Inhibitory Effect Of Some Spices On Growth And Aflatoxin Production Aspergillus flavus In Liquid Media

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Summary:

The effect of different concentrations of rosemary cinnamon and green pepper against the growth of toxigenic isolate of Aflavus 86 76 and the production of aflatoxin. The results showed that cinnamon caused high inhibition for mycelium growth and aflatoxin production when it was added to liquid media directly. Low inhibition occurred for Mycelium growth only with 26 1.5 cinnamon and %2 rosemary No effect for mycelium growth and aflatoxin production was observed when the liquid! Media contains up to 2% green pepper. The data indicate that use of spices librates with the media were less inhibitory for mycelium growth and aflatoxin production compared with the hy spices added to the media at all concentrations used.

Introduction:

Aflatoxins are secondary metabolites produced primarily by some strains of Aspergillus flavus and A. parasiticus, which can grow in a wide variety of agricultural commodities. The presence of aflatoxin in foods and feeds is potentially hazardous to the health of both humans and animals (4, 8). Preservatives are used worldwide to protect food against microbial deterioration and to inhibit the growth of fungi and mycotoxin production. Some articles refer to the effects of some spices and herbs on microbial growth in foods (11, 1, 7). Many spices are used in foods today mainly for their flavor and aroma (11, 1). Some spices and herbs have been known to possess antibacterial activities. They contain many components such as aldehydes, phenols, esters, and essential oils, which have inhibitory properties against many kinds of foodborne bacteria (11, 3). In 1971 (12) it was reported that over 150 million pounds of spices are consumed annually in the United States, and 20% of these are used in the meat industry. Several recent articles have been known on the effect of spices on the growth of fungi and mycotoxins production (6, 5).

The objective of the present study was to obtain data on the ability of different concentrations of rosemary cinnamon and green pepper to inhibit the growth of aflatoxigenin strain of Aspergillus flavus 8676 and the production of aflatoxin B in liquid media

Materials and Methods:

Spices: ground rosemary, cinnamon, and green pepper used in this study were purchased from the local market in Basrah, Iraq, in 2003. Two procedures were employed for the preparation of the media. In the first, dry spices at various concentrations (0.5, 1, 1.5, and 2% by weight) were added to yeast extract-sucrose medium. The second dry spices (0, 0.5, 1, 1.5, and 2%) were added to distilled water heated for 10 min at 120°C and filtered through Whatmann No. 1 filter paper, and the spices filter ate was added to the media.

Media and Culture:

The aflatoxigenic isolate of Aflavus 86 76 procured from M. Saito, Japan, was used for aflatoxin production in semi-synthetic media (150 g sucrose, 20 g yeast extract, and 1 liter of distilled water). Aflatoxin production was effected at 25 ± 1 °C for 14 days in stationary culture in 250 ml Erlenmeyer flasks containing 50 ml medium.

Dry mass of mycelium:

To determine the dry weight of A. flavus mycelium at the end of the incubation period, the mycelium was separated by filtration through Whatman No. 1 and whashed twice with distilled water, and the dry mass was determined after drying at 105 °C for 12 hours (6).

Determination of Aflatoxin:

Aflatoxins were determined according to (6). The culture filtrate was extracted twice with chloroform (15 ml each). Then the combined chloroform extract was filtered through anhydrous sodium sulfate. The solvent was evaporated to dryness on a water bath. Thin layer

Chromoography was used to separate aflatoxins by using silica gel and a solvent mixture of chloroform-methanol (50:1, v/v). Aflatoxin B and B_1 were located by UV light (365 nm). Aflatoxin B and B were determined as aflatoxin B (6). The quantity of aflatoxins was estimated according to (10).

Results and Discussion:

Table 1 shows the effect of different concentrations of three spices on the growth of A. flavus and aflatoxin production in yeast extract-sucrose medium.

