

# Intoxication of newly implanted coffee plants by simulated drift of the dicamba herbicide

Fabio Alessandro Muniz Pires<sup>1</sup>; Caroliny Pereira Santos<sup>2</sup>; Chayenne de Lira Ferreira<sup>3</sup>; Saul Jorge Pinto de Carvalho<sup>4</sup>

<sup>1</sup>Instituto Federal de Educação, Ciência e Tecnologia do Sul de Minas Gerais, Campus Machado, graduated in Agricultural Engineering, E-mail: fabiopires12@icloud.com

<sup>2</sup> Instituto Federal de Educação, Ciência e Tecnologia do Sul de Minas Gerais, Campus Machado, undergraduate student in Agricultural Engineering, E-mail: carolinyengagro@gmail.com

<sup>3</sup>Instituto Federal de Educação, Ciência e Tecnologia do Sul de Minas Gerais, Campus Machado, graduated in Agricultural Engineering, E-mail:chayenneferreira\_@hotmail.com

<sup>4</sup> Instituto Federal de Educação, Ciência e Tecnologia do Sul de Minas Gerais, Campus Machado, Professor Doutor, E-mail: sjpcarvalho@yahoo.com.br

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## Abstract

After the release of soybean cultivars resistant to the herbicide dicamba, this product should be used more frequently in weed management programs, which may increase the risk of drift to neighboring crops, since the molecules are volatile and might cause damage in non-target plants. This work was developed to evaluate the effects of sub-doses of the herbicide dicamba on young coffee plants, simulating a drift situation. The experiment was carried out in a greenhouse, with a randomized block design of eight treatments and five replications. Each plot consisted of a 4 L pot, filled with sieved clayey soil and tanned manure (3:1 v/v), with a seedling of Catuaí IAC 144 coffee. The treatments used were: 0 (check plots), 0.0001, 0.001, 0.01, 0.01, 0.1, 1.0, 10.0, and 100.0 g ha<sup>-1</sup> of dicamba. Plant's percentual phytotoxicity was evaluated up to 49 days after application (DAA), SPAD index (14, 28, and 42 DAA), and dry matter biomass, at 49 DAA. In all evaluations, no differences were observed regarding phytotoxicity for dicamba sub-doses of up to 0.01 g ha<sup>-1</sup>. The highest doses of dicamba caused visual injuries of up to 31%. At 49 DAA, there were no differences regarding dry matter biomass. Leaf symptoms caused by the herbicide were observed, such as curling, wrinkling, and epinasty. No plant death was observed. Compared to other crops, coffee might be considered more tolerant to sub-doses of dicamba, with adequate recovery capacity for reduced doses of this herbicide.

Keywords: Coffea arabica, phytotoxicity, auxinics, injury.

#### Introduction

Weeds are one of the main restrictive biotic factors when establishing crops of economic interest, since they may compete for space, light, water, and nutrients, thus hindering the development of the crop (CORRÊA et al., 2016). In addition, weeds present allelopathy, which prevents the development of other endemic species in the areas (MARINHO et al., 2017).

In recent decades, the use of herbicides has been considered the main form of weed control within production systems due to practicality, low cost, and management efficiency, when compared to manual and mechanical methods. Another factor that has promoted high rates of herbicide adoption is the introduction of genetically modified crops resistant to specific herbicides, such as glyphosate (GRUBE et al., 2011). However, the continuous use of a given same herbicide in incorrect dosages has favored the emergence of resistant weeds in agricultural areas (ZHOU et al., 2016).

Today, in Brazil, there are nine species of glyphosate-resistant weeds: *Amaranthus palmeri*, *Amaranthus hybridus*, *Chloris elata*, *Conyza bonariensis*, *Conyza canadensis*, *Conyza sumatrensis*, *Digitaria insularis*, *Eleusine indica*, and *Lolium multiflorum* (HEAP, 2020). The development of genetically modified crops resistant to herbicides with other mechanisms of action provides new technologies in crops management, such as the new varieties of Intacta 2 Xtend<sup>®</sup> soybean, which are resistant to the dicamba herbicide (BEHRENS, 2007; MELHORANÇA FILHO, PEREIRA, MARTINS, 2011; MOHSENI-MOGHADAM; DOOHAN, 2015). This modified soybean could be adopted as part of a management program for eudicot weeds resistant to herbicides commonly used in crops, such as glyphosate (SILVA et al., 2018).

