

Promoting branching of a potential biofuel crop *Jatropha curcas* L. by foliar application of plant growth regulators

Hafiz A. Abdelgadir · Steven D. Johnson ·
Johannes Van Staden

Received: 20 October 2008 / Accepted: 31 March 2009 / Published online: 16 April 2009
© Springer Science+Business Media B.V. 2009

Abstract *Jatropha curcas* L. (Euphorbiaceae) has the potential to become a key biofuel crop. Manual pruning (MP) is one of the major management practices in commercial plantations of this crop, resulting in production of more branches and thus increased potential for more inflorescences leading to a higher seed yield. However, this method is time-consuming, labour-intensive and expensive. This study was conducted to determine the potential of different plant growth regulators (PGRs) to increase the number of lateral branches of *J. curcas*. A single foliar application of *N*⁶-benzyladenine (BA) at 12 mM significantly increased branches in both the pot (4.0) and field (13.2) trials compared to MP (1.8 and 5.7, respectively) and control (no new branches) plants. In the field, a single foliar application of 1.0 mM 2,3,5-triiodobenzoic acid (TIBA) resulted in a significant increment in the number of branches (15.9) after 7 months. Of all the PGRs examined, 2,3:4,6-di-*O*-isopropylidene-2-keto-L-gluconic acid (dikegulac; DK) at 2.0 mM produced the maximum number of branches (18.0) in the field 7 months after application. Concentrations of 2.0 and 3.0 mM of 1,2-dihydro-3,6-pyridazinedione (maleic hydrazide; MH) significantly increased the number of branches, 4 and 7 months after spraying in both the pot trial in the shade house and field,

respectively. Under field conditions, *J. curcas* plants responded better to all the PGRs (DK < TIBA < BA < MH) when treated once, with insignificant variations in other growth parameters. This study indicates that a single foliar application of PGRs under field conditions can be an alternative method to MP for increasing the number of lateral branches of *J. curcas*.

Keywords Chemical pruning · Dikegulac · *Jatropha* · Lateral branching · Maleic hydrazide · Manual pruning · TIBA

Abbreviations

| | |
|------|--|
| BA | <i>N</i> ⁶ -benzyladenine |
| Ck | Cytokinin |
| DK | Dikegulac (2,3:4,6-di- <i>O</i> -isopropylidene-2-keto-L-gulonic acid) |
| MH | Maleic hydrazide (1,2-dihydro-3,6-pyridazinedione, coline salt) |
| MP | Manual pruning |
| PGR | Plant growth regulator |
| TIBA | 2,3,5-triiodobenzoic acid |

Introduction

Declining availability of fossil fuels is driving the current search for alternative sources of energy. Biofuels offer promise, but are controversial because of their requirement of large land area for production, potential competition with food production and marginal economic viability in the absence of subsidies (Gressel 2008). These potential negative impacts could be reduced and profitability increased if production could be made more efficient.

H. A. Abdelgadir · J. Van Staden (✉)
Research Centre for Plant Growth and Development,
School of Biological and Conservation Sciences, University of
KwaZulu-Natal Pietermaritzburg, Private Bag X01,
Scottsville 3209, South Africa
e-mail: rcpgd@ukzn.ac.za

S. D. Johnson
School of Biological and Conservation Sciences,
University of KwaZulu-Natal Pietermaritzburg,
Private Bag X01, Scottsville 3209, South Africa

A crop with a good potential for biofuel production in arid and semi-arid regions is the physic nut, *Jatropha curcas* L. (Heller 1996; Augustus et al. 2002; Azam et al. 2005; Achten et al. 2008). It was suggested that the oil yield from *J. curcas* nuts can be improved if the number of seed-bearing branches could be increased. The pruning of apical buds of the main stem of one-year-old plants can increase the number of main and secondary branches (Kureel 2006). Proper pruning of *J. curcas* helps in producing more branches with healthy inflorescences. This enhances flowering and fruit set that ultimately increases yield (Gour 2006). However, the cost, convenience and efficiency of manual pruning in large-scale plantations still remains a major concern.

Plant growth regulators (PGRs) have been an important component in agricultural production. They are used on millions of hectares worldwide on a diversity of crops. Most of these applications are, however, confined to high-value horticultural crops rather than field crops, although there are significant exceptions. Significant opportunities exist for the development of PGRs to increase yield in the major crops. PGRs are useful because they can in some way modify plant development. This may occur by interfering with biosynthesis, metabolism, or translocation of plant hormones, or the PGRs may replace or supplement the plant hormones when their endogenous levels are below that needed to change the course of plant development (Gianfagna 1995). A number of PGRs can serve as powerful tools for manipulating tree growth and yield (Lovat 2006). In contrast to the inhibitory action of auxin, cytokinin (Ck) can directly promote bud growth. Exogenous application of Ck to buds promotes their outgrowth (Miguel et al. 1998; Chatfield et al. 2000), and Ck levels increase in buds as they activate (Emery et al. 1998). The Ck, BA (N^6 -benzyladenine) is known to release apical dominance and promote new lateral branches (Richards and Wilkinson 1984; Svenson 1991) by altering the auxin to Ck ratio in shoot tips (Cline 1991). The auxin transport inhibitor TIBA (2,3,5-triiodobenzoic acid) promotes the activation of auxiliary buds under a wide range of experimental and field conditions (Morey and Dahl 1975; Bangerth 1993). Dikegulac (DK; 2,3:4,6-di-*O*-isopropylidene-2-keto-L-gulonic acid) is used to overcome apical dominance and increase axillary shoot production (Banko and Stefani 1996). Maleic hydrazide (MH; 1,2-dihydro-3,6-pyridazinedione, coline salt) acts as an anti-auxin or a regulator of auxin metabolism (Hoffman and Parups 1964), suggesting the possibility that it may increase Ck levels in lateral buds and stimulate shoot elongation (Ito et al. 2000). This study was undertaken to evaluate the effect of these proven, widely used, PGRs on branching of *J. curcas* plants as a possible substitution to manual pruning.

