

# Measuring population fluctuation of jatropha stem-borer [*Cophes notaticeps* (Marshall)] in the Brazilian Cerrado using a new trap

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## Abstract

This study aimed to monitor the population fluctuation of *Cophes notaticeps* (Marshall) (Coleoptera: Curculionidae) in a jatropha (*Jatropha curcas* L.) plantation in the Federal District, Brazil, through the use of a new trap, combined with different attractive lures and trap colors. The study was conducted at Embrapa Cerrados (Planaltina/DF) in a field trial with 720 plants. The new trap, named CPAC16, was made with PVC pipes (100 mm in diameter) of about 40 cm in length, with a window (8 × 10 cm<sup>2</sup>) in its center to enable insect access. A lure compartment was fixed on the inside of the trap's top, and a pot, containing water and detergent, was placed at its bottom to collect the insects. The traps were painted in red, yellow, blue and green. Molasses, ethanol and pineapple were used as attractive lures. The traps were in the field trial area between May 2013 and April 2014 and the insects were collected weekly. The incidence of plants presenting damage caused by *C. notaticeps* was evaluated in the beginning and at the end of the study. The CPAC16 trap proved efficient in monitoring *C. notaticeps*. About 3494 of *C. notaticeps* adults were collected during the study. There were no significant differences among traps painted with different colors. The most attractive lure was molasses collecting 75.2% ( $n = 2627$ ) of the specimens. Although the population peak occurred in December ( $n = 1162$ ), *C. notaticeps* were collected throughout the year. The incidence of plants attacked by *C. notaticeps* ranged from 66.4% (start) to 100% (end).

**Keywords:** *Jatropha curcas* L., population dynamics, biofuel, baits, colors

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## Introduction

Global climate change has led many countries to look for different options in their energy matrix, mainly alternatives to reduce the use of fossil fuels, which are pointed out as

partially responsible for the greenhouse effect (Baede *et al.*, 2001; Zickfeld *et al.*, 2012; Friedlingstein *et al.*, 2014). Among the raw materials for biodiesel production, the National Program for Production and Use of Biodiesel (NPPUB) in Brazil (Brasil, 2004, 2005) included jatropha (*Jatropha curcas* L.) (Euphorbiaceae) as a promising species (Ferreira & Cristo, 2006). The expectations were that, once jatropha was grown commercially, it would have high oil productivity, combined with low production costs and resistance to water stress (Ferreira & Cristo, 2006).

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However, commercial cultivation of jatropha in Brazil lacks basic agronomic information to verify its economic viability. In this context, studies related to plant health, such as the occurrence of pests and the development of management strategies for this species, are fundamental to the successful expansion of this crop. In central Brazil, an insect pest species *Cophes notaticeps* (Marshall) (Coleoptera: Curculionidae), commonly known as jatropha stem-borer, was identified (Oliveira *et al.*, 2011). This species was first recorded in jatropha plants by Bondar in the state of Bahia (Costa Lima, 1922, 1956; Silva *et al.*, 1968). This insect, whose larva develops inside the plant tissue, mainly at the base of the stem, offers few alternatives to curative control (Oliveira *et al.*, 2011). Preventive control measures, based on adult behavior may be feasible to avoid plant infestation. Therefore, developing knowledge about the bio-ecological aspects of this pest is essential, especially its population dynamics.

Control measures for Curculionidae pests, mainly on annual crops, has been restricted to cultural measures such as crop rotation, removal of infested plant organs, destruction of crop residues and alternative host plants, since chemical control is inefficient (Callan, 1942; Monte, 1940, 1945; Jansson, 1992). However, these practices cannot be applied to jatropha, because it is a perennial plant and *C. notaticeps* mainly attacks the trunk (Oliveira *et al.*, 2011).