The dicamba herbicide (3,6-dichloro-2methoxybenzoic acid) is a hormone growth regulator, which belongs to the class of herbicides that mimic auxin, the chemical group of benzoic acids (ZHOU et al., 2016). Due to the higher volatility of molecules, dicamba is more subject to drift, which is a deviation from the trajectory of particles, caused by drops of reduced size and wind influence, that may reach unwanted areas (USEPA, 2019). This drift of herbicides can reach crops far from the application site, reducing the efficiency of the application and negatively affecting the production of neighboring crops (SILVA et al., 2018).

Auxin herbicides act similarly to indoleacetic acid (IAA); however, they are more persistent and more active, causing damage to more sensitive crops even at very low concentrations (OLIVEIRA JÚNIOR, 2011; CARVALHO et al., 2022). Even at low doses, auxin herbicides present great efficacy in the control of broadleaf weeds. However, the occurrence of drift and the consequences for sensitive crops have been one of the problems regarding the use of transgenic crops resistant to auxin herbicides (EGAN; MORTENSEN, 2012; MUELLER; WRIGHT; Remund, 2013).

According to Lima e Silva (2020), this new technology may cause problems due to herbicide drift in adjacent crops, as soon as cultivars resistant to the dicamba herbicide begin to be commercialized in Brazil. The drift problem may mainly affect locations with increased soybean crops in recent years, such as south of Minas Gerais, where the crop is often adjacent to coffee plantations, which are not genetically modified to tolerate herbicide exposure. Therefore, this work aims to evaluate the susceptibility of young coffee plants exposed to sub-doses of the dicamba herbicide, simulating a drift condition.

### Material and methods

This study was carried out in a greenhouse of the Instituto Federal de Educação, Ciência e Tecnologia do Sul de Minas Gerais (IFSULDEMINAS), Machado campus (21°45'S; 45°55'W; 850 m altitude), from March to November 2019. Each experimental unit consisted of a 4 L vessel, with perforated bottom, filled with sieved clay soil mixed with tanned cattle manure, in a ratio of 3:1 v/v. On March 13, 2019, coffee seedlings (*Coffea arabica* L.) cv. Catuaí Vermelho IAC 144, with approximately nine months of cultivation and seven to eight pairs of leaves, were transplanted into pots, where they remained until the conclusion of the experiment, one plant per pot.

After transplantation, the plants were kept in a greenhouse for acclimatization until September 2019, when herbicide treatments were administered. The plots were properly fertilized with one gram of commercial fertilizer at 15:15:20 (N:P:K + micronutrients) on March 27, 2019. Additionally, a spraying with copper hydroxide (6 mL / 400 mL  $H_2O$ + 0.5% Of Agral) was carried out on August 14, 2019. The plants developed adequately, without water or nutritional deficiency. The greenhouse includes automated irrigation, properly regulated, which ensures the proper water maintenance of plants.

A randomized block design was adopted with eight treatments and five replicates, totaling 40 plots. Even after the selection of homogeneous material, each experimental block was defined by a set of plants similar in size and number of leaves to avoid any possible interference of plant size in the experimental results. The treatments consisted of different doses of dicamba (g ha<sup>-1</sup>): 0.0001, 0.001, 0.01, 0.1, 1.0, 10.0, and 100.0, as well as check plots without application (zero).