Materials and methods

Study sites and plant materials

Field: The experiments were conducted in a monoculture plantation at the University of KwaZulu-Natal Agricultural Research Station (Ukulinga) Pietermaritzburg, South Africa (30°41'E, 29°67'S and 781 m a. s. l). The plantation was established from seeds. The seeds were obtained from collections made by the Department of Agriculture at the Owen Sithole College of Agriculture, South Africa. The original seeds came from Malawi (Everson and Everson 2006). The average lowest and highest temperature at the time when the experiments were conducted was 15 and 27°C, respectively.

Shade house: Seeds were collected from the study site mentioned above and sown (3 seeds each) in plastic pots (size 20 cm). The soil mixture (v/v) in each pot was 80% compost: 20% bark (chipped and decomposed): 0.5% LAN (limestone ammonium nitrate): 0.5% NPK (nitrogen, phosphorus, potassium). The pots were placed in a shade house at the University of KwaZulu-Natal Botanical Garden, Pietermaritzburg, South Africa. The mean temperature and solar radiation at mid-day in the shade house were $28^\circ\text{C} \pm 3$ and $331 \pm 16 \mu\text{mol m}^{-2} \text{s}^{-1}$, respectively. The plants were watered every 14 days and weeded once a month.

Shade house experiment

Five months after sowing, when the plants were at similar height and stem diameter they were treated with PGRs. The foliar PGR-treatments consisted of BA (3.0, 6.0, 9.0 and 12.0 mM), TIBA (0.5, 1.0, 1.5 and 2.0 mM), DK (2.0, 4.0, 6.0 and 8.0 mM) and MH (2.0, 3.0 and 4.0 mM). A small volume (3.0 ml) of sodium hydroxide (0.1 M) was used to solubilize PGRs before adding water. Plants sprayed with distilled water plus an equivalent amount of 0.1 M NaOH (Merck, Germany) served as control. A few drops (2.0 ml) of Tween[®] 20 (Merck, Germany) were added as surfactant. Plant growth regulators BA, TIBA and DK were purchased from Sigma-Aldrich Ltd, South Africa, and MH was obtained from Koch-Light Laboratories Ltd, UK. The plants were treated once on 20 October 2007 (foliar application of 50 ml of test solution per seedling) using a new plastic sprayer (500 ml) for each PGR. Manual pruning was done on the same day as that of the foliar treatments. Each treatment consisted of sixteen plants considering a single plant as one replicate arranged randomly. The distance between any two pots was 45 cm. Plant height, shoot length, number of lateral branches and leaves were recorded before and after 1 and 4 months' PGR

treatments. Growth of a lateral branch was considered when it had elongated more than 3 cm.

Field experiment

The experiment was conducted at the study site described above. The foliar PGR-treatments consisted of BA (3.0, 6.0, 9.0 and 12.0 mM), TIBA (0.5, 1.0, 1.5 and 2.0 mM), DK (2.0, 4.0, 6.0 and 8.0 mM) and MH (2.0, 3.0 and 4.0 mM). Sodium hydroxide (0.1 M) and Tween[®] 20 were used as mentioned above. The plants were treated once on 20 May 2007, and each plant received 200 ml of respective test solution using a new plastic sprayer (500 ml) for each PGR. The foliar spray was done in such a way that it covered the entire plant to contact leaves, stems and meristem. Manual pruning was done on the same day as that of the foliar treatment. Each treatment consisted of twelve plants considering a single plant as one replicate laid out randomly. The distance between the plants was 2.5 m. Plant height, plant crown diameter, number of lateral branches and stem diameter at the base were recorded before and after 3 and 7 months' PGR treatments.

Statistical analysis

Data were analyzed using SPSS[®] version 15 (SPSS Inc., Chicago, USA) statistical software. Effects of treatments on plant growth were analyzed using one-way analysis of variance (ANOVA). Tukey's Honestly Significant Difference (HSD) test was used to compare the significance of differences among treatments at $P \leq 0.05$.

Results

Shade house experiment

Foliar applications of BA had significantly increased the number of branches after 1 and 4 months in comparison to the control where no new branches developed (Fig. 1). The number of leaves produced by plants treated with BA at 15 mM was significantly higher than the control after 1 month (Table 1). Of all the concentrations of TIBA tested, only 1.5 mM led to a significant increase in the number of branches (1.6) compared to the control, with no branching after four months. On the other hand, manual pruning produced significantly more branches (1.8) than the control and other concentrations of TIBA applied (Fig. 1). After 1 and 4 months, TIBA application did not significantly improve plant height, shoot length and number of leaves compared to the control plants (Table 2). Spraying of plants with DK (6.0 mM) and MH (2.0 mM) resulted in a significantly higher number of branches (4.7

and 2.8, respectively) compared to control plants after 4 months (Fig. 1). High concentrations of DK (8.0 mM) and MH (4.0 mM) significantly decreased plant height, shoot length and number of leaves in comparison to the untreated plants after 4 months (Tables 3, 4 respectively). There were some growth abnormalities, such as short yellowish leaves and stunted plants.

