Effects of Plant Bioregulators on Nutrients, Insect Resistance, and Yield of Corn (*Zea mays* L.)

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A field test was conducted at Mississippi State, MS, in 1986 to evaluate 15 natural or synthetic plant growth regulators (plant bioregulators) applied two times at two levels to corn, *Zea mays* (L.), in the midwhorl (V_{8-10}) stage of growth. The bioregulators were evaluated for their effects on levels of nutrients (protein, fat, fiber, ash, free sugars, total fatty acids, and the ratio of linoleic to linolenic acid), on any induced plant resistance to the southwestern corn borer *Diatraea grandiosella* Dyar, and on yield. Three bioregulators, glyphosine, BAS-109 (*all-cis*-8-(4-chlorophenyl)-3,4,8-triazatetracyclo[4.3.1.$0^{2,5}$.$0^{7,9}$]dec-3-ene), and V-2307 (3-chlorobenzyl 3,6-dichloro-2-methoxybenzoate), decreased the yield of southwestern corn borer infested and uninfested plots, while none increased yields significantly. None of the bioregulators had an effect on insect resistance. One bioregulator, gibberellic acid (GA_3), caused a decrease in the protein content of whorls. Of the bioregulators 12 increased the whorl levels of crude fat, total fatty acids, crude fiber, and/or ash. The most interesting result was that four bioregulators, GA_3, indole-3-acetic acid (IAA), Burst (a commercial mixture of cytokinins), and DCPTA [2-(diethylamino)ethyl 3,4-dichlorophenyl ether], decreased the ratio of linoleic to linolenic acid (18:2/18:3), a presumptive desirable indicator of cold hardiness of the plant and improved corn oil (less saturated) for human consumption.

There is an extensive literature about the use of either natural or synthetic plant bioregulators on cereals including corn, *Zea mays* L. (Nickell, 1983; Jung, 1984). Natural bioregulators including notably the cytokinin zeatin [6-[(4-hydroxy-3-methylbut-2-enyl)amino]purine] have been found in corn. Campbell et al. (1984) applied a synthetic bioregulator, Dinoseb (2-*sec*-butyl-4,6-dinitrophenol), to corn plants, resulting in a significant reduction in the growth of the corn earworm. Dinoseb has also been found to increase corn yields. Other bioregulators used on corn and other cereal crops include several GA antagonists and tetcyclacis (Jung, 1984). Burst AgriTech Inc., Overland Park, KS, has recommended their product Burst, a mixture of zeatins (cytokinins), to retard multiple earing. Of related plants within Graminaceae, CCC [(2-chloroethyl)trimethylammonium chloride] has been found relatively effective as a stem length retardant in wheat (*Triticum spp.*) but less so in rice (*Oryza sativa* L.) (Jung, 1984). Glyphosine [*N*,*N*-bis(phosphonomethyl)glycine] has been used as a ripener and sugar enhancer in sugar cane *Saccharum officinarum* L. (Nickell, 1984). In 1986, a field test was conducted at this location to evaluate a number of plant bioregulators when applied to growing corn for their effects on levels of nutrients, on any induced plant resistance to the southwestern corn borer, *Diatraea grandiosella* (Dyar), and on grain yield. Two applications of 15 bioregulators were applied to growing corn in the midwhorl V_{8-10} (Ritchie and Hanway, 1982) stage at three rates (zero, low, high) in statistically designed field plots. Plots were infected with insects, plant tissues were harvested and analyzed, insect damage was scored, and the corn was harvested to determine grain yield. This report summarizes the results.

MATERIALS AND METHODS

Field Plots. The corn hybrid Pioneer Brand 3165, a commercial variety commonly grown in the midsouth, was planted on 1 April 1986 on a marietta sandy loam (fine-loamy, siliceous thermic Fluvaquentic Entrochrepts) soil at Mississippi State, MS. The corn was planted in four-row plots with a border row between plots. A split plot design was used with bioregulators as the main plot and concentration–insect infestations combinations as subplots. The test was replicated four times. Thus, each compound required 0.0158 ha at each level infested and noninfested. Two applications of 15 plant bioregulators (identified in a later section) were applied to corn at the midwhorl stage of growth at two rates (low and high plus a control; see Table I of Hedin et al. (1988) for exact rates) on 30 April and again on 14 May. On both days, the temperature was 20–27 °C under clear skies and minimal wind velocity.

Infestations with southwestern corn borer larvae were made on 21 and 22 May with an average of 30 neonate larvae/plant applied to the whorl (Davis and Williams, 1980). Half of the plots were infested. Leaf feeding ratings were taken 14 days after infestation on a visual scale of 1–9 where 1 = highly resistant and 9 = highly susceptible (Davis et al., 1973).

