The Effect of Gibberellic Acid and Gibberellin Inhibitors on Cassava

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ABSTRACT

The effect of gibberellic acid and gibberellin inhibitors (Alar and RSW-0411) on the physiology of greenhouse grown cassava (Manihot esculenta Crantz) was investigated. Foliar application of gibberellic acid and gibberellin inhibitors markedly affected shoot and tuber growth. Gibberellic acid applied to a dwarf cultivar (MSAF2) stimulated shoot growth. This resulted in a decrease in tuber dry mass. Following gibberellic acid treatment, abscisic acid and cytokinin activity in the tuber decreased. Application of gibberellin inhibitors to a tall cultivar (MSAF1) decreased elongation and shoot dry mass. This decreased shoot growth resulted in a significant increase of tuber dry mass.

Tuberization in plants is to a large extent determined by the activities of plant growth regulators. Cytokinins have been considered as being essential for tuber initiation (Tizio and Biain, 1973). This view is supported by the fact that application of kinetin to potato plants induces tuberization (Smith and Palmer, 1970; Mingo-Castel et al., 1976) and by the findings that the endogenous cytokinin activity in potato plants increased when the plants were placed under tuber inducing conditions (Sattelmacher and Marschner, 1978). Kannangara and Booth (1978) also found an increase in cytokinin in the roots of dahlia with tuberization. The level of cytokinin activity did, however, decrease after tuber initiation. Van Staden and Dimalla (1976) found higher activity of cytokinins in "little potatoes" (formed in storage on emerging stolons) than in the mother tubers. It was suggested that the cytokinins act as a storage metabolic sink in this systems.

While considerable attention has been given to the possible role of cytokinins in the tuberization process, less in known about the other hormones. Following tuber initiation the activity of abscisic acid in potato stolons and tubers increased (Krauss, 1978). In experiments where ABA was applied to the leaves, tuber growth was promoted (Abdullah and Ahmed, 1980). These results suggest that increased ABA levels enhances some aspects of the tuberization process.

There are reports that gibberellins influence tuberization by influencing shoot growth (Menzel, 1980; Kuman et al., 1981). Applying gibberellic acid to the tops stimulates leaf and stem growth, while indirectly reducing tuber growth. This suggests that high levels of gibberellins in leaves promote the use of newly formed carbohydrates for shoot expansion. This process can be reversed by gibberellin inhibitors. Cassava yield increases have been reported with CCC (Das Gupta, 1976) and ALAR (Muthukrishnan, 1976).

In this investigation the effect of two gibberellin inhibitors on the growth of a tall cultivar and the effect of gibberellic acid on a dwarf cultivar of cassava was studied. The effect of GA, on the endogenous cytokinin and abscisic acid levels was also recorded.

Materials and Methods

The experiments used local cassava (Manihot esculenta Crantz) cultivars MSAF1 (tall) and MSAF2 (dwarf). Stakes of 40 g were planted in 12 kg loamy topsoil. A complete nutrient solution was given every 2 weeks. After 3.5 months the nutrient solution was withheld from MSAF2 to limit shoot growth.

Gibberellic acid (500 ppm to saturation) was applied to MSAF2, 5 months after planting. Gibberellin inhibitors RSW-0411 (500 ppm to saturation) and Alar (2000 ppm to saturation) were applied to MSAF1, 3 months after planting. RSW-0411 was supplied by Bayer S.A. This regulator belongs to the azole group of chemicals and is at the last stages of development.

Shoot length, leaf area, number of leaves, fresh and dry mass of MSAF1 and MSAF2 were measured at harvest which occurred 86 days and 45 days after treatment, respectively. To determine their chlorophyll content, sections of young leaves were extracted in 100% methanol for 24 h and the absorbance measured at 435 um and 665 um.