Field experiment

One-year-old plants treated with BA at 12 mM produced a significantly higher number of branches after 3 and 7 months (5.5 and 13.2, respectively) than the untreated plants (1.2 and 3.8, respectively) (Fig. 1). Manual pruning did not show any significant increase in the number of branches compared to this BA concentration after 7 months (Fig. 1). Although some concentrations of BA tested increased plant height, crown- and stem diameter, these results were not significantly different from those obtained for the control (Table 1). Foliar application of TIBA at 1.0 and 2.0 mM produced a significantly higher number of branches (15.9 and 15, respectively) compared to the control (3.8) and manually pruned (5.7) plants after 7 months (Fig. 1). Plant height, crown- and stem diameter were not significantly higher than the control with the application of TIBA (Table 2). A concentration of 2.0 mM of DK produced more branches (18.1) compared to the control (3.8) and manual pruning (5.7) after 7 months (Fig. 1). MH at 3.0 mM also produced a significantly higher number of branches (11.7) compared to the control (3.8) and manual pruning (5.7) 7 months after foliar application. However, plant height, crown- and stem diameter did not significantly increase when compared to the control plants (Table 4).

Discussion

Shoot branching is the process by which buds, located in the axil of a leaf, develop and form new flowers or branches (Ongaro and Leyser 2008). Cytokinins and auxins are part of the plants' hormonal mechanism of enforcing apical dominance. Apical dominance, however, can be interrupted in several ways. One way is by reduction of the internal ratio of auxin to Ck through application of external Cks. A second way is by application of a chemical that inhibits auxin production or transport. A third way is by killing the apical meristem which halts auxin production (Bangerth 1993; Bangerth et al. 2000). Application of Cks interrupts the balance in favour of Cks, which result in lateral buds escaping apical dominance and breaking (Lalouem and Fox 1989). As growth inhibitors, we used the anti-auxins TIBA and maleic hydrazide. These substances work by halting

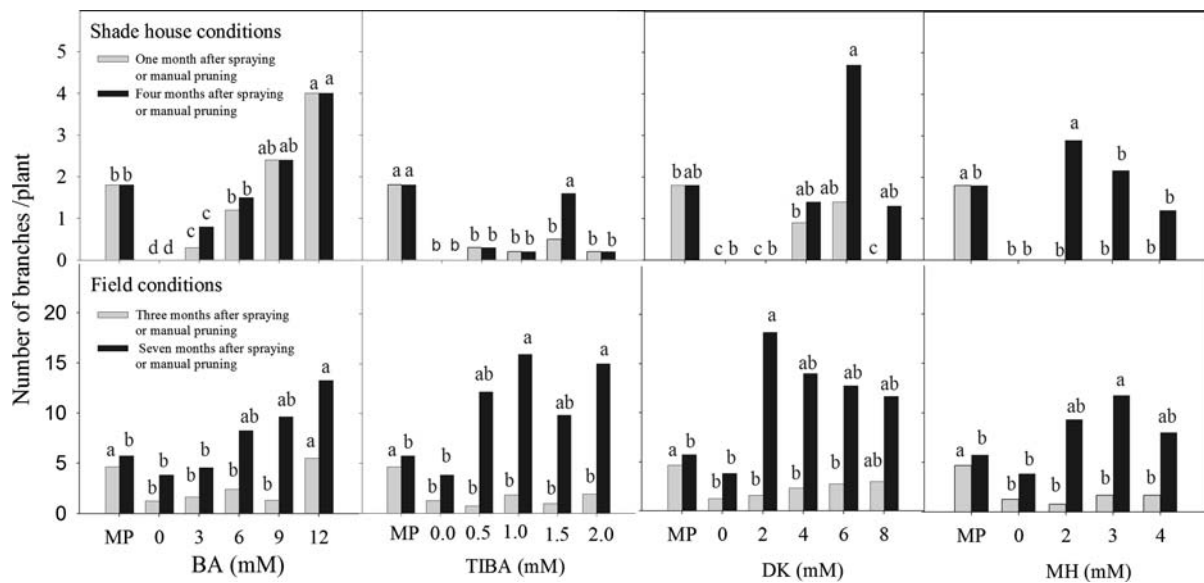


Fig. 1 Influence of foliar application of plant growth regulators on lateral branching of *Jatropha curcas* under shade house and field conditions. Means with common letters are not significantly different

at $P \leq 0.05$, according to Tukey's HSD test. BA *N*⁶-benzyladenine, DK dikegulac, MH maleic hydrazide, TIBA 2,3,5-triiodobenzoic acid