Collections of plant samples for chemical analysis were made on 29 May. Ten whorl samples were made from the fourth row in each subplot that was uninfested. Samples were immediately frozen until analyzed. The corn was machine harvested on 30 September by a picker–sheller. The stated yields are those of shelled grain.

Chemical Analyses. AOAC methods (Horwitz, 1975) were used for the following analyses: total Solids (moisture), 14.083; crude fat, 14.019; crude fiber, 14.118; ash, 14.114; total protein, 2.049 (percent nitrogen × 6.25); nitrogen-free extract (NFE), by difference from 100%. Total sugars were determined by the colorimetric anthrone procedure (DuBois et al., 1956). Fatty acids were analyzed after methylation by GLC on a 0.2 × 182 cm column packed with 10% Silar-10 C on 100–120-mesh Gas Chrom Q operated at 180 °C in a N_2 flow of 20 mL/min. All results are reported on a dry-weight basis.

Procurement of Bioregulators. The trivial names, nomenclature, structures, sources of procurement, recommended treatments, and rates of application are given in Table I and in Figure 1 of our previous report (Hedin et al., 1988) except that compound **16**, Treflan, was not used. Each compound was weighed and dissolved in 5–10 mL of specified solvent (see Hedin et al. (1988), Table I). Portions of 1 mL each of the emulsifiers Span 80 and

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Table I. Proximate Analyses[a] of Fatty Acids with Ratio of Linoleic to Linolenic Acid, Sugars, and SWCB Leaf Feeding Ratings of Bioregulator-Treated Corn Whorls[b] and Seed Yield