Abscisic acid extraction and bioassay

Samples of fresh tubers were extracted in 80% methanol for 24 h at 5°C, dried down, purified with PVP followed by solvent extraction with ethyl acetate at pH 3.5. The water fraction was hydrolysed at pH 11 and solvent extracted at pH 3.5. All ethyl acetate fractions were pooled and after drying down separated on TLC plates using toluene: ethyl acetate: acetic acid (40:15:2 v/v). The chromatograms were divided into eight equal Rf strips and the inhibitor activity determined using the wheat embryo test (Eeuwens and Schwabe, 1975).

Cytokinin extraction and bioassay

Samples of fresh tubers were extracted in 80% ethanol for 24 h at 5°C, purified on a Dowex 50W-X8 column, separated on paper using iso-propanol: 25% ammonium hydroxide:water (10:1:1 v/v). The Rf zone 0.2 - 0.9 was eluted and separated on an LH-20 Sephadex column using 10% methanol as solvent. The fractions were assayed for cytokinin activity using the soybean callus bioassay (Miller, 1965).

Results and Discussion

The effect of treatments on growth and tuberization

Application of gibberellin inhibitors to MSAF1 caused a marked change in dry matter distribution (Table 1). The total dry mass was not affected by the treatments, but shoot mass was 35% and 16% lower than in the control for RSW-0411 and Alar, respectively. The carbohydrates were apparently diverted to the tubers. Shoot:root ratio decreased significantly. Although the leaf area was reduced by RSW-0411 application, the mass/cm^2 of the young leaves was significantly higher. Differences in chlorophyll absorbance were found, but were not significant.

RSW-0411 inhibited shoot elongation more than Alar (Figure 1). The rate of leaf appearance (Figure 2) was not affected by either of the inhibitors. Both regulators have a long residual effect.

Table 1. Dry mass, mass per cm^2, and chlorophyll content of leaves of cassava cultivar MSAF1 as affected by gibberellin inhibitors.

	Control	RSW-0411	Alar	LSD (0,05)
Dry shoot mass (g)	44.2	28.7	37.0	9.1
Dry mass tubers (g)	25.4	50.7	31.5	10.8
Shoot/root ration	1.77	0.57	1.44	0.81
Dry mass increase shoots after application (g)	19.8	8.0	13.8	6.2
Total leaf area (cm^2)	2,932	2,434	2,889	6.8
Leaf mass mg/cm^2 young leaves	12.8	15.9	13.6	1.8
% chlorophyll absorbance towards control at				
435 nm	-	+ 7.0%	+ 7.7%	n.s.
665 nm	-	+14.7%	+ 6.7%	n.s.

Shoots of MSAF2 plants had stopped growing after 5 months due to withholding of the nutrient solution. After application of gibberellic acid there was a rapid elongation of the shoots (Table 2). Tubers which apparently acted as sinks before the hormone application now became a source making carbohydrate available for the rapidly growing shoots. The shoot:root ratio changed significantly.

Table 2. Dry mass, cytokinin, and inhibitor levels of cassava cultivar MSAF2 as affected by gibberellic acid application.

	Treated	Untreated
Dry mass shoot (g)	42.2	32.3 n.s.
Dry mass tuber (g)	48.6	57.6**
Zeatin ng/g	3.5	33.7
Ribosylzeatin ng/g	6.9	273.7
Abscisic acid ng/g	1.5	15.8

The effect of gibberellic acid treatment on endogenous hormone levels

Gibberellic acid application to the shoot caused a decrease of both the inhibiter activity (Figure 3) and the cytokinin level (Figure 4). The inhibiter activity is found in the Rf zone that co-chromatographed with authentic abscisic acid. The inhibition around Rf 0.5 might be due to other inhibitors of the

