

INHIBITION OF GROWTH AND TOXIN PRODUCTION BY LAURIC ACID DERIVATIVES IN *ASPERGILLUS* SPECIES

G. RAMA DEVI AND H. POLASA

Department of Microbiology, Osmania University, Hyderabad 500 007, India

SEVERAL approaches¹⁻³ have been suggested to protect foods and feeds from mold growth and toxin production. One of them is to preserve the food materials with food-grade chemicals which appear to be safe because of their non-toxic nature to the consumer. Reports are available showing the inhibition of growth and toxin

production by some fatty acid derivatives⁴, antioxidants⁵ and certain natural substances; like lauricidin (a monoglyceride of lauric acid) and butylated hydroxy anisole.

The monolaurin derivatives of lauric acid viz lauricidin-012, lauricidin-812 and lauricidin-1012 are considered as generally recognised as safe (GRAS) chemicals. The efficacy of these compounds as antifungal agents and toxin inhibitors is not known. Their efficacy as antifungal agents was therefore studied with special reference to three toxigenic *Aspergillus* species of *A. flavus*, *A. ochraceus* and *A. versicolor* (isolated from maize) which produced aflatoxin, ochratoxin and sterigmatocystin, respectively⁶. These were grown on

Table 1 Effect of lauricidin-012, lauricidin-812 and lauricidin-1012 on growth and toxin production by *Aspergillus* species

Compound	Conc. (ppm.)	Growth substrate	<i>A. flavus</i>		<i>A. ochraceus</i>		<i>A. versicolor</i>	
			Growth (mg)	Aflatoxin B ₁ (µg)	Growth (mg)	Ochratoxin A (µg)	Growth (mg)	Sterigmatocystin (mg)
Control	0	Liquid medium (LM)	2519 ± 93	208.6 ± 6.5	2394 ± 13	114.5 ± 2.2	2119 ± 29	197.7 ± 3.0
		Maize (M)	55.5 ± 0.5	42.2 ± 1.6	56.8 ± 1.0	46.8 ± 1.5	89.0 ± 1.0	53.5 ± 1.5
		Maize flour (MF)	66.0 ± 0.7	50.5 ± 0.2	60.5 ± 2.0	48.0 ± 1.2	118.5 ± 1.0	67.5 ± 0.2
Lauricidin-012	250	LM	604 ± 18	Traces	497 ± 52	Traces	206 ± 36	0
		M	36.9 ± 1.0	26.5 ± 4.5	42.5 ± 1.0	20.5 ± 1.0	62.0 ± 0.5	24.0 ± 1.5
		MF	68.0 ± 0.2	54.5 ± 0.5	62.5 ± 0.2	44.0 ± 0.5	120.0 ± 0.6	70.0 ± 2.0
	500	LM	0	0	0	0	0	0
		M	23.5 ± 1.0	Traces	21.5 ± 0.4	0	32.5 ± 1.5	0
		MF	47.0 ± 0.8	32.5 ± 1.6	44.0 ± 0.4	Traces	83.5 ± 0.5	32.0 ± 0.2
1000	LM and M	0	0	0	0	0	0	
	MF	28.0 ± 0.5	0	21.0 ± 0.5	0	2.5 ± 0.0	0	
	LM, M and MF	0	0	0	0	0	0	
Lauricidin-812	500	LM	139 ± 26	Traces	818 ± 11	26.1 ± 1.0	0	0
		M	45.5 ± 1.0	33.5 ± 2.0	40.0 ± 1.0	20.6 ± 0.5	56.3 ± 1.0	ND
		MF	39.0 ± 0.6	22.5 ± 1.6	67.5 ± 0.4	48.5 ± 0.5	98.0 ± 1.0	32.0 ± 1.5
	1000	LM	0	0	92.0 ± 0	0	0	0
		M	26.5 ± 1.0	0	22.0 ± 0.5	0	34.0 ± 0	0
		MF	39.0 ± 0.0	22.5 ± 0.5	52.0 ± 0.5	30.0 ± 1.0	60.0 ± 1.0	0
2000	LM and M	0	0	0	0	0	0	
	MF	32.0 ± 1.5	0	33.0 ± 1.0	0	12.0 ± 0	0	
	LM and M	0	0	0	0	0	0	
3000	MF	9.0 ± 0	0	13.0 ± 0	0	0	0	
	LM, M and MF	0	0	0	0	0	0	
	0	0	0	0	0	0	0	
Lauricidin-1012	500	LM	366 ± 48	Traces	521 ± 40	Traces	191 ± 16	0
		M	30.0 ± 1.0	Traces	25.0 ± 0.5	11.0 ± 0.0	65.0 ± 2.0	30.0 ± 1.0
		MF	94.0 ± 1.0	27.50 ± 0.4	50.0 ± 1.6	36.0 ± 1.0	98.4 ± 1.5	32.5 ± 1.5
	1000	LM	0	0	0	0	0	0
		M	15.0 ± 0.5	0	7.5 ± 0.0	0	40.0 ± 1.0	Traces
		MF	27.0 ± 0.5	Traces	34.0 ± 3.0	Traces	51.0 ± 1.8	Traces
2000	LM and M	0	0	0	0	0	0	
	MF	19.0 ± 0.5	0	16.0 ± 0.0	0	22.0 ± 1.0	0	
	LM, M and MF	0	0	0	0	0	0	