			anal., %								leaf	yield,[g] kg/ha	
no.	sample[c]	rate[d]	protein	fat	fiber	ash	NFE	sugar[e]	fatty acid	18:2/18:3	feeding[f] score	uninfested	infested
1	CCC	0	13.8	1.1	18.8	6.5	59.8	3.0	0.12	0.87	6.01	8025	5051
		L	14.1	1.1	23.9	6.8	54.1	3.0	0.42	1.83	5.94	6563	5409
		H	13.8	2.4	24.0	7.2	52.6	3.1	0.25	1.00	6.14	7585	4524
2	BAS-105	0	13.9	1.2	22.0	6.5	56.6	3.1	0.19	1.54	6.10	7393	5573
		L	13.3	1.4	16.5	7.8	61.0	3.1	0.13	2.20	6.19	7283	5073
		H	13.8	2.2	21.2	8.8	54.0	3.1	0.16	1.42	6.17	7958	5057
3	BAS-109	0	13.8	1.1	21.8	6.6	56.7	3.1	0.16	1.40	6.19	8217	5227
		L	14.8	1.2	15.6	7.6	60.8	3.0	0.31	1.56	6.17	6392 (S)	5409
		H	12.8	1.8	22.8	6.2	56.6	2.8	0.17	1.18	6.05	6612	5156
4	PIX	0	14.4	1.5	24.2	6.8	53.1	3.1	0.14	1.18	6.00	6939	4963
		L	13.1	1.2	27.4	8.4	48.9	3.0	0.30	1.63	5.97	6530	5128
		H	13.4	2.2	26.6	6.5	51.6	3.0	0.34	1.11	6.15	7327	5128
5	Dinoseb	0	14.4	0.8	25.4	5.8	53.6	3.1	0.12	1.24	6.22	7056	5332
		L	13.1	0.8	27.8	7.6	50.7	3.1	0.37	1.23	6.32	6464	5266
		H	13.4	2.4	27.0	7.0	50.2	3.1	0.18	1.52	6.05	7563	4694
6	Burst	0	13.4	1.0	21.2	6.1	58.3	3.0	0.10	1.19	6.06	8047	5227
		L	14.1	1.1	25.6	7.0	52.2	2.8	0.22	0.97	6.30	6601	5661
		H	14.4	2.0	18.2	7.8	57.6	2.9	0.11	0.88	6.12	7837	4941
7	XE-1019	0	13.8	1.1	22.8	6.4	55.9	3.2	0.12	1.04	6.12	7629	5464
		L	13.9	1.1	27.6	7.4	50.0	3.0	0.24	0.91	6.19	7937	5601
		H	13.6	1.4	21.6	9.2	54.2	3.0	0.14	1.19	6.16	6953	5568
8	V-2307	0	13.9	1.3	23.3	6.3	55.2	3.0	0.18	1.47	6.21	7217	6310
		L	15.0	1.2	21.4	7.4	55.0	2.9	0.15	1.51	6.17	7354	5738
		H	13.4	1.6	25.6	8.0	51.4	3.0	0.13	1.12	6.12	6838	4040 (S)
9	DCPTA	0	13.8	1.4	23.3	6.5	55.0	3.1	0.12	1.47	6.11	8041	5304
		L	14.4	1.3	20.8	8.4	55.1	3.0	0.18	1.28	5.89	6508	4667
		H	13.1	2.0	21.4	8.4	55.1	3.0	0.11	1.00	6.20	8074	5161
10	Glyphosine	0	15.0	1.2	21.6	6.3	55.9	3.1	0.10	0.86	6.07	8673	6217
		L	14.1	1.6	23.4	6.8	54.1	3.0	0.17	1.42	6.09	6513 (S)	5101
		H	13.9	2.2	24.2	9.0	50.7	3.0	0.11	1.18	6.08	6315 (S)	3837 (S)
11	V-3183	0	13.0	1.1	18.5	6.1	61.3	3.2	0.17	1.28	6.04	7360	4177
		L	14.2	2.0	21.6	6.0	56.2	3.0	0.13	1.30	6.02	7701	5502
		H	13.8	1.8	27.8	7.8	48.8	3.0	0.12	1.13	6.14	6063	4974
12	GA_3	0	14.7	1.2	22.2	6.7	55.2	3.1	0.13	1.38	5.95	6695	5216
		L	13.6	1.8	23.4	6.0	55.2	3.0	0.15	1.09	5.91	6750	5117
		H	12.8	1.4	23.4	8.4	54.0	3.0	0.08	0.58	5.95	7085	5546
13	IAA	0	14.7	1.2	19.2	8.2	56.7	3.2	0.12	1.05	6.15	7228	4452
		L	14.4	1.4	23.6	5.2	55.4	3.1	0.16	1.10	6.14	6464	5321
		H	13.6	1.6	24.2	7.6	53.0	2.9	0.08	0.42	6.08	6024	5749
14	Kinetin	0	14.7	1.2	22.6	8.0	53.5	3.1	0.17	1.09	6.27	7365	5189
		L	14.1	1.8	18.8	6.6	58.7	3.0	0.13	1.28	6.28	7481	5628
		H	14.1	1.6	25.4	9.0	49.9	3.0	0.10	1.33	6.19	6805	4853
15	Arabinogalactan	0	14.4	1.3	21.3	7.6	55.4	3.1	0.16	1.03	6.05	6310	4804
		L	14.1	2.0	17.2	7.6	59.1	3.0	0.09	0.99	6.11	7283	4974
		H	12.8	1.8	24.6	8.2	52.6	2.9	0.07	0.88	6.16	6673	5310
	av of control & SEM	0	14.1 ± 0.2	1.2 ± 0.2	21.9 ± 0.9	6.7 ± 0.4	56.1 ± 0.6	3.1 ± 0.1	0.12 ± 0.1	1.07 ± 0.06	6.11	7480	5234

[a] Proximate analyses of 10 freeze-dried, ground, composited whorls performed in duplicate. All other analyses based on four replicates. [b] Bioregulators applied 4/30 and 5/14, corn infested 5/21 and 5/22, whorls gathered 5/29, leaf feeding scores assigned on 6/4 and 6/5. [c] See Table I and Figure 1 of Hedin et al. (1988) for nomenclature and structures. [d] Key: 0 = control, L = low, H = high. See Table I for rates of application. [e] Sugars increased as day progressed: rep 4, 0900 h, 2.6%; rep 3, 1030, 2.7%; rep 2, 1330, 3.5%; rep 1, 1500, 3.6%. Table gives averages of four replicates. [f] For comparisons among concentrations within individual bioregulators, LSD (0.05) = 0.25. [g] For comparisons among concentrations within individual bioregulators, LSD (0.05) = 1684, (S) = significant.

Tween 80 were added. The solution was made up to 2 L with water and stored at 4 °C until used.

Statistical Methods. Data obtained from the proximate analyses (see Chemical Analyses) were duplicates. Calculations of the control values were then carried out to obtain means and standard errors of the means. Analysis for fatty acids and sugars were based on four replicates. Means and standard errors of the means were calculated for controls only. Data obtained from the determination of yield and insect leaf feeding scores were subjected to ANOVA ($P = 0.05$), and LSD values were calculated.