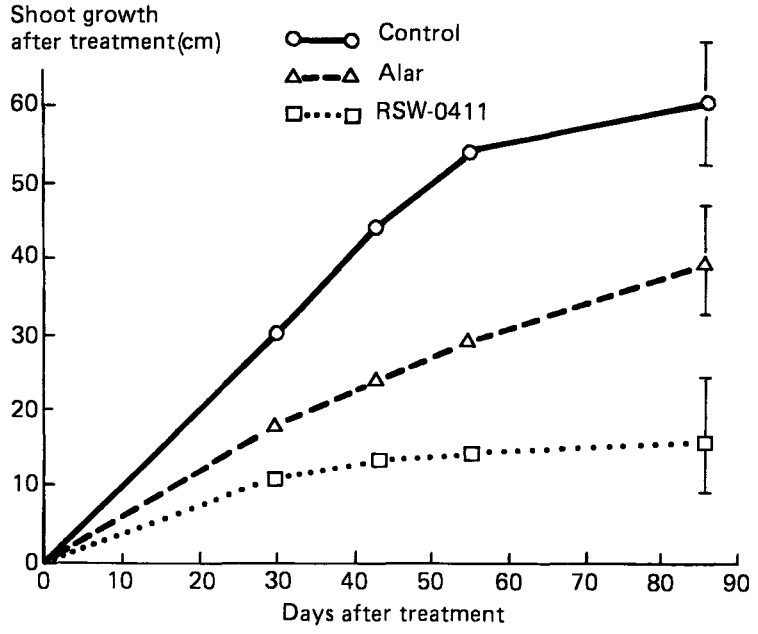

Figure 1. Effect of gibberellin inhibitors on shoot growth of cassava cultivar MSAF1.

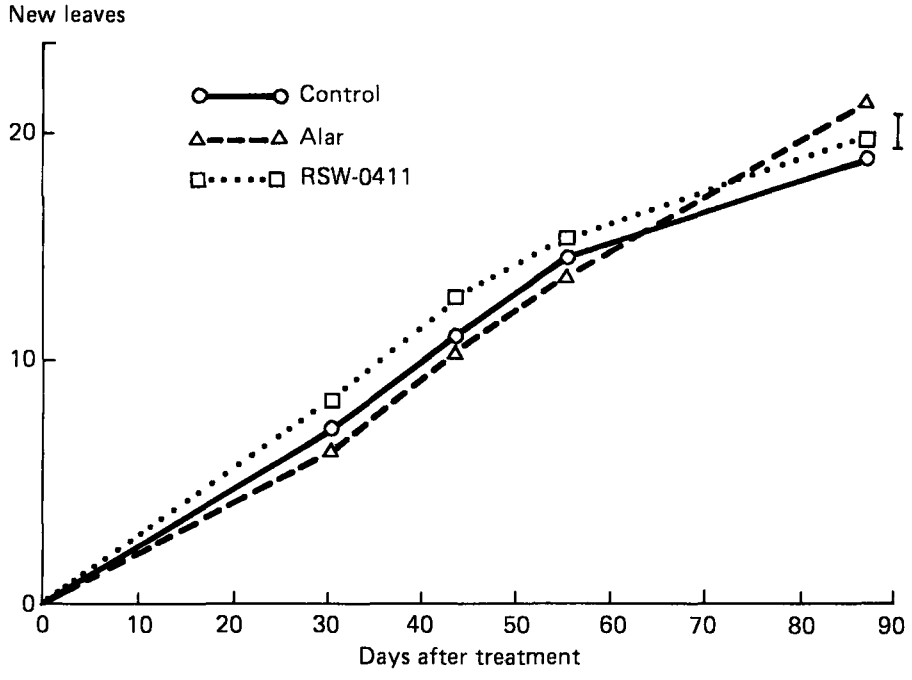

Figure 2. Effect of gibberellin inhibitors on the rate of leaf appearance of cassava cultivar MSAF1.