Table 1 Effects of a single foliar application of *N*⁶-benzyladenine (BA) and manual pruning (MP) on different growth parameters of *J. curcas*

| BA (mM) | Five-month-old plants (shade house conditions) | | | Twelve-month-old plants (field conditions) | | |
|---------|--|--------------------------------|---------------|--|---------------------|--------------------|
| | Plant height (cm) | Shoot length (cm) ^a | No. of leaves | Plant height (cm) | Crown diameter (cm) | Stem diameter (cm) |
| | One month after spraying | | | Three months after spraying | | |
| 0 | 19.2 ab | 2.9 ^b | 9.7 b | 5.5 ^b | 18.4 ab | 3.3 ^b |
| 3 | 25.0 a | 3.3 | 15.5 b | 7.0 | 14.3 ab | 1.9 |
| 6 | 9.5 ab | 1.7 | 12.9 b | 3.3 | 17.9 ab | 2.8 |
| 9 | 3.1 b | 0.7 | 10.6 b | 6.0 | 28.7 a | 3.5 |
| 12 | 3.4 b | 1.2 | 13.8 b | 9.2 | 16.6 ab | 3.0 |
| 15 | 3.5 b | 4.2 | 26.2 a | — | — | — |
| MP | 10.0 ab | 1.6 | 9.9 b | 4.0 | 5.1 b | 2.1 |
| | Four months after spraying | | | Seven months after spraying | | |
| 0 | 32.8 a | 24.4 a | 32.8 a | 54.5 a | 220.0 ^b | 2.6 ab |
| 3 | 34.5 a | 27.5 a | 18.8 b | 52.6 a | 231.0 | 1.9 ab |
| 6 | 16.7 b | 11.0 bc | 13.8 bc | 61.2 a | 241.0 | 2.3 ab |
| 9 | 10.2 b | 9.6 bc | 17.5 b | 63.4 a | 244.0 | 2.9 ab |
| 12 | 16.6 b | 12.6 bc | 13.8 b | 75.4 a | 235.0 | 2.9 a |
| 15 | 14.9 b | 20.1 ab | 17.4 b | — | — | — |
| MP | 3.7 b | 2.3 c | 3.7 c | 16.6 b | 142.0 | 1.6 b |

^a Measurement from the first leaf nodes

^b No significant difference at $P \leq 0.05$, according to *F* test

Means with common letters are not significantly different at $P \leq 0.05$, according to Tukey's HSD test

the biosynthesis of auxins or by interrupting its transport to the site of action, subsequently breaking apical dominance and stimulating branching. Dikegulac is known to reduce stem growth and break apical dominance (Kwon and Criley 1991). Foliar application of dikegulac to the shoot apices

inhibits DNA synthesis, thus restricting cell division and new growth (Arzee et al. 1977). This has the effect on preventing the apical meristem from growing and thus breaks apical dominance and stimulates lateral buds to grow. This results in denser and generally shorter plants. It

Table 2 Effects of a single foliar application of 2,3,5-triiodo benzoic acid (TIBA) and manual pruning (MP) on different growth parameters of *J. curcas*

| TIBA (mM) | Five-month-old plants (shade house conditions) | | | Twelve-month-old plants (field conditions) | | |
|-----------|--|--------------------------------|------------------|--|---------------------|--------------------|
| | Plant height (cm) | Shoot length (cm) ^a | No. of leaves | Plant height (cm) | Crown diameter (cm) | Stem diameter (cm) |
| | One month after spraying | | | Three months after spraying | | |
| 0 | 19.2 ^b | 2.9 ^b | 9.7 ^b | 5.5 ^b | 18.4 a | 3.3 ^b |
| 0.5 | 21.5 | 2.1 | 7.4 | 6.3 | 17.3 a | 3.5 |
| 1.0 | 32.7 | 2.5 | 5.9 | 9.0 | 26.2 a | 3.7 |
| 1.5 | 16.2 | 3.6 | 7.5 | 5.0 | 39.9 a | 3.4 |
| 2.0 | 15.8 | 3.5 | 7.2 | 6.5 | 24.5 a | 2.2 |
| MP | 10.0 | 1.6 | 9.9 | 4.0 | 5.1 b | 2.1 |
| | Four months after spraying | | | Seven months after spraying | | |
| 0 | 32.8 a | 24.4a | 32.8a | 54.5 ^b | 220 ^b | 2.6 ^b |
| 0.5 | 27.2 ab | 22.4ab | 22.4a | 39.4 | 228 | 1.6 |
| 1.0 | 15.8b | 10.3 9b | 8.7b | 52.0 | 236 | 1.7 |
| 1.5 | 16.4b | 15.6ab | 20.6a | 53.0 | 258 | 2.7 |
| 2.0 | 21.1ab | 22.2ab | 15.4b | 27.0 | 251 | 2.4 |
| MP | 3.7c | 2.3c | 3.7c | 16.6 | 142 | 1.6 |

^a Measurement from the first leaf nodes^b No significant difference at $P \leq 0.05$, according to F testMeans with common letters are not significantly different at $P \leq 0.05$, according to Tukey's HSD test**Table 3** Effects of a single foliar application of dikegulac (DK) and manual pruning (MP) on different growth parameters of *J. curcas*