The sprays were carried out on September 11, 2019, directly on the plants in formation, with the first pairs of plagiotropic branches starting to grow. For this, the plants were removed from the greenhouse and sprayed in an external environment, with lateral bulkheads to the pots to allow drift chance. A coastal spray pressurized by carbon dioxide ( $CO_2$ ) was used coupled to a spray bar with two tips of the TTI 110.02 fan type, spaced at 0.50 m, operating with a constant pressure of 2.5 bar, appropriately calibrated for 200 L ha<sup>-1</sup> spray volume and positioned at 0.5 m from the plants. In all applications, deionized water was used as a vehicle.

After complete drying of the spray, the plants were transported back to the greenhouse for further evaluation of phytotoxicity at 7, 14, 21, 28, 42, and 49 days after application (DAA). Phytotoxicity was evaluated considering a visual scale of variable symptoms between zero and 100% of damage, in which zero represented healthy plants, without symptoms, and 100% represented dead plants (SBCPD, 1995). At 14, 28, and 42 DAA, the Soil Plant Analysis Development (SPAD) index was evaluated using the Konica-Minolta502 Plus device. In this measurement, the average sampling of three leaves per plot was conducted, two new leaves (top third of plants) and one old leaf (middle third of plants).

At 49 DAA, all plots were washed in running water for root cleaning and, then, sent for drying. In the laboratory, all plants were dried in a greenhouse with forced air circulation, at 70 °C, for 72 hours, to obtain dry matter mass. After the procedure, the aerial part and roots of each treatment were weighed. All data were submitted to the F test application in variance analysis, followed by the Scott-Knott clustering test, both with 5% significance. Even with the existence of quantitative treatments, regressions

were adopted due to the low amplitude of the sampled results and greater discrimination of the data promoted by the means test.

## **Results and discussion**

Table 1 shows the intoxicating data of the simulated drift of dicamba on coffee seedlings in the first two evaluations. At 7 DAA, low levels of intoxication were observed at the lowest doses, that is, no significant differences were observed regarding the check plots for sub-doses of up to 0.01 g ha<sup>-1</sup>. On the other hand, at the higher doses, intoxication levels reached 10.6% and 20%, demonstrating significance according to the F test. Lima e Silva (2020) observed up to 9.7% phytotoxicity at seven DAA in young *Eucalyptus urograndis* plants with doses of up to 120 g ha<sup>-1</sup>. This demonstrates that coffee is more sensitive to dicamba drift than eucalyptus crops.

Dicamba is a product of growing importance since it is an alternative for the control of certain glyphosate-resistant weeds, such as plants of the genus *Conyza* and other problematic plants to the agroecosystem, and since few plants are resistant to its mechanism of action (SANTOS, 2017). The introduction of dicamba-tolerant crops in the agricultural market has raised the high rates of commercialization and adoption of this product outside the country, increasing the risk of the drift of this herbicide on non-target plants (VIEIRA et al., 2020).

From 14 DAA, doses above 1.0 g ha<sup>-1</sup> were considered different from the check plots according to the Scott-Knott test. Intoxication values increased between treatments, with the peak of phytotoxicity at 31% damage, this being the highest level among treatments identified for the highest dose. A similar behavior occurs in soybean plants, since, according to Costa (2019), the maximum level of injury by the herbicide in the crop is perceived in the 14-day evaluation.

Treatments (g ha <sup>-1</sup> )	Phytotoxicity		SPAD
	7 DAA	14 DAA	14 DAA
Check plots	0.0 A	0.0 A	41.34
0.0001	0.4 A	1.0 A	38.90
0.001	1.2 A	1.6 A	39.44
0.01	0.0 A	0.6 A	41.32
0.1	3.0 B	2.6 A	42.14
1	3.2 B	5.2 B	42.26
10	10.6 C	13.4 B	32.66
100	20.0 D	31.0 C	37.92
F Test	61.423*	85.814*	1.036 <sup>NS</sup>
CV (%)	41.96	37.14	17.71

**Table 1.** Phytotoxicity<sup>1</sup> of dicamba on freshly planted coffee seedlings, evaluated at 7 and 14 days after application (DAA), and Soil Plant Analysis Development (SPAD) index evaluated at 14 DAA. Machado, MG, 2019.