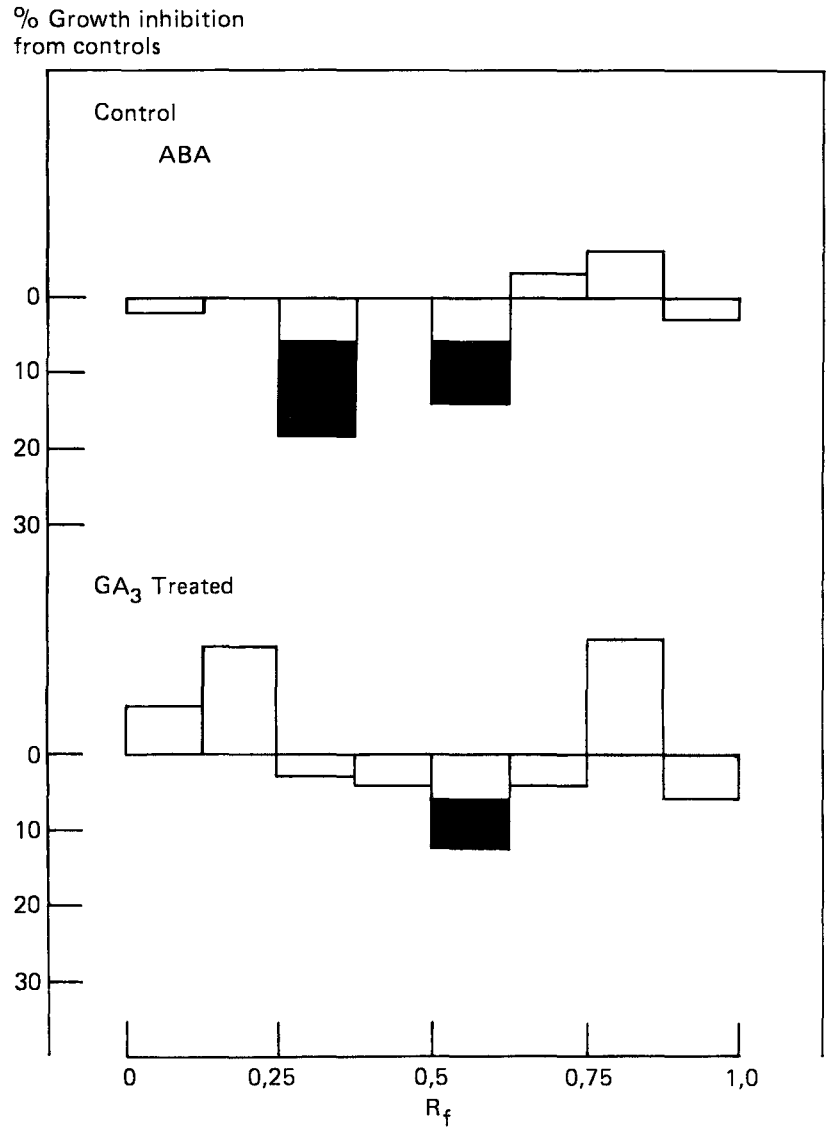

Figure 3. Wheat embryo bioassay for inhibitors extracted from 20 g fresh tubers of gibberellic acid treated and untreated plants. The shaded areas represent regions significantly different from the controls at the 1% level. ABA = abscisic acid.