The genus *Cophes* Champion occurs throughout the Americas, from Canada to Argentina (Alonso-Zarazaga & Lyal, 1999). This genus is composed of 242 species, with 202 occurring in South America, and currently includes the species that once belonged to the genera *Graphonotus* Chevrolat, *Coelosternus* Schoenherr and *Sternocoelus* Kuschel (Wibmer & O'Brien 1986; Alonso-Zarazaga & Lyal, 1999). Although it has already been reported as a jatropha (Peixoto, 1973; Siqueira Franco & Gabriel, 2008) and cassava pest in Brazil (Monte, 1940, 1945), there is no information about the biology and ecology of *C. notaticeps*. Data concerning bio-ecological aspects of this genus are restricted to *C. granicollis* (Pierce) (= *Coelostenus granicollis*), an insect pest of cassava (*Manihot sculenta* Crantz). For this species the larval stage, which develops inside cassava stems, lasts about 2 months and the insect completes its cycle in about 3 months (up to four generations can occur per year), with adult population peaking in December and January (Monte, 1940, 1945; Oliveira & Paula-Moraes, 2011). Monitoring the populations of these coleborers in Brazil has been accomplished with traps that use cassava roots "in natura" as an attractant (Carvalho *et al.*, 2009).

The use of baited traps, which release volatile lures, such as alcohol or plant material in fermentation, has been widely used for monitoring populations of insects from the Curculionidae family (Coleoptera) (Vacas *et al.*, 2013; Miller *et al.*, 2015; Flechtmann & Atkinson, 2016). The capture efficiency, however, may depend on the trap type, bait and adult behavior of each species studied.

Our hypothesis is that *C. notaticeps* may have an adult population peak in a given time of the year, which would be detected through insect monitoring by baited traps. This would direct the adult control measures in that period, reducing the infestation of jatropha plants.

The aim of this study was to determine the population fluctuation of *C. notaticeps* in a jatropha plantation in the Cerrado of the Federal District, through the use of a new trap, and different types of attractive lures and trap colors.

## Materials and methods

### Trial area

The study was conducted in an area of about 1.3 ha, with 5-year old jatropha plants, located at Embrapa Cerrados in Planaltina/DF (15°35'42.7"S; 47°44'14.8"W; 1039 m). The experimental area consisted of 15 rows, 12 m in length, with 48 plants per row, totaling 720 plants. This site was chosen because *C. notaticeps* activity had previously been reported in the area.

### New trap

Based on *C. notaticeps* habit of damaging the region around the base of the stem (Oliveira *et al.*, 2011), the traps were installed close to the plants, at the ground level, rather than suspended, as most of the Curculionidae traps. The traps, called CPAC16, were made of a 100 mm in diameter PVC pipe, cut into sections of 40 cm in length with a window of 10 × 8 cm in the center, to enable insect access. A cap was placed at the top of each section. A bait compartment, consisting of a plastic pot (10 cm tall and 6 cm in diameter) with holes on the sides to help the lures smell dissipate, was fixed on the inside of the cap. A plastic pot (9 cm tall and 10 cm in diameter), containing water and detergent, was placed at the bottom of the trap, directly under the bait, to collect the insects attracted to the trap (fig. 1). The traps were painted with spray paint in red (code 8761 – wavelength range for highest % reflectance: 630–740 nm), yellow (code 8591 – wavelength range for highest % reflectance: 580–590 nm), blue (code 8621 – wavelength range for highest % reflectance: 450–480 nm) and green (code 8731 – wavelength range for highest % reflectance: 510–550 nm) (Colorgin<sup>®</sup>, Sherwin-Williams do Brasil, Taboão da Serra/SP, Brazil). We selected the primary colors (red, yellow and blue) and green, which resembled the color of the vegetation. Since many Curculionidae are attracted by compounds such as alcohols and esters obtained from the fermentation of plant material, molasses from sugarcane, ethanol (92.8° GL) and pineapple (macerated) were used as attractive lures for *C. notaticeps* adults. Each bait compartment received about 100 ml of lure solution. Baits were replaced weekly.