RESULTS AND DISCUSSION

Corn whorls were analyzed for protein, crude fat, crude fiber, ash, nitrogen-free extract (by difference), fatty acids (with ratio of 18:2/18:3), and sugars. These data plus grain yields and insect leaf feeding scores are given in Table I. An abbreviated summary of the data in Table I identifying probable trends is given in Table II.

Only three of the bioregulators had a significant effect on yield, all adverse. Glyphosine decreased the yield in both southwestern corn borer (SWCB) infested and uninfested plots. BAS-109 at the low rate decreased the yield of uninfested plots, while the high rate of V-2307 decreased the yield of infested plots. Though not significant, there was an upward yield trend of IAA-treated, insect-infested plots. None of the bioregulators had an effect on SWCB resistance (Table I).

The bioregulators did not appear to affect protein levels, although there was a slight downward trend with GA. On

Table II. Abbreviated Summary of the Data in Table I Identifying Bioregulators Having a Probable Effect on Nutrients and Yields

compd	protein	fat	fiber	ash	fatty acid	18:2/18:3	sugar	yield uninfested	yield infested
CCC		↑	↑	↑	↑				
BAS-105		↑		↑		↑?			
BAS-109		↑	↑		↑		↓	↓(S)	
PIX		↑	↑		↑	↑?			
Dinoseb		↑			↑	↑			
Burst		↑		↑		↓			
XE-1019				↑					
V-2307				↑					↓(S)
DCPTA		↑		↑		↓			
Glyphosine		↑	↑	↑				↓(S)	↓(S)
V-3183		↑	↑	↑					
GA_3	↓				↓	↓			
IAA					↓	↓		↓	↑
Kinetin					↓	↑			
Arabinogalactan		↑			↓				

the other hand, we have observed increases of protein in leaves and squares (buds) of cotton plants treated with PIX (Hedin et al., 1984a) and with PIX, CCC, and V-3183 in a recent test (Hedin et al., 1988). Yokoyama (1984) reported an increase of protein in soybean plants treated with DCPTA. Rittig (1987) reported that bioregulators can increase protein in alfalfa.

Of the bioregulators 12 increased the fat, fiber, and/or ash content (Table II). The total fatty acid content was increased by four plant bioregulators and decreased by four others (Table II). BAS-105 has been reported to decrease lipoxygenase activity and to moderately decrease free sugars and oligosaccharides in peanuts (Ory et al., 1984). BAS-105 has also been reported to block desaturation of linoleic acid to linolenic acid in cotton seedlings and small grains, decreasing cold hardiness in the latter (Christiansen and St. John, 1984). In this test, BAS-105 increased 18:2/18:3 only at the low treatment level (Table I). PIX, Dinoseb, and kinetin increased in ratio. On the other hand, Burst, DCPTA, GA, and IAA decreased the ratio, presumably increasing cold hardiness. If the linolenic acid content of the kernel were also increased (ratio of 18:2/18:3 decreased), this could result in a less saturated corn oil for human consumption.

The free sugar content decreased only when BAS-109 was applied (Table I). It was interesting to note that whorl sugars, though not affected by bioregulators, increased as could be expected as the day progressed (Table I, footnote *e*). Replicate 4, which was harvested at approximately 0900 h contained 2.6%, while replicate 1, which was harvested at 1500 h contained 3.6%, evidence of biosynthesis of sugars during the daylight hours.

It is difficult to speculate about why none of the bioregulators increased yield or insect resistance because they have shown some activity at similar rates when applied by spraying to other crops including cereal grains. Applications were made 4 and 2 weeks before insect infestation in an attempt to elicit any latent resistance. If additional bioregulators are to be investigated, they perhaps should be selected for their known capabilities to affect biosynthesis of hemicellulose, protein, sugars, and polyphenol oxidase, factors found to be different in susceptible and resistant lines (Hedin et al., 1984b). While several of the bioregulators appeared to effect some changes in nutrient levels, the decrease in the 18:2/18:3 ratio of corn treated with BURST, DCPTA, GA_3, and IAA was perhaps the most interesting change. This could impart into the corn plant increased cold hardiness and, if the change also occurred in the kernel, an improved corn oil (less saturated) for human consumption.

Registry No. 1, 999-81-5; **2**, 3707-98-0; **3**, 77788-93-3; **4**, 24307-26-4; **5**, 88-85-7; **6**, 114718-87-5; **7**, 83657-22-1; **8**, 101191-06-4; **9**, 65202-07-5; **10**, 2439-99-8; **11**, 68157-60-8; **12**, 77-06-5; **13**, 87-51-4; 14, 525-79-1; **15**, 9036-66-2; 18:2, 60-33-3; 18:3, 463-40-1.

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