<sup>1</sup>Averages followed by equal letters in the columns do not differ from each other according to the 5% significance Scott-Knott test; CV – coefficient of variation; \*Significant F test at 5%; <sup>NS</sup>F test not significant. **Source:** By the authors (2020).

Table 2 shows, at 21 DAA, a gradual reduction in plant intoxication values, indicating the ability of coffee plants to overcome the damage caused by the herbicide molecule. According to Costa (2019), a similar result occurs with soybeans, demonstrating that the effects of dicamba take a certain time to manifest and that plants display mechanisms that allow them to recover, at least in part, from the damage suffered by dicamba drift.

From 28 DAA, phytotocity levels ranging from 0 to 24% were observed among the treatments. According to Lima and Silva (2020), eucalyptus plants also have mechanisms that allow the gradual recovery of the damage caused after exposure to the herbicide. Similar behavior was observed in coffee since, after the peak of intoxication (14 DAA), there was a gradual reduction in symptoms.

From 42 DAA (Table 3), no symptoms of damage were observed at the lowest dosages (0.0001, 0.001, and 0.01 g ha<sup>-1</sup>). Symptoms were noted only on plants that received dicamba drift from 1.0 g ha<sup>-1</sup> or higher. At 49 DAA, low levels of phytotoxicity were observed in

most treatments. Plants that received higher herbicide dosages presented a higher reduction of symptoms compared to previous evaluations.

After the evaluation of the treatments, symptoms were more prevalent and accentuated in the young tissues of the plants. According to Vidal (1997), the older leaves of some plant species are less affected after exposure to auxin herbicides due to the low presence of meristematic tissues, where the herbicides act.

The Minolta SPAD-502 equipment, which measures the intensity of green leaf coloration, has been used for the quantification of chlorophylls due to its speed, its simplicity, and, mainly, its nondestructive evaluation of leaf tissue (RAMOS, MONNERAT, PINHO, 2013). According to the evaluations, the lowest value obtained was 32.66 (Table 1), and the highest was 56.9 (Table 3). In general, no significant differences regarding to chlorophyll contents were observed to herbicide treatments, except for the evaluation of 42 DAA, at doses of 0.01 and 100.0 g ha<sup>-1</sup>.

Regarding the dry biomass of coffee (Table 4), the significance of the F test was detected only for

Treatments (g ha <sup>-1</sup> )	Phytotoxicity		SPAD
	21 DAA	28 DAA	28 DAA
Check plots	0.0 A	0.0 A	50.96
0.0001	0.6 A	0.8 A	44.26
0.001	2.0 A	2.4 A	45.22
0.01	0.0 A	2.2 A	46.00
0.1	2.2 A	5.8 B	49.34
1	0.6 A	7.0 B	48.04
10	10.4 B	17.2 C	49.62
100	22.6 C	24.0 D	51.58
F Test	48.291	37.577	0.701 <sup>NS</sup>
CV (%)	53.40	42.53	14.79

**Table 2.** Phytotoxicity<sup>1</sup> of dicamba on freshly planted coffee seedlings, evaluated at 21 and 28 days after application (DAA), and Soil Plant Analysis Development (SPAD) index evaluated at 28 DAA. Machado, MG, 2019.

<sup>1</sup>Averages followed by equal letters in the columns do not differ from each other according to the 5% significance Scott-Knott test; CV – coefficient of variation; \*Significant F test at 5%; <sup>NS</sup>F test not significant. **Source:** By the authors (2020).

the root mass, in which the herbicide treatments were always equal to or superior to the check plots. These data show a possible hormetic effect of dicamba on root growth, up to the dose of  $0.01 \text{ g} \text{ ha}^{-1}$ ; however, no constancy was observed between treatments, since, this effect

was not presented at a dose of 0.001 g ha<sup>-1</sup>. It is known that the hormonal activity of auxinmimicking herbicides, such as dicamba, could cause diverse manifestations in plant physiology, including hormesis; however, this analysis does not fit within the scope of this study.

