Bulletin of Environment, Pharmacology and Life Sciences Bull. Env. Pharmacol. Life Sci., Vol 11 [8] July 2022 : 72-74 ©2022 Academy for Environment and Life Sciences, India Online ISSN 2277-1808 Journal's URL:http://www.bepls.com CODEN: BEPLAD ORIGINAL ARTICLE



# Organic acids suppress aflatoxin production in kalonji (*Nigella sativa* L.) seeds

## Chitra Kumari and Shyam Govind Singh

Department of Botany, Agra College, Agra (U.P.) India

#### ABSTRACT

Kalonji (Nigella sativa L.) is one of the most important spices, which is used in pickels, bakery products and culinary purposes. The seed samples of kalonji were found to be naturally contaminated with aflatoxin  $B_1$ ,  $B_2$  and  $G_1$ . Some organic acids i.e., benzoic acid, lauric acid, propionic acid, sodium benzoate and sorbic acid have been screened to inhibit mould incidence and aflatoxin in kalonji seeds. The maximum inhibition of mould and aflatoxin  $B_1$  contamination was recorded at 0.4% concentration of propionic acid and the minimum inhibition of mould and aflatoxin contamination was noted with sodium benzoate. The benzoic acid inhibited aflatoxin  $B_2$  and  $G_1$  at the concentration of 0.4% but aflatoxin  $B_1$  could be inhibited only upto the extent of 96.60% at 0.5% concentration and the mould count was also completely suppressed at that concentration. More or less similar findings have been reported for lauric acid and sorbic acid. These acids completely inhibited aflatoxin  $B_2$  and  $G_1$  at the concentration  $B_1$  was inhibited to the extent of 94.70 and 95.80 at 0.5% concentration respectively. Thus, propionic acid at 0.4% can be safely used for complete inhibition of moulds and aflatoxin in kalonji seeds.

Keywords : Aflatoxin, kalonji seeds, organic acids

Received 16.04.2021

Revised 22.06.2021

Accepted 27.07. 2022

#### INTRODUCTION

Kalonji (*Nigella sativa* L.) is an important spice crop of the world and is much used in pickles, culinary purposes and food preservative in India. It has a broad range of medicinal use. The seeds are commonly contaminated with fungi during storage.

During storage, species of *Aspergillus, Penicillium, Fusarium* and *Alternaria* are more frequently associated with its seed. The fungal contamination causes decrease in weight, nutrition value, germinability [13] and also produce highly toxic secondary metabolites known as mycotoxins.

Among the mycotoxins, aflatoxins are the most dangerous and highly toxic secondary metabolites produced by *Aspergillus flavus* and *A. parasiticus* [10]. Aflatoxin  $B_1$ ,  $B_2$ ,  $G_1$  and  $G_2$  are synthesized by these fungi, among these toxins, aflatoxin  $B_1$  is highly toxic and has carcinogenic nature and is considered as the most carcinogenic compound by IARC the International Agency for Research as Cancer, [3,4].

Consumption of aflatoxin contaminated food by human beings and animals lead to many pathological disorders, immune suppression [14], liver cancer [8], liver cirrohsis followed by rapid death [2].

Recently, Singh and Cotty [12] noted the aflatoxin B and G in dried red chilies (*Capsicum* spp.) Wagas *et al.* [15] recorded aflatoxin  $B_1$  in all edible vegetable seeds samples. In view of the growing concern about health hazards of aflatoxins, present investigation was carried out to find the effect of some organic acids on fungal count and aflatoxin production by *Aspergillus flavus* and *A. parasiticus* on kalonji seeds.

#### MATERIAL AND METHODS

In the present investigation seed samples from traders were collected and treated with 5 organic acids viz., benzoic acid, lauric acid, propionic acid, sodium benzoate and sorbic acid at the rate of 0.1, 0.2, 0.3, 0.4 and 0.5% by weight following Rani and Singh [11]. After adding the chemicals, the seeds were shaken thoroughly to disperse organic acids uniformly and then stored at room temperature for 6 months. Untreated seeds served as control. After the stipulated storage period both treated and untreated seeds were analysed for the association of fungi and aflatoxin contamination.

The fungi associated with seeds were isolated by seed plating method following rules of ISTA [5] using PDA medium for each seed sample, 200 seeds were taken and plated at the rate of 10 seeds per plate on PDA medium and incubated at  $28 \pm 2^{\circ}$ C for 6 days in an alternating cycle of 12 hours fluorescent light and 12 hours darkness. After incubation period the seeds were examined under stereobinocular microscope

#### Kumari and Singh

for the presence of fungi and finally fungal population was determined. Further, seed samples were analysed for presence of aflatoxin by thin layer chromatography following method outlined by Jones [6].