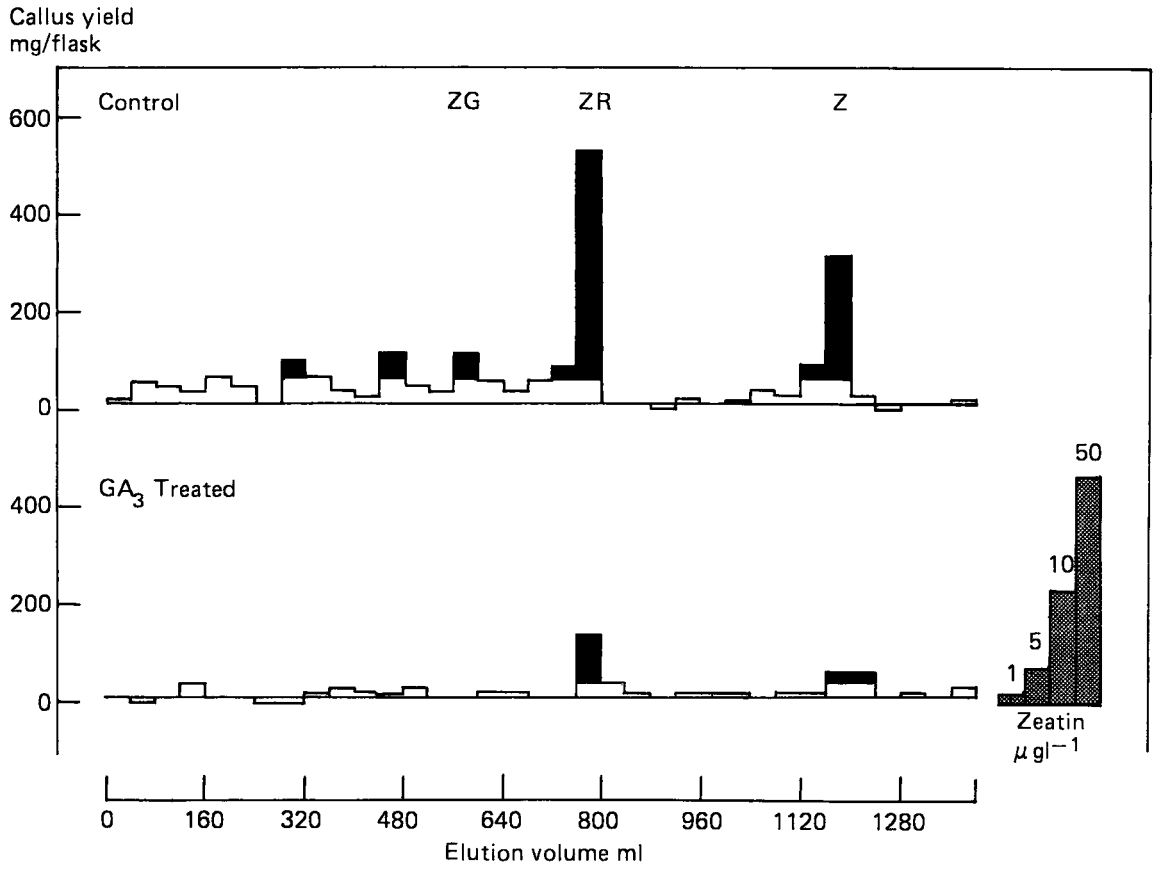

Figure 4. Soybean callus bioassay of extracts from 16 g cassava tubers which were obtained from gibberellic acid treated and untreated plants. After purification the extract was fractionated on a Sephadex LH-20 column eluted with 10% methanol. The shaded areas represent regions significantly different from the mean at the 5% levels. Z = zeatin, ZR = ribosylzeatin, ZG = glucozylzeatin.