| DK (mM) | Five-month-old plants (shade house conditions) | | | Five-month-old plants (shade house conditions) | | |
|---------|--|--------------------------------|---------------|--|--------------------------------|------------------------|
| | Plant height (cm) | Shoot length (cm) ^a | No. of leaves | Plant height (cm) | Shoot length (cm) ^a | Stem diameter (cm) |
| | One month after spraying | | | Three months after spraying | | |
| 0 | 19.2ab | 2.9a | 9.7a | 5.5 ^b | 18.4ab | 3.3 ± 1.0 ^b |
| 2 | 31.3a | 0.83b | 6.3ab | 6.2 | 18.0 ab | 2.1 ± 0.5 |
| 4 | 10.1b | 1.9a | 10.7a | 5.3 | 19.9a | 3.3.8 ± 0.8 |
| 6 | 10.9ab | 1.2ab | 8.4a | 2.58 | 7.53ab | 2.7 ± 0.7 |
| 8 | 3.4c | 0.66b | 4.8b | 2.75 | 6.96ab | 2.8 ± 0.6 |
| MP | 10.0b | 1.6ab | 9.9a | 4.0 | 5.1b | 2.1 ± 0.6 |
| | Four months after spraying | | | Seven months after spraying | | |
| 0 | 32.8a | 24.4a | 32.8a | 54.5 ab | 220ab | 2.6ab |
| 2 | 29.5ab | 22.8ab | 10.4c | 74.7a | 283a | 4.0a |
| 4 | 14.6b | 11.3b | 21.8b | 26.7b | 223ab | 2.4b |
| 6 | 18.5ab | 17.0ab | 18.0bc | 47.7ab | 245ab | 2.1b |
| 8 | 2.2c | 1.2c | 6.0 cd | 54.4ab | 239ab | 2.5ab |
| MP | 3.7c | 2.3c | 3.7d | 16.6b | 142b | 1.6b |

^a Measurement from the first leaf nodes^b No significant difference at $P \leq 0.05$, according to F testMeans with common letters are not significantly different at $P \leq 0.05$, according to Tukey's HSD test

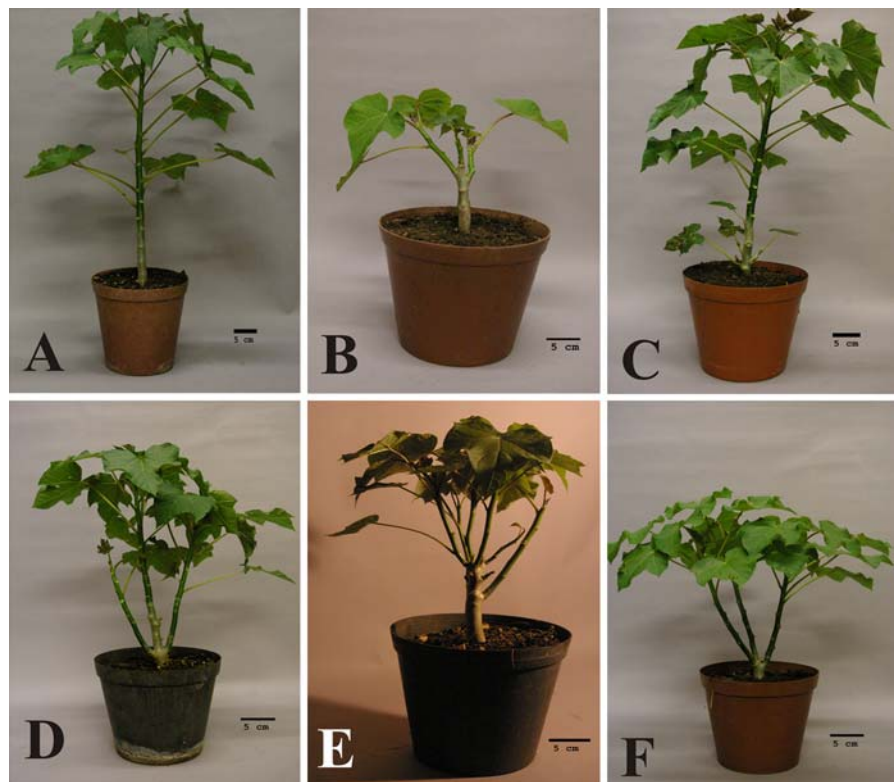
can cause some foliar chlorosis, and can even delay resumption of growth.

In the shade house (Fig. 2), foliar application of BA (12 mM) to five-month-old plants of *J. curcas* was very

effective in producing a maximum number of lateral branches. Consistent with this, Sansberro et al. (2006) showed an increase in number of branches of *Ilex paraguariensis* St. Hil. seedlings following foliar application of BA. At most of

Table 4 Effects of a single foliar application of maleic hydrazide (MH) and manual pruning (MP) on different growth parameters of *J. curcas*