Note: One 100 ml of liquid medium and 50 g of maize and flour each were used in all the experiments. The above values represent the mean of six experiments with standard deviation (±).

sterilized chemically defined media⁷⁻⁹, maize grain and maize flour, the latter two usually found naturally contaminated with these fungi and toxins.

Each test compound was incorporated at various concentrations in the above growth substrates and were inoculated with 1 ml of spore suspension of the test organism containing 10⁵ spores. These were incubated for 8, 10 and 14 days, at which optimal growth and toxin were recorded for *A. flavus*, *A. ochraceus* and *A. versicolor* respectively. After the incubation period, dry weights of the mycelial mats were taken by separating the mycelial growth from the liquid phase and drying them at 100°C for 24 hr. Fungal growth on the two solid substrates (maize and its flour) was estimated biochemically by assaying the amount of glucosamine by employing known methods¹⁰. Toxin was estimated^{11,12} by the method of TLC using known toxin standards.

The results showed that lauricidin-012, lauricidin-812 and lauricidin-1012 were effective inhibitors of fungal growth and toxin production by the three *Aspergilli*, when grown on the three different growth substrates as shown in table 1.

It may be noted that higher concentration of each test compound was required for inhibition of both growth and toxin production when the three test fungi were grown on maize flour than on maize grain and liquid medium. Further 100% toxin production was inhibited by all the three compounds at lesser concentrations than required for growth inhibition. Similar results were reported⁴ in the case of biosynthesis of aflatoxin on liquid medium by sorbic acid and derivatives of fatty acids.

Among the three lauric acid derivatives lauricidin-

012 was more effective in inhibiting the growth and toxin production by the three test fungi (table 1).

The authors thank Prof. J. J. Kabara, Department of Bio-mechanics, Michigan State University for supplying lauricidin 012, 812 and 1012.

1 December 1983; Revised 10 May 1984

1. Applegate, K. L. and Chipley, J. R., *Mycologia*, 1973, **66**, 436.
2. Applegate, K. L. and Chipley, J. R., *J. Appl. Bacteriol.*, 1974, **37**, 359.
3. Uraiah, N., Cassity, T. R. and Chipley, J. R., *Can. J. Microbiol.*, 1977, **23**, 1580.
4. Chipley, J. R., Story, L. D., Todd, P. T. and Kabara, J. J., *J. Food. Safety*, 1981, **2**, 109.
5. Chang, H. C. and Branen, A. L., *J. Food. Sci.*, 1975, **40**, 349.
6. Rama Devi, G. and Polasa, H., *Curr. Sci.*, 1982, **51**, 751.
7. Reddy, T. V., Vishwanathan, L. and Venkatasubramanian, T. A., *Appl. Microbiol.*, 1971, **22**, 393.
8. Ferriera, N. P., *Antonie Van Leeuwenhoek*, 1968, **34**, 433.
9. Hitokoto, H., Morozumi, S., Wauke, T., Sakai, S. and Yoshikawa, S., *Mycopathologia*, 1982, **78**, 99.
10. Ride, J. P. and Drysdale, R. B., *Physiol. Plant Pathol.*, 1972, **2**, 7.
11. Roberts, B. A. and Patterson, D. S. P., *J. Assoc. Official. Anal. Chem.*, 1975, **58**, 1178.
12. Seitz, L. M. and Mohr, H. E., *Cereal Chem.*, 1977, **54**, 179.