#### **RESULTS AND DISCUSSION**

Among all the organic acids, propionic acid was found to be most effective organic acid in reducing fungal contamination as well as aflatoxin production. The propionic acid completely inhibited the production of aflatoxin  $B_1$ ,  $B_2$  and  $G_1$  at 0.4% concentration and this acid completely inhibited fungal incidence at 0.5% concentration . However, 0.1% concentration of propionic acid completely inhibited aflatoxin  $G_1$  production.

Furthermore, aflatoxin  $B_2$  could not be reported beyond 0.1% concentration but aflatoxin  $B_1$  has been reported upto 0.3% concentration of propionic acid.

After propionic acid, benzoic acid was the next effective organic acid. It inhibited aflatoxin  $B_2$  at 0.4% concentration and aflatoxin  $G_1$  was completely inhibited at 0.1% concentration but aflatoxin  $B_1$  could be inhibited only upto the extent of 96.60% even at 0.5% concentration.

Other organic acids i.e. lauric acid, sodium benzoate and sorbic acid entirely inhibited aflatoxin  $B_2$  at 0.4% concentration, and aflatoxin  $G_1$  was totally inhibited at 0.3% concentration in case of sorbic acid and lauric acid. but aflatoxin  $B_1$  could not be inhibited totally even at 0.5% concentration of these organic acids. The least effective organic acid was sodium benzoate, which inhibited aflatoxin  $B_1$  to the extent of 93.80 only. Thus it can be concluded that propionic acid is the best organic acids like benzoic acid (96.60%), sorbic acid (95.80%), lauric acid (94.70%) and sodium benzoate (93.80%) can reduce aflatoxin  $B_1$  contamination at 0.5% concentration. Thus, it can be safely recommended for prevention of moulds and aflatoxins in kalonji seeds.

S. N.	Organic Acid	Conc. of organic acid (%)	No. of moulds	Amount of aflatoxin (in ppb)				% inhibition in aflatoxin production			
				<b>B</b> <sub>1</sub>	<b>B</b> <sub>2</sub>	G1	G <sub>2</sub>	<b>B</b> <sub>1</sub>	<b>B</b> <sub>2</sub>	<b>G</b> 1	<b>G</b> <sub>2</sub>
1.	Control	-	32	2050	640	520	-				
	Propionic acid	0.1	18	820	110			65.57	84.79	100.00	-
		0.2	14	670				73.27	100.00	100.00	-
2.		0.3	08	150				91.50	100.00	100.00	-
		0.4	01					100.00	100.00	100.00	-
		0.5	-					100.00	100.00	100.00	-
	Benzoic acid	0.1	20	870	130			62.02	70.59	100.00	-
		0.2	18	740	85			65.71	85.40	100.00	-
3.		0.3	10	700	70			69.06	91.32	100.00	-
		0.4	08	250				87.36	100.00	100.00	-
		0.5	-	90				96.60	100.00	100.00	-
	Sorbic Acid	0.1	25	950	180	180		58.89	52.00	68.70	-
		0.2	20	820	120	70		64.57	78.90	84.20	-
4.		0.3	15	740	85			67.58	88.64	100.00	-
		0.4	03	400				80.34	100.00	100.00	-
		0.5	01	120				95.80	100.00	100.00	-
	Lauric Acid	0.1	22	1100	340	200		54.03	44.80	50.00	-
		0.2	16	860	180	100		64.40	52.00	78.00	-
5.		0.3	10	780	90			69.56	87.40	100.00	-
		0.4	02	450				82.42	100.00	100.00	-
		0.5	-	110				94.70	100.00	100.00	-
	Sodium Benzoate	0.1	20	1120	430	300		52.00	45.06	48.00	-
		0.2	15	900	180	150		58.20	50.00	75.00	-
6.		0.3	10	800	100	60		66.75	84.79	84.00	-
		0.4	04	500				78.64	100.00	100.00	-
		0.5	01	140				93.80	100.00	100.00	-

 Table 1 : Efficacy of some organic acids against moulds and aflatoxin contamination in Kalonji seeds.

The propionic acid at 0.9% concentration significantly reduced aflatoxin [7]. At 0.5% concentration, propionic acid entirely inhibited fungal growth, Likewise, benzoic acid and sorbic acid at same concentration also inhibited aflatoxin production [9].