Treatments (g ha <sup>-1</sup> )	Phytotoxicity		SPAD
	42 DAA	49 DAA	42 DAA
Check plots	0.0 A	0.0 A	43.1 B
0.0001	0.0 A	0.0 A	44.9 B
0.001	0.0 A	0.4 A	42.9 B
0.01	0.0 A	0.0 A	50.5 A
0.1	0.0 A	1.8 A	48.1 B
1	1.6 A	1.2 A	46.4 B
10	10.0 B	4.2 B	43.2 B
100	25.6 C	18.6 C	56.9 A
F Test	93.929*	64.455*	3.757*

**Table 3.** Phytotoxicity<sup>1</sup> of dicamba on freshly planted coffee seedlings, evaluated at 42 and 49 days after application (DAA), and Soil Plant Analysis Development (SPAD) index evaluated at 42 DAA. Machado, MG, 2019.

<sup>1</sup>Averages followed by equal letters in the columns do not differ from each other according to the 5% significance Scott-Knott test; CV - coefficient of variation; \*Significant F test at 5%.

54.04

45.36

Source: By the authors (2020).

CV (%)

11.80

Treatments (g ha <sup>-1</sup> )	Dry Matter Mass (g plot <sup>-1</sup> )			
	Roots	Aerial part	Total	
Check plots	11.1 B	27.4	38.5	
0.0001	20.1 A	29.6	49.7	
0.001	13.0 B	28.9	41.8	
0.01	20.2 A	27.8	48.0	
0.1	12.4 B	28.7	41.0	
1	9.4 B	28.6	38.1	
10	11.3 B	29.0	40.3	
100	13.8 B	28.4	42.2	
F Test	3.466*	0.167 <sup>NS</sup>	1.590 <sup>NS</sup>	
CV (%)	35.17	13.42	17.71	

**Table 4.** Roots and aerial part dry matter mass and total seedlings of freshly planted coffee plants submitted to sub-doses of dicamba herbicide evaluated at 49 days after application (DAA). Machado, MG, 2019.

<sup>1</sup>Averages followed by equal letters in the columns do not differ from each other according to the 5% significance Scott-Knott test; CV – coefficient of variation; \*Significant F test at 5%; <sup>NS</sup>F test not significant. **Source:** By the authors (2020).

Regarding to aerial and total biomass, no significance regarding to F test was identified, that is, all treatments tested reached aerial dry matter biomass equal to the check plots, as well as total dry matter biomass. According to Christoffoleti et al. (2015), after herbicide exposure, the following symptoms were visually noted in sensitive plants: tumors in the apical meristem region; abnormality of growth tissues, such as wrinkling and shriveling of the stem and leaves; epinasty; thickening of the stem and multiplication of the root system. In the aereial part of the coffee, symptoms such as epinasty, wrinkling and shriveling of leaves were observed.

Dicamba is an auxin-mimicking herbicide and, as such, its presence impacts several plant physiological processes naturally sensitive to auxin hormones, such as plant growth, meristematic differentiations, leaf deformations, and calluses in stems. Some of these manifestations were present in coffee seedlings at a lower intensity than is commonly observed in more sensitive crops, such as genetically modified soybean (COSTA, 2019; CARVALHO et al., 2022). In general, it is possible that coffee is less sensitive to the dicamba molecule due to its metabolic response to the herbicide presence (detoxification), or even due to lower rates of herbicide absorption and translocation in coffee plant tissues, which culminated in a lower general manifestation of symptoms.

### Conclusions

No significant differences regarding dicamba herbicide phytotoxicity were observed for subdoses of up to 0.01 g ha<sup>-1</sup> in all evaluations performed. The doses of 10.0 and 100.0 g ha<sup>-1</sup> promoted greater damage in young coffee plants, with a damage level of up to 31%. The main symptoms observed were epinasty, wrinkling, and wilting of leaves.

The peak of phytotoxicity was observed at 14 DAA, recovery of damages caused by drift after 14 DAA, and no plant death at the tested doses. No differences regarding dry matter biomass were observed.

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