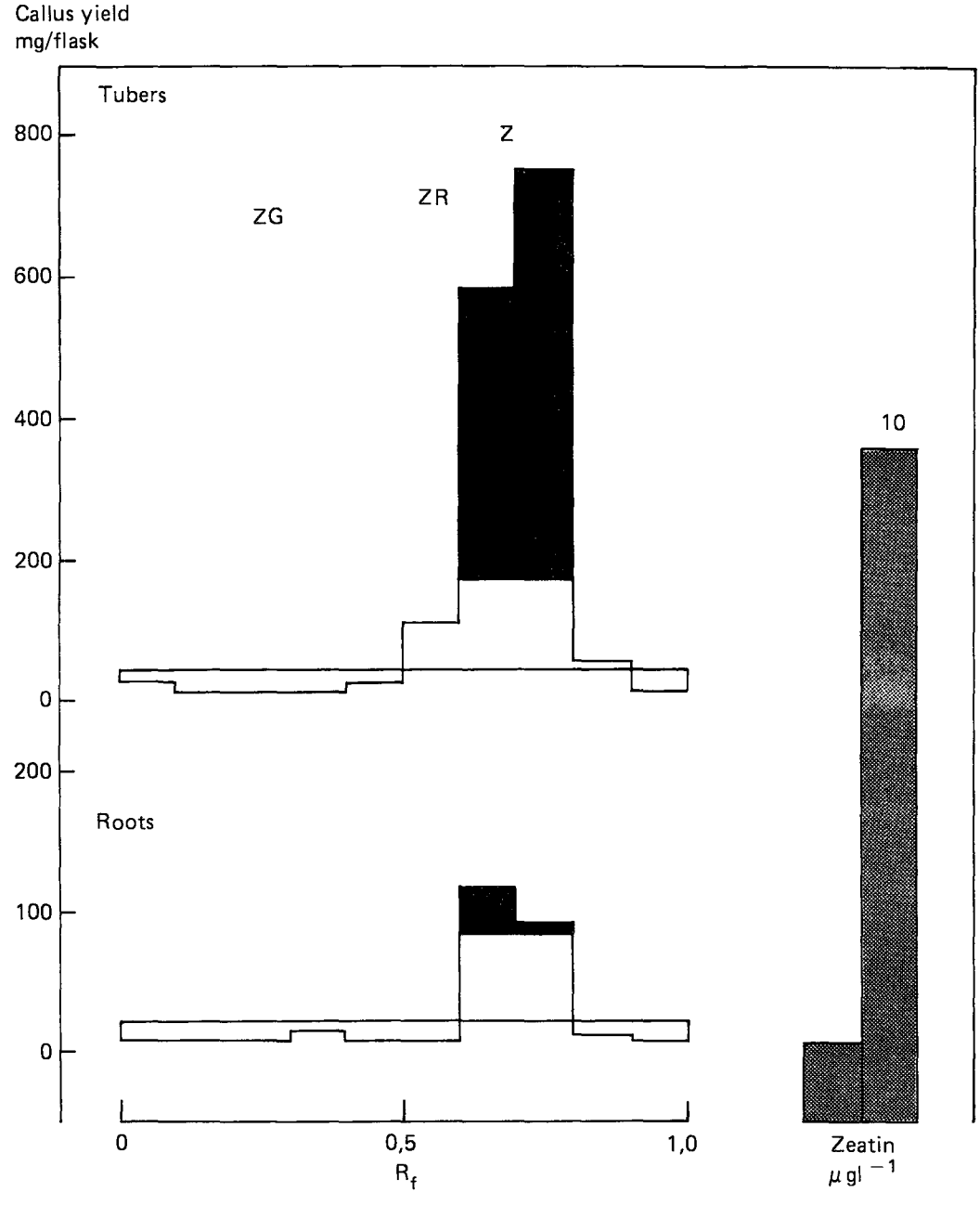

Figure 5. Soybean callus bioassay of extracts from 16 g fresh cassava tubers and roots. Chromatograms were developed in iso-propanol: 25% ammonium hydroxide:water (10:1:1:v/v). The shaded areas represent regions significantly different from the mean at the 5% levels. Z = zeatin, ZR = ribosylzeatin, ZG = glucosylzeatin.

β-inhibitor complex. High activity of both zeatin and ribosylzeatin and some activity of glucosylzeatin were found in expanding tubers.

High activity of inhibitors and cytokinins appears to be associated with sink activity. It is known that cytokinins are involved in cell division. In rapidly growing tubers many cell divisions take place. A comparison of cytokinin activity in roots and tubers of cassava shows much higher activity in the tubers (Figure 5).

Gibberellic acid and gibberellin inhibitors have a drastic influence on the physiology of cassava. Application of these regulators alters the dry matter distribution between the shoot and the tubers. In practice the gibberellin inhibitors can be used to limit excessive leaf growth and increase the tuber yield.

Plant hormones appear to play a key role in the source-sink relations in the plant. Gibberellic acid favors the sink activity of the shoots, while cytokinins and abscisic acid have been found to be associated with sink activity in the tubers.

Acknowledgements

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