#### Kumari and Singh

The propionic acid, benzoic acid, sodium benzoate, lauricidin and sorbic acid inhibited mould infestation and aflatoxin production in Isabgol husk (*Plantago ovata* Forsk.) [1].

Therefore, the above document makes it clear that some organic acids show inhibitory effect for mould incidence and aflatoxin production during storage of seeds. Furthermore, they are ecofriendly, cheap, have no residual toxicity and used as food preservatives, thus can be safely used for stored edible seeds like kalonji and other species.

### ACKNOWLEDGMENTS

Authors are highly thankful to Dr. Anurag Shukla, the Principal, Agra College, Agra and to Dr. P. B. Jha, the head of the Botany Department for providing facilities and constant encouragement. Thanks are also due to Dr. Surendra Singh for critical suggestions.

#### REFERENCES

- 1. Agrawal R. and Singh S. (2017). Effects of organic acids on aflatoxin in isabgol (*Plantago ovata* Forsk). *Indian Research Journal of Genetics and Biotechnology*, 9(02): 236.
- 2. CDU (2004). Out break of aflatoxin poisoning-eastern and central provinces, Kenya, January-July 2004, *Morb. Mortal. wkly Rep.* 53, 790-793.
- 3. IARC (1982). Monographs on the evaluation of the carcinogenic risk of chemicals to humans, supplement No. 4, IARC, 1-29. Lyon : International Agency for Research on Cancer.
- 4. International Agency for Research on Cancer (2002). Traditional herbal medicines, Some mycotoxins, naphthalene and styrene. In : IARC monograph on the evulation of carcinogenic risk of chemicals to humans, 82.
- 5. ISTA (1966). International rules for seed testing. *Proc. Int. Seed. Test. Association*, 32: 1-152.
- 6. Jones B.D. (1972). Methods of aflatoxin analysis. *Bulletin Tropical Products Institute*, London, 9-70.
- Jubeen F., Sher F., Hazafa A., Zafar F., Ameen M. and Rasheed T. (2020). Evaluation and detoxification of aflatoxins in groundnut and tree nuts using food grade organic acids. *Biocatalysis and Agricultural Biotechnology*, 29.
- 8. Liu and Wu, F. (2010). Global burden of aflatoxin-induced hepatocellular carcinoma : a risk assessment. *Environ. Health Perspect.* 118: 818-824.
- 9. Moon Y.S., Kim H.M., Chun H.S. and Lee S.E. (2018). Organic acids suppress aflatoxin production via lowering expression of aflatoxin biosynthesis-related genes in *Aspergillus flavus*, *Food Control*, 88(2) : 207-216.
- 10. Payne, G.A. and Brown, M.P. (1998). Genetics and physiology of aflatoxin biosynthesis. *Annu. Rev. Phytopathol.* 36: 329-362.
- 11. Rani N. and Singh S. (1992). Effect of lauricidin derivatives on aflatoxin elaboration by *Aspergillus flavus, Nat. Acad. Sci. Letters*, 15(1): 3-5.
- 12. Singh, P. and Cotty P.J. (2019). Characterization of *Aspergilli* from dried red chilies (*Capsicum* spp.) : insights into the etiology of aflatoxin contamination, *International Journal of Food Microbiology*, 289: 145-153.
- 13. Sinha K.K. and Sinha A.K. (1993). Effect of aflatoxin B1 on germination index and seedling growth in wheat varieties. *Mycotoxicoses and Mycotoxins*, 123: 165-169.
- 14. Turner, P.C. Moore, S.E., Hall, A.J., Prentice A.M., Wild C.P., (2003). Modification of immune function through exposure to dietary aflatoxin in Gambian children, *Environ, Health Perspect*, 111: 217-220.
- 15. Waqas, M., Iqbal, S.Z., Abdul Razis A.F.A., Pervaiz W., Ahmad T., Usman S., Ali N.B. and Asi M.R. (2021). Occurrence of aflatoxins in edible vegetable seeds and oil samples available in Pakistani retail markets and estimation of dietary intake in consumers, International Journal of Environmental Research and Public Health, 18: 8015.

## **CITATION OF THIS ARTICLE**

C Kumari and S G Singh. Organic acids suppress aflatoxin production in kalonji (*Nigella sativa* L.) seeds.. Bull. Env.Pharmacol. Life Sci., Vol 11 [8] July 2022 :72-74