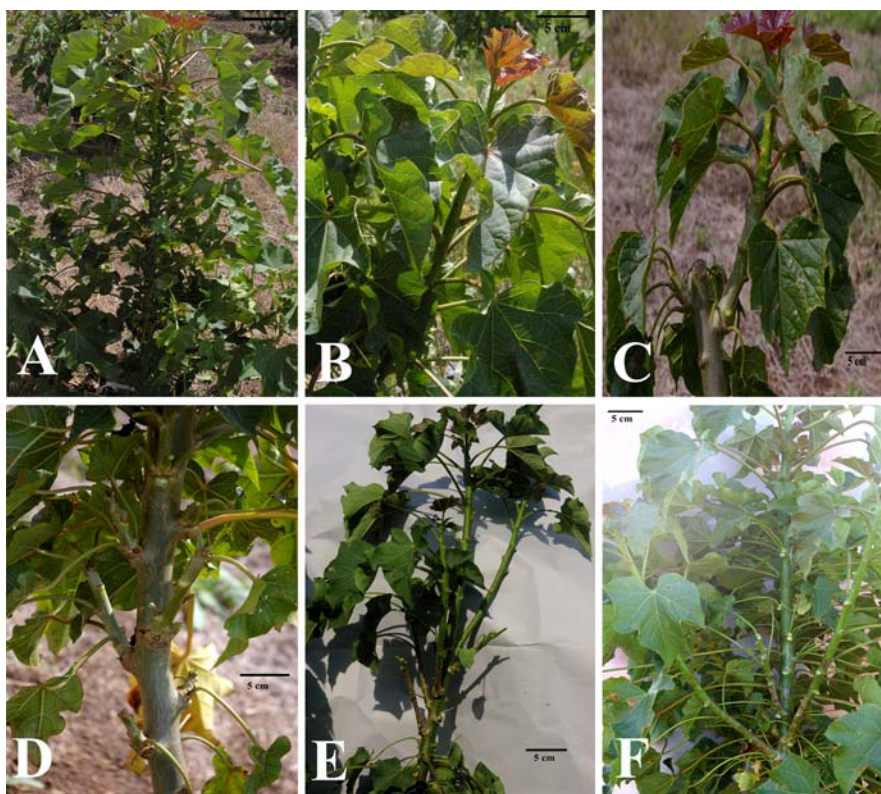
| MH (mM) | Five-month-old plants (shade house conditions) | | | Twelve-month-old plants (field conditions) | | |
|---------|--|--------------------------------|---------------|--|---------------------|--------------------|
| | Plant height (cm) | Shoot length (cm) ^a | No. of leaves | Plant height (cm) | Crown diameter (cm) | Stem diameter (cm) |
| | One month after spraying | | | Three months after spraying | | |
| 0 | 19.2a | 2.9 a | 9.7a | 5.5 ± 1.6 ^b | 18.4ab | 3.3 ^b |
| 2 | 5.24b | 1.2ab | 3.1b | 5.8 ± 2.3 | 19.77a | 2.9 |
| 3 | 8.41ab | 0.25b | 0.3c | 5.1 ± 1.4 | 5.33ab | 2.8 |
| 4 | 16.68a | 0.55b | 0.3c | 2.8 ± 0.6 | 4.75b | 3.1 |
| MP | 10.0 ab | 1.6ab | 9.9a | 4.0 ± 1.2 | 5.1b | 2.1 |
| | Four months after spraying | | | Seven months after spraying | | |
| 0 | 32.8a | 24.4a | 32.8a | 54.5ab ± 9.8ab | 220 ^b | 2.6 ^b |
| 2 | 25.4a | 17.7ab | 20.6b | 57.5a ± 5.5a | 239 | 2.1 |
| 3 | 13.7b | 11.9bc | 18.2b | 43.0ab ± 6.6ab | 215 | 2.4 |
| 4 | 10.9b | 13.2bc | 20.5b | 45.4ab ± 4.9ab | 217 | 2.3 |
| MP | 3.7b | 2.3c | 3.7c | 16.6b ± 13.6b ^b | 142 | 1.6 |

^a Measurement from the first leaf nodes^b No significant difference at $P \leq 0.05$, according to *F* testMeans with common letters are not significantly different at $P \leq 0.05$, according to Tukey's HSD test**Fig. 2** Influence of foliar application of plant growth regulators on lateral branching of *Jatropha curcas* under shade house conditions after 4 months. **(a)** one-year-old plant. **(b)** control. **(c)** manual pruning. **(d)** 12.0 mM BA. **(e)** 1.5 mM TIBA. **(f)** 6.0 mM DK. Bar scale = 5 cm. Acronyms same as in Fig. 1

the concentrations tested, BA decreased plant height and shoot length of *J. curcas* grown under shade house conditions. These results are in agreement with the findings of Henny (1986) who reported that BA increased lateral branches and decreased plant height of *Peperomia obtusifolia* L., resulting in shorter and more compact plants. Exogenous

application of BA had positive effects on the shoot growth of *Welkeria dieffenbachia* (Wilson and Nell 1983) and *Pinus* species (Boe 1990). Under field conditions (Fig. 3), after 7 months following BA treatment (12 mM), there was a significant improvement in number of branches in *J. curcas*. However, unlike the results of the shade house experiment,

Fig. 3 Influence of foliar application of plant growth regulators on lateral branching of *Jatropha curcas* (one-year-old) under field conditions after 7 months. (a) control. (b) manual pruning. (c) 12.0 mM BA 12 mM. (d) 1.0 mM TIBA. (e) 2.0 mM DK. Bar scale = 5 cm. Acronyms same as in Fig. 1



there was a non-significant increase in plant height and crown diameter with most concentrations of BA applied. This result suggests that the growth of BA-treated *J. curcas* plants may differ under shade house and field conditions.

Foliar application of TIBA to *J. curcas* plants at 1.5 mM under shade house conditions (after 4 months) and at 1.0 mM under field conditions (after 7 months) yielded more branches than the control. With both these growth conditions this effect was not immediate as the number of branches did not improve at early growth stages. Similarly, primary shoot development was delayed in *I. paraguariensis* with TIBA treatment (Sansberro et al. 2006). In the shade house, after 4 months' treatment, some concentrations of TIBA inhibited plant height, shoot length and number of leaves of *J. curcas* plants; however, under field conditions there were no significant effects.