### Sampling

Traps were sampled weekly for a period of 12 months between May 2013 and April 2014. The field plot design was a randomized block in a 4 × 3 factorial. The first factor being the four colors (red, yellow, blue and green) and the second factor the three types of bait (molasses, alcohol and pineapple). There were five repetitions totaling 60 traps. The traps were installed near the base of the jatropha plants. The distance between traps in the same row was approximately 8 m and 12 m between rows. They were placed in steel screened cages to prevent the baits from being taken by wild animals, especially small primates (Oliveira & Mendonça, 2011). After sampling, the insects were transported to the laboratory of Entomology at Embrapa Cerrados, where they were screened using a stereoscopic microscope, separating the Coleoptera and *C. notaticeps* specimens of other insects.

The identification of *C. notaticeps* was based on comparisons with specimens previously identified by Dr Sergio Vanin (Department of Zoology, Institute of Biosciences, University of São Paulo, São Paulo, SP) [Diagnostic characters

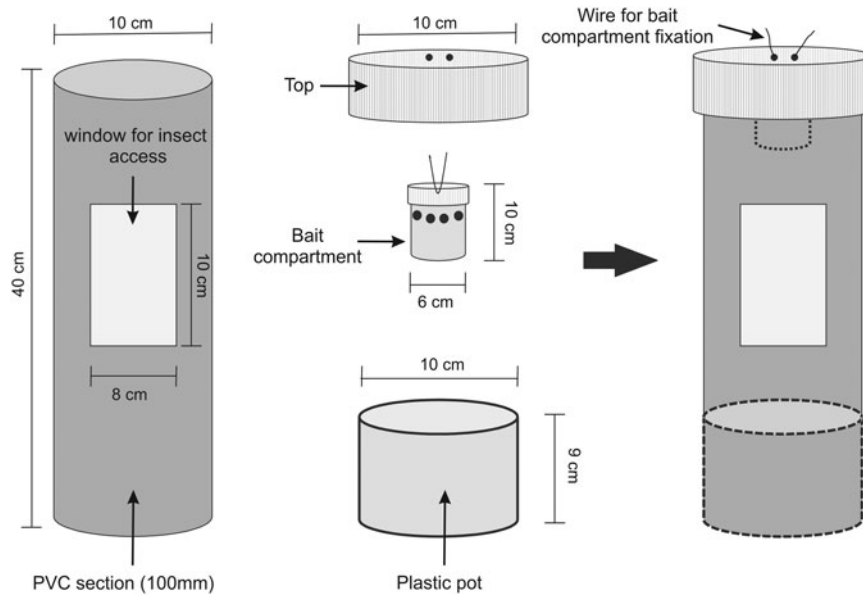


Fig. 1. Trap model CPAC 16 used to collect *Cophes notaticeps* in the jatropha trial area in the Federal District, Brazil, between May 2013 and April 2014.

of the species and keys for taxonomic identification can be found at Fiedler (1942, 1943)]. Specimens vouchers of the insects collected, were mounted on entomological pins, labeled and deposited in the Entomological Museum of Embrapa Cerrados (CPAC).

To characterize the presence of the jatropha stem-borer and the development of the epidemics in the experimental area, a survey was done, before the start and at the end of the sampling period, to calculate the percentage of plants showing signs of the infestation, mainly the presence of holes near the base of the plant and resin exudation or accumulation of sawdust (Oliveira *et al.*, 2011).

#### Data analysis

As the data related to *C. notaticeps* adults collected during the experiment represent a quantitative variable with a cumulative frequency distribution, the weekly numbers of insects obtained for each trap, were added during the collection period. Box-Cox transformation was applied to the data set (Box & Cox, 1964) and normality was verified by the Shapiro–Wilk test; Kolmogorov–Smirnov; Cramer-von Mises and Anderson-Darling. Data were subjected to an ANOVA using the general linear model PROC GLM (SAS Institute, 2001). The objective of this analysis was to determine whether there are statistically significant differences in the number of *C. notaticeps* adults collected in the sample period in relation to colors and types of bait used in traps, and verify the existence of interaction between these two factors. The averages for each factor (color and bait) were compared by Tukey's test at the 0.05 level of probability (SAS Institute, 2001). To measure the degree of association between the dependent variable (quantitative) 'number of *C. notaticeps* collected per month' and the monthly values of the independent variables (climate data: average monthly temperature, average monthly relative humidity and monthly cumulative rainfall) a correlation analysis (Spearman) was calculated using the Statistica program

version 13 (Dell, 2015). This non-parametric analysis, which is used for quantitative discrete variables (i.e., number of adult insects collected per trap) does not require data normality or linearity.

