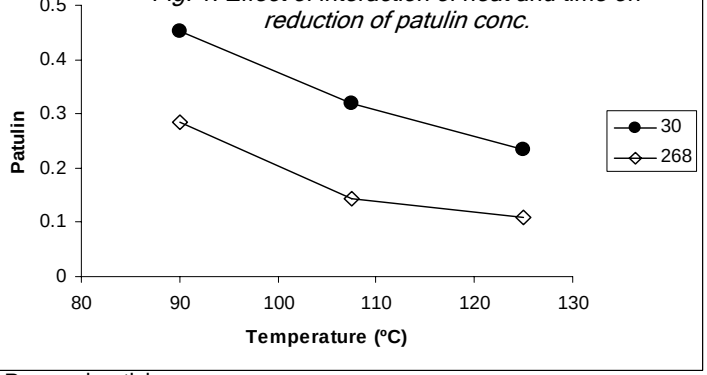


Fig. 1. Effect of interaction of heat and time on reduction of patulin conc.



Conclusion

To sum it up, heat as pasteurization is not considered as an eradication factor for patulin in apple juice but SO_2 treatment is beneficial. The combined treatment of heat and SO_2 can be recommended as the most effortless method for apple fruit juice industries.

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