In comparison to the shade house-treated plants, *J. curcas* plants that were treated in the field with DK (2.0 mM) yielded a maximum number of branches after 7 months. This indicates that under field conditions *J. curcas* plants responded much better to a lower concentration of DK (2.0 mM) than at an higher concentration (6.0 mM) in the shade house for enhancing the number of lateral branches. This positive effect of a low concentration of DK in the field may be associated with the age and maturity of *J. curcas* plants. Increasing concentrations of DK decreased plant height, shoot length and number of leaves when compared to untreated plants in the shade

house after 4 months. Similar results were reported for *Lonicera heckrotti* Rehd. (Bruner et al. 2000) and *Hedera helix* (Al-Juboory and Williams 1991) where DK increased the number of shoots and decreased shoot length. This effect was not consistent in field-treated plants. The observation of abnormal growth of *J. curcas* plants in pots (shade house) caused by high concentrations of DK supports the findings of Sansberro et al. (2006) that phytotoxicity is concentration-dependent in *I. paraguariensis*. Similar observations were also reported by Banko and Stefani (1996) who noticed slight chlorosis and leaf deformity in *Salvia farinacea* with foliar application of DK. In this study, foliar application of a low concentration of DK (2.0 mM) did not show any growth abnormalities in the field, suggesting that DK at low concentrations can be safely used in the field to promote branching of *J. curcas* plants.

Maleic hydrazide increased the number of lateral branches when compared to manual pruning and control plants after 4 and 7 months under shade house (2.0 mM) and field conditions (3.0 mM), respectively. These results indicate that the positive effect of MH on branching is only observed after a longer period, as in both cases there were no improvement at early growth stages. In the shade house after 4 months, MH (4.0 mM) suppressed plant height and number of leaves with some growth abnormalities. Studies have revealed that MH-treated plants lose or show impaired apical dominance (Naylor and Davis 1950). On

the other hand, no significant effect was noticed in the field-grown plants that exhibited normal growth.

Bearing fruit bunches at the apex of the branches is the normal fruiting behaviour of *J. curcas* plants. Therefore, limited branching is considered as one of the major factors limiting yield in *J. curcas*. Traditionally manual pruning is practiced to promote branching in this plant. This study was conducted to determine the potential of different PGRs to increase the number of lateral branches of *J. curcas* as possible substitution to the time-consuming, labour-intensive and expensive manual pruning. In the subsequent year following the foliar application of the PGRs, various parameters, such as flowering, fruit set, fruit characteristics, seed total oil content and oil free fatty acid (FFA) content were evaluated. All these parameters were significantly affected in response to different PGR treatments (H. Abdelgadir, unpublished data). This gives an indication that PGRs can be used in *J. curcas* for promotion of branching and increase of seed and oil yield. However, we cannot overlook the fact that this study presents only a preliminary guide to the doses and the effectiveness of the PGRs used.

Conclusions

Plant growth regulators tested in this study were effective in promoting branching of *J. curcas*. The influence of these PGRs was much more pronounced in the field, achieving significantly greater numbers of branches than manual pruning and untreated plants. Under field conditions DK (2.0 mM) was the best treatment followed by TIBA (1.0 mM), BA (12 mM) and MH (3.0 mM) respectively. This study suggests that PGRs can become a valuable tool for promoting *J. curcas* branching under field conditions.

Acknowledgments We thank Verus Farming Ltd, South Africa and the University of KwaZulu-Natal Pietermaritzburg for financial support. Professor Colin Everson generously made available the plants used in the field trials.