#### Results

Of all the Coleoptera captured, 3494 specimens were *C. notaticeps* adults (30.1%). Among the Coleoptera, except for *C. notaticeps*, the most abundant species belonged to the Nitidulidae and Histeridae families. The population peak of the jatropha stem-borer occurred in December, when 1162 specimens, representing 33.3% of the total, were captured. The month with the lowest numbers was October, when only 24 specimens (0.7%) were captured. Therefore, *C. notaticeps* adults were present at the trial area during the entire sampling period (fig. 2). The CPAC16 trap captured, on average, about 1.2 *C. notaticeps* adults per trap, ranging from 0.4 to 19.4 adults per trap. There was no significant interaction between the four trap colors used and the three attractive lures ( $F = 0.86$ ;  $P = 0.53$ ).

It was observed that data presented normality (Shapiro–Wilk  $W = 0.98$ ,  $P = 0.36$ ; Kolmogorov–Smirnov  $D = 0.07$ ,  $P > 0.15$ ; Cramer-von Mises  $W-Sq = 0.04$ ,  $P > 0.25$  and Anderson-Darling  $A-Sq = 0.29$ ,  $P > 0.25$ ) and there was no interaction between the factors 'colors' and 'lures' used in the experiment ( $F = 0.82$ ;  $P = 0.56$ ).

Although there was no statistically significant differences among the four colors used to paint the traps ( $F = 1.53$ ,  $P = 0.21$ ), the ones painted in red captured the highest absolute number of specimens ( $n = 1200$ ), representing 34.3% of the total, followed by the yellow color ( $n = 953$  or 27.3%), blue ( $n = 720$  or 20.6%) and green ( $n = 621$  or 17.8%) (Table 1).

There were statistically significant differences among the attractive baits ( $F = 28.46$ ,  $P < 0.0001$ ) (Table 1). Traps baited with sugarcane molasses captured significantly more adult *C. notaticeps* ( $n = 2627$  or 75.2%) than those which used ethyl

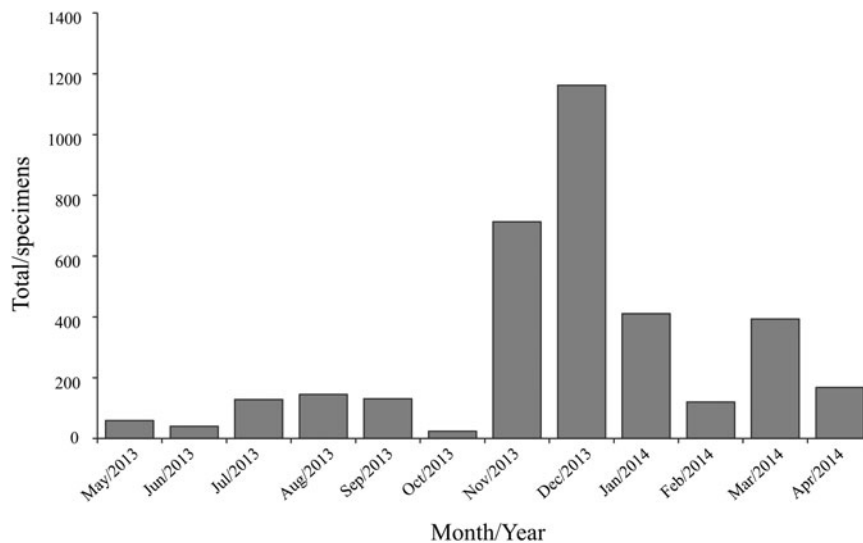


Fig. 2. Total monthly number of *Cophes notaticeps* adults captured with trap model CPAC16, using different baits [sugarcane molasses, ethanol (98.7° GL) and pineapple (macerated)] and colors (red, yellow, blue and green) in a jatropha trial area in the Federal District, Brazil, between May 2013 and April 2014.