The degree of inhibition for mycelium growth and aflatoxin production differed according to the concentrations Kind and edding methods of spices to medium concentration (0.5 to 1.5%) of rosemary had no effect on mycelium growth and aflatoxin production, while the 2% concentration caused low inhibition for mycelium growth and no effect for aflatoxin production. The weight of dry mycelium was 1.93 g/50 ml when the media did not contain any spices (control), and reduced to 1.82 g/50 ml at 2% concentration of rosemary. High inhibition of both mycelium growth and aflatoxin production occurred at 1.5 and 2% concentrations of cinnamon. The concentration that reduced the weight of mycelium from 1.93 g/50 ml (control) to 1.25 g/50 ml was 2%, and the decrease in aflatoxin production from 118.5 ug/50 ml (control) to 95.3 ug/50 ml at the same concentration of cinnamon. Table 1. This finding agrees with those obtained from (6.5) who referred to that some spices such as cinnamon, thyme and clove contain some essential oils which caused inhibition for mycelium.

Growth aflatoxin production and the growth of dermatephyte (2) also reported that cinamon could be used for inhibition of aflatoxin. In another study, (3) concluded that cinamic aldehyde and eugenol are the major antifungal substances of cinnamon oil. In the other study, (9) referred to the inhibitory factor of cinnamon and identified it as O-methoxy cinamic aldehyde and indicated that this substance had a strong inhibitory effect on the growth of dermatophytes. No effect for aflatoxin production was observed at concentrations of 0.5% to 2% of green pepper, and less inhibition for mycelium growth was observed at a 2% concentration of green pepper, which reduced the weight of.

Table 1. Effect of dry ground spices on the growth of A. flavus and aflatoxin production in liquid media.

Concentration of spices (%) in media	Mycelium wet/50 ml	Concentration of aflatoxin B (ug/50ml)
0	1.93	118.5
Resomary 0.5	1.92	118.2
1.0	ND	111.8
1.5	1.88	116.4
2.0	1.82	116.0
Cinnamon 0.5	1.90	117.3
1.0	1.84	ND
1.5	1.50	101.2
2.0	1.24	95.3
Green pepper 0.5	1.92	118.3
1.0	1.91	ND
1.5	1.89	117.0
2.0	1.82	116.8

Not detected = ND

Mycelium from 1.93 g/50 ml (control) to 1.82 g/50 ml

Table 1.

The results obtained from this study showed (Table 2.) that the media with spices filters were less inhibiting for both mycelium growth and aflatoxin production for all concentrations of spices than those containing dry spices. These results agree with (11), who studied the sensitivity of some common food-borne bacteria to certain spices.

In conclusion, of the three spices examined for antifungal activity, green pepper was the least effective, whereas rosemary was effective against mycelium growth and cinnamon was effective on aflatoxin production in addition to mycelium growth. There was no clear effect for both mycelium growth and aflatoxin production when we added dry green pepper to liquid media.

Table 2. Effect of spices filters on the growth of A flavus and atlatoxin production in liquid media.

Concentration of spices (%) in media	Mycelium wet/50 ml	Concentration of aflatoxin B (ug/50ml)
0	1.93	118.5
Resomary 0.5	1.93	118.3
1.0	ND	117.5
1.5	1.90	ND
2.0	1.82	116.0
Cinnamon 0.5	1.92	118.3
1.0	ND	ND
1.5	1.83	118.2
2.0	1.75	110.1
Green pepper 0.5	ND	118.5
1.0	1.92	ND
1.5	ND	118.0
2.0	1.88	117.6

Not detected = ND

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التأثير التثبيطي لبعض التوابل على نمو الفطر

Aspergillus flavus

وانتاج الافلاتوكسينات في الوسط الغذائي السائل

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الخلاصة:

تم دراسة التأثير التشبيضي لتتبع مختلف التوابل المنت القرفة.

الدارسين، والفلفل الأخضر على نمو الفطر Aspergillus flavus وانتاج الافلاتوكسينات النوع B من العزلة ٧٦-٨٦ فينان المنح للأفلاتوكسينات في الوسط الصناعي مستخلص الحميرة والدكستروز.

اظهرت نتائج الدراسة استخدام 2% من أعشاب الدارسين على نمو الفطر وانتاج الأفلاتوكسينات عند اضافته مباشرة إلى الوسط الزراعي، بينما انخفضت نسبة أعشاب الدارسين إلى أقل من 1.5% من أعشاب الدارسين و2% من أعشاب القرفة للوسط الزراعي، وتم إضافة الأعشاب إلى نمو الفطر ولا على إنتاج الأفلاتوكسينات عندما احتوى الوسط على تركيز 0.5% إلى 2% من الفلفل الأخضر، وبالتالي تم إضافة الأعشاب إلى الوسط الزراعي .