References

- Achten WMJ, Verchot L, Franken YJ, Mathijs E, Singh VP, Aerts R, Muys B (2008) *Jatropha* bio-diesel production and use. *Biomass Bioenergy* 32:1063–1084. doi:[10.1016/j.biombioe.2008.03.003](https://doi.org/10.1016/j.biombioe.2008.03.003)
- Al-Juboory K, Williams D (1991) Effects of two growth regulators on lateral branching and shoot growth of English ivy *Hedera helix*. *Plant Growth Regul Soc Am Q* 19:35–40
- Arzee T, Langenauer H, Gressel J (1977) Effects of dikegulac, a new growth regulator, on apical growth and development of three Compositae. *Bot Gaz* 138:18–28. doi:[10.1086/336891](https://doi.org/10.1086/336891)
- Augustus GDPS, Jayubalan M, Seiler GJ (2002) Evaluation and bioinduction of energy components of *Jatropha curcas*. *Biomass Bioenergy* 23:161–164. doi:[10.1016/S0961-9534\(02\)00044-2](https://doi.org/10.1016/S0961-9534(02)00044-2)
- Azam MM, Waris A, Nahar NM (2005) Prospects and potential of fatty acid methyl esters of some non-traditional seed oils for use as biodiesel in India. *Biomass Bioenergy* 29:293–302. doi:[10.1016/j.biombioe.2005.05.001](https://doi.org/10.1016/j.biombioe.2005.05.001)
- Bangerth F (1993) Polar auxin transport as a signal in the regulation of tree and fruit development. *Acta Hort* 329:70–76
- Bangerth F, Li CJ, Gruber J (2000) Mutual interaction of auxin and cytokinins in regulating correlative dominance. *Plant Growth Regul* 32:205–217. doi:[10.1023/A:1010742721004](https://doi.org/10.1023/A:1010742721004)
- Banko T, Stefani M (1996) Chemical growth control of *Salvia farinacea* ‘Victoria Blue’. *Proc SNA Res Conf* 41:214–215
- Boe A (1990) Effects of application of BA to seedlings of *Pinus sylvatica* and *Pinus nigra*. *J Environ Hort* 8:212–214
- Bruner L, Keever G, Kessler R, Gilliam C (2000) Growth regulation effects of *Lonicera heckrotti* (Goldflame Honeysuckle). *Proc SNA Res Conf* 45:263–264
- Chatfield SP, Stirnberg P, Frode BG, Lyeser O (2000) The hormonal regulation of axillary bud growth in *Arabidopsis*. *Plant J* 24:159–169. doi:[10.1046/j.1365-3113x.2000.00862.x](https://doi.org/10.1046/j.1365-3113x.2000.00862.x)
- Cline M (1991) Apical dominance. *Bot Rev* 57:318–358. doi:[10.1007/BF02858771](https://doi.org/10.1007/BF02858771)
- Emery RJN, Longnecker NE, Atkins CA (1998) Branch development in *Lupinus angustifolius* L. II. Relationship with endogenous ABA, IAA and cytokinins in auxiliary and main stem buds. *J Exp Bot* 49:555–562. doi:[10.1093/jexbot/49.320.555](https://doi.org/10.1093/jexbot/49.320.555)
- Everson C, Everson T (2006) Agroforestry systems for improved food production through the efficient use of water: Implementation of on-station trials. Deliverable submitted to the Water Research Commission—Project K5/148
- Gianfagna T (1995) Natural and synthetic growth regulators and their use in horticultural and agronomic crops. In: Davies PJ (ed) *Plant hormones: physiology, biochemistry and molecular biology*. Kluwer Academic Publishers, Dordrecht, pp 751–773
- Gour VK (2006) Production practices including post-harvest management of *J. curcas*. In: Singh B, Swaminathan R, Ponraj V (eds) *Biodiesel conference toward energy independence-focus of Jatropha*, Hyderabad, India, June 9–10. Rashtrapati Bhawan, New Delhi, India, pp 223–251
- Gressel J (2008) Transgenics are imperative for biofuel crops. *Plant Sci* 174:246–263. doi:[10.1016/j.plantsci.2007.11.009](https://doi.org/10.1016/j.plantsci.2007.11.009)
- Heller J (1996) Physic nut. *Jatropha curcas* L. 1. Promoting the conservation and use of underutilized and neglected crops. Institute of Plant Genetics and Crop Plants Research. Gate-rlsleben/International Plant Genetic Resources Institute, Rome, Italy, pp 21–22
- Henny RJ (1986) BA induces lateral branching of *Peperomia obtusifolia*. *J Am Soc Hort Sci* 20:115–116
- Hoffman I, Parups E (1964) Mode of action of maleic hydrazide in relation to residues in crops and soils. *Residue Rev* 7:96–113
- Ito A, Hayama H, Yoshioka H (2000) Effect of maleic hydrazide on endogenous cytokinin contents in lateral buds, and its possible role in flower bud formation on the Japanese pear shoot. *Sci Hort* (Amsterdam) 87:199–205. doi:[10.1016/S0304-4238\(00\)00174-6](https://doi.org/10.1016/S0304-4238(00)00174-6)
- Kureel RS (2006) Prospects and potential of *Jatropha curcas* for biodiesel production. In: Singh B, Swaminathan R, Ponraj V (eds) *Biodiesel conference toward energy independence-focus of Jatropha*, Hyderabad, India, June 9–10. Rashtrapati Bhawan, New Delhi, India, pp 43–74
- Kwon E, Criley RA (1991) Cytokinin and ethephon induce greater branching of pruned *Plumeria*. *Hortic Dig* 93:6–8
- Lalouem M, Fox JE (1989) Cytokinin oxidase from wheat. Partial purification and general properties. *Plant Physiol* 90:899–906. doi:[10.1104/pp.90.3.899](https://doi.org/10.1104/pp.90.3.899)
- Lovatt C (2006) Plant growth regulators for avocado production. *Plant Growth Regul Soc Am Q* 34:34–35

- Miguel LC, Longnecker NE, Ma Q, Osborne L, Atkins CA (1998) Branch development in *Lupinus angustifolius* L.I. Not all branch have the same potential growth rate. J Exp Bot 49:547–553. doi: [10.1093/jexbot/49.320.547](https://doi.org/10.1093/jexbot/49.320.547)
- Morey PR, Dahl BE (1975) Histological and morphological effects of auxin transport inhibitors on honey mesquite. Bot Gaz 136:274–280. doi: [10.1086/336814](https://doi.org/10.1086/336814)
- Naylor A, Davis E (1950) Maleic hydrazide as a plant growth inhibitor. Bot Gaz 112:112–113. doi: [10.1086/335632](https://doi.org/10.1086/335632)
- Ongaro V, Leyser O (2008) Hormonal control of shoot branching. J Exp Bot 59:67–74. doi: [10.1093/jxb/erm134](https://doi.org/10.1093/jxb/erm134)
- Richards D, Wilkinson RI (1984) Effect of manual pinching, potting-on and cytokinins on branching and flowering of *Camellia*, *Rhododendron*, and *Rosa*. Sci Hortic (Amsterdam) 23:75–83. doi: [10.1016/0304-4238\(84\)90047-5](https://doi.org/10.1016/0304-4238(84)90047-5)
- Sansberro PA, Mroginski CLA, Bottini RB (2006) Stimulation of lateral branch formation on *Ilex paraguariensis* (Aquifoliaceae) seedlings. Aust J Exp Agric 46:707–710. doi: [10.1071/EA04178](https://doi.org/10.1071/EA04178)
- Svenson S (1991) Rooting and lateral shoot elongation of *Verbena* following benzyl aminopurine application. J Am Soc Hortic Sci 26:391–392
- Wilson M, Nell T (1983) Foliar applications of BA increase branching of *Welkeri dieffenbachia*. J Am Soc Hortic Sci 18: 447–448