

## Efficacy of GA<sub>3</sub> on Amino acids contents in Aflatoxin B<sub>1</sub> treated Maize seeds (*Zea mays* L.)

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### ABSTRACT

Effect of Aflatoxin B<sub>1</sub> & GA<sub>3</sub> Concentration at 2000ppb in alone and various combinations (*i.e.* 1:1, 1:2, 2:1, 1:3 and 3:1, V/V) on amino acids was studied in maize (C. V. Madhuri-01). The maximum and minimum inhibitions were recorded in this at 3:1 & 1:3 combinations ratio of AFT-B<sub>1</sub> and GA<sub>3</sub> respectively. However, L- Methionine, DC(+) Alanine and Aspartic acid were completely lost at 2000 ppb concentration of Aflatoxin B<sub>1</sub>.

**Keywords:** AFTB<sub>1</sub>, Amino acids (kits), Phytohormones, *Zea mays*

### INTRODUCTION

Maize is an important cereal crop of India and has been shown to be naturally contaminated with different levels of aflatoxin (Sinha, 1990; Bilgrami *et al.*, 1990; Bilgrami and Jeswal, 1992). AFTB<sub>1</sub> have been shown to influence adversely on the physiological processes of different crop system. In plants, seed generation, seedling growth as well as other physiological processes due to aflatoxin B<sub>1</sub> reported in *Zea mays*; *Vigna radiata* (Sinha and Kumari 1990; Prasad *et al.*, 1994;1995), *Sorghum* and *Maize* (Tripathi and Mishra, 1983), *Mung bean* (Sinha and Kumari, 1989) and *Mustard* and *Gram* (Sinha *et al.*,1993).

Aflatoxin B<sub>1</sub> is known to interfere with the synthesis of GA<sub>3</sub>, which stimulates the activities of Lipase and  $\alpha$ - amylase enzyme , primarily required during seed germination and seedling growth (Black and Altschul,1965; Saxena and Maheshwari,1979 and Jones *et al.*,1967) as well as enhances the fresh and dry weight of seedling (Sharma, 1978).

Since only few reports are available about the impact of GA<sub>3</sub> on aflatoxin treated seeds, an attempt has, therefore been made to investigate the effect of GA<sub>3</sub> and Aflatoxin B<sub>1</sub> in alone and in combination on amino acids in maize germinating seeds.

### MATERIALS AND METHODS

Seeds of *Zea mays* L. were obtained from Apurva seeds hybrid maize madhuri-01 from Dayal traders manures and storage, Kadirabad, Darbhanga, India. Stock (powder) solution of AFT-B<sub>1</sub> and phytohormones (GA<sub>3</sub>) were obtained from the local scientific stores, Darbhanga, India. Stock solution were initially prepared 1 cm<sup>3</sup> ethanol from which the dilution (100, 250, 500, 1000, 2000ppb) were made in sterilized distilled water solution of these toxin and phytohormone were mixed alone and in combination ratio like 1:1, 1: 2. 2:1, 1:3 & 3:1 V/V at 2000 ppb in order to record their combined effects. Seeds were steeped

initially in distilled water for one hour and subsequently in different combinations of these toxin and phytohormones for 20 hours. For each treatment 100 seeds were taken in triplicate. The steeped seeds were subsequently placed on moist blotting paper and kept in germinator (B.O.D.) at  $28\pm 20^{\circ}\text{C}$ . The qualitative and quantitative analysis of amino acids will be measured with the help of amino quant followed by thin layer chromatography( T.L.C. ) plates.

## **RESULTS AND DISCUSSION**

Aflatoxin B<sub>1</sub> and phytohormone GA<sub>3</sub> individually and in combination exhibited marked depletion in amino acid contents of germinating maize seeds. Inhibition by AFT-B<sub>1</sub> was however, more pronounced (Table 1).

Five amino acids i.e. L- Leucine, L- methionine, DL( +) Alanine, Glutamine acid and Aspartic acid were prominently detected in the control and treated maize seeds. Amino acids were found to be decreased with the increase in the concentration of the toxin (AFT-B<sub>1</sub>). The maximum inhibition in L-Leucine and Glutamic acid contents was 79.24 and 77.78% respectively at 2000ppb concentrations of AFT-B<sub>1</sub>; L- Methionine, DL (+) Alanine and Aspartic acid were completely inhibited at that concentrations of AFT-B<sub>1</sub>. Minimum inhibition 22.64, 20.00, 30.64, 43.13 and 14.28 % was recorded in L- Leucine, L- Methionine, DL (+) Alanine, Glutamic acid and Aspartic acid due to 100µg/l concentration of AFT-B<sub>1</sub> respectively ( Table -1). Change in amino acid contents due to treatments of various concentration of GA<sub>3</sub> were also evident in (Table -2). The combined effect of AFT-B<sub>1</sub> and GA<sub>3</sub> 2000ppb concentration in various ratios on amino acid contents was depicted in (Table- 3). The maximum inhibition L- Leucine and Glutamic acid 79.24 and 77.12% at 3:1 ratio of these toxin AFT-B<sub>1</sub> and GA<sub>3</sub>, respectively. More or less complete inhibition in L – Methionine, DL(+) Alanine and Aspartic acid were recorded at various ratios of these combinations. The minimum inhibition were recorded at 1:3 ratio in L- Leucine *i.e.* 23.58%.

A highly significant fall in the levels of amino acids in maize seedlings was recorded with the treatment of different levels of AFT-B<sub>1</sub> and phytohormone individually as well as various combinations ( Table-1 and 3). The maximum inhibition in L-Leucine & Glutamic acid were upto 79.24 and 77.78 % at 2000ppb of AFT-B<sub>1</sub> respectively. However, L- Methionine, DL (+) Alanine and Aspartic acid were completely lost at that concentration. Different combinations of these toxin and phytohormone also reduced/ increase the levels of amino acid was 79.24 and 77.12 % at 3:1 ratio of these combination, respectively. More or less complete inhibition in L-methionine, DL(+) Alanine and Aspartic acid was recorded at other ratio of these combinations.

Some of the amino acids disappeared completely or expressed a sharp fall in quantity at high concentrations of AFT-B<sub>1</sub> treatment. This may probably due to partial utilizations of these amino acid in the germination processes. This assumption can further strength on the basis of RQ values of the treated seeds in my PhD work (Prasad 1995). Which were less than 1 in all the cases, inhibition in Protein and amino acid levels suggested that these components are finally converted in to Keto acids which participated in the respiratory cycle. Annapurna and Rao (1983) however, reported that the level of amino acid continue to decrease as the concentration of the test chemical (Meta seed 50) increased.

**Table 1**  
**Impact of Aflatoxin B<sub>1</sub> on Amino Acid Contents of Germinating Maize seeds (CV. Madhuri- 01)**

Conc. Of AFT-B <sub>1</sub> (µg/l) (ppb)	*L-Leucine#		*L-Methionine#		DL(+) Alanine#		Glutamic acid		Aspartic Acid#	
	Amount (S.E.)	Difference With control(% inhibition)	Amount (S.E.)	Difference With control(% inhibition)	Amount (S.E.)	Difference With control(% inhibition)	Amount (S.E.)	Difference With control(% inhibition)	Amount (S.E.)	Difference With control(% inhibition)
00	1.06 (0.05)	-	0.15 (0.03)	-	0.62 (0.03)	-	1.53 (0.07)	-	0.49 (0.04)	-
100	0.82 (0.03)	0.24 (22.64)	0.12 (0.02)	0.03 (20.00)	0.43 (0.03)	0.19 (30.64)	0.87 (0.04)	0.66 (43.13)	0.42 (0.04)	0.07 (14.28)
250	0.58 (0.04)	0.48 (45.28)	0.08 (0.02)	0.07 (46.67)	0.31 (0.03)	0.31 (50.00)	0.61 (0.03)	0.92 (60.13)	0.36 (0.03)	0.13 (26.53)
500	0.43 (0.03)	0.63 (59.43)	0.08 (0.01)	0.07 (46.67)	0.14 (0.02)	0.48 (77.42)	0.43 (0.03)	1.10 (71.89)	0.22 (0.02)	0.27 (55.10)
1000	0.25 (0.03)	0.81 (76.41)	0.05 (0.01)	0.10 (66.67)	-	-	0.38 (0.03)	1.15 (75.16)	0.09 (0.01)	0.04 (81.63)
2000	0.22 (0.01)	0.88 (79.24)	-	-	-	-	0.34 (0.03)	1.19 (77.78)	-	-

\*Essential amino acids

#( mg/100mg.)

**Table 2**  
**Impact of GA<sub>3</sub> on Amino Acid Contents of Germinating Maize seeds (CV. Madhuri- 01)**

Conc. Of GA <sub>3</sub> (µg/l) (ppb)	*L-Leucine#		*L-Methionine#		*DL(+) Alanine#		Glutamic acid		Aspartic Acid#	
	Amount (S.E.)	Difference With control(% inhibition)	Amount (S.E.)	Difference With control(% inhibition)	Amount (S.E.)	Difference With control(% inhibition)	Amount (S.E.)	Difference With control(% inhibition)	Amount (S.E.)	Difference With control(% inhibition)
00	1.06 (0.07)	-	0.10 (0.23)	-	0.61 (0.04)	-	1.52 (0.12)	-	0.48 (0.12)	-
100	1.05 (0.04)	0.09 (8.49)	0.14 (0.08)	0.03 (20.00)	0.63 (0.08)	0.09 (14.52)	1.06 (0.12)	0.47 (30.71)	0.45 (0.12)	0.06 (12.24)
250	1.05 (0.04)	0.43 (40.56)	0.13 (0.08)	0.03 (20.00)	0.66 (0.16)	0.18 (29.03)	0.86 (0.08)	0.65 (42.48)	0.40 (0.14)	0.11 (22.45)
500	1.06 (0.04)	0.49 (46.22)	0.10 (0.23)	0.05 (33.34)	0.62 (0.12)	0.24 (38.71)	0.72 (0.10)	0.78 (50.98)	0.24 (0.19)	0.29 (59.18)
1000	1.07 (0.08)	0.65 (61.32)	0.12 (0.12)	0.07 (46.67)	0.62 (0.04)	0.45 (72.58)	0.61 (0.25)	0.89 (58.17)	0.15 (0.21)	0.38 (77.55)
2000	1.05 (0.08)	0.72 (67.92)	0.12 (0.12)	-	0.60 (0.27)	0.59 (95.16)	0.31 (0.12)	1.15 (75.16)	-	-

\*Essential amino acids

#( mg/100mg.)

**Table 3**  
**Combined effect of AFT-B<sub>1</sub> & GA<sub>3</sub> on Amino Acid Contents of Germinating Maize seeds (CV. Madhuri- 01)**

Conc. Of 2000ppb (AFTB <sub>1</sub> : GA <sub>3</sub> )(V/V)	*L-Leucine#		*L-Methionine#		*DL(+) Alanine#		Glutamic acid#		Aspartic Acid#	
	Amount (S.E.)	Difference With control (% inhibition)	Amount (S.E.)	Difference With control (% inhibition)	Amount (S.E.)	Difference With control (% inhibition)	Amount (S.E.)	Difference With control (% inhibition)	Amount (S.E.)	Difference With control (% inhibition)
0:0	1.06 (0.12)	-	0.12 (0.12)	-	0.60 (0.09)	-	1.52 (0.12)	-	0.49 (0.04)	-
1:1	0.25 (0.10)	0.48 (73.58)	0.03 (0.12)	-	0.35 (0.04)	-	1.00 (0.02)	1.14 (74.50)	-	-
1:2	0.34 (0.12)	0.65 (65.69)	0.05 (0.14)	-	0.42 (0.09)	0.50 (80.64)	1.29 (0.16)	1.08 (70.58)	0.10 (0.09)	-
2:1	0.22 (0.12)	0.30 (76.41)	-	-	-	-	0.22 (0.12)	1.08 (76.47)	-	-
1:3	0.81 (0.10)	0.78 (23.58)	0.07 (0.08)	0.10 (66.67)	0.57 (0.12)	0.47 (75.80)	1.46 (0.24)	0.64 (41.83)	0.37 (0.12)	0.39 (79.59)
3:1	0.19 (0.16)	0.25 (79.24)	-	-	-	-	0.34 (0.16)	1.18 (77.12)	-	-

\*Essential amino acids

#( mg/100mg.)

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