Table 1. Mean number/trap/week of *Cophes notaticeps* adults captured with different types of baits [sugarcane molasses, ethanol (98.7° GL) and pineapple (macerated)] and colors (red, yellow, blue or green) using the trap model CPAC16 in a jatropha trial area in the Federal District, Brazil, between May 2013 and April 2014.

Bait/Color	Red	Yellow	Blue	Green	Average <sup>1</sup>
Molasses	3.71	3.02	2.11	1.88	2.68A
Ethanol	0.98	0.70	0.55	0.46	0.67B
Pineapple	0.21	0.17	0.28	0.20	0.21C
Average <sup>1</sup>	1.63A	1.30A	0.98A	0.84A	1.19

<sup>1</sup>Means followed by the same letter in the column (bait) or in the line (color) do not differ by Tukey's test at  $P < 0.05$ .

alcohol or pineapple. Traps baited with ethyl alcohol ( $n = 657$  or 18.8%) captured significantly higher numbers than those using pineapple ( $n = 210$  or 6.0%).

There was no correlation (Spearman) between the meteorological data (rainfall  $R^2 = 0.50$ ,  $P = 0.10$ ; temperature  $R^2 = 0.24$ ,  $P = 0.45$ ; and relative humidity  $R^2 = 0.30$ ,  $P = 0.34$ ) and the monthly average of *C. notaticeps* captured during monitoring.

The survey done at the beginning of the trial (May 2013) identified 66.4% plants with signs of attack by *C. notaticeps*. After 1 year (April 2014) signs were detected in all plants of the experimental area (100%).

## Discussion

For most insects, population fluctuation is directly influenced by abiotic factors. Among these factors, the distribution of rainfall appears to be one that has the greatest importance, for directly influencing changes in temperature, relative humidity, plant growth, and also promoting an increase in the population of natural enemies (Oliveira & Frizzas, 2008; Silva *et al.*, 2011). In the central region of Brazil the distribution of rainfall follows a pattern where, approximately, 87% (1212

mm) of it occurs between October and March, and 13% (185 mm) between April and September (Silva *et al.*, 2008). Most insect species in this biome concentrate their adult populations in the first half of the rainy season (Oliveira & Frizzas, 2008; Silva *et al.*, 2011).

Our results demonstrated that *C. notaticeps* adults also have a higher abundance during the rainy season, when 80.8% were trapped. Of the total number of specimens collected during the trial, 53.7% were captured in the months of November and December. However, unlike many other Coleoptera whose adult populations are observed only in the rainy season (Oliveira *et al.*, 2007, 2008), *C. notaticeps* representatives were captured during the entire sampling period, indicating that, for this species, development appears to be continuous during the year. However, in the rainy season, the jatropha plants resume vegetative growth and this physiological state of the plant seems to be more suitable for *C. notaticeps*, leading to a population increase. Population studies with Curculionidae in Brazil, have pointed out the presence of adults throughout the year, peaking in the rainy season (Monte, 1940; Tironi *et al.*, 2005), as observed for the species *C. granicollis* (Monte, 1940), *Cyrtomon luridus* Boheman (Tironi *et al.*, 2005), *Neochetina eichhorniae* Warner, *N. bruchi* Hustache (Sousa *et al.*, 2011) and *Rhynchophorus palmarum* L. (Takada *et al.*, 2011). The absence of abrupt climatic changes, such as severe winters with freezing temperatures, and the cryptic life habit of most Curculionidae seem to allow adults to develop throughout the year in Brazil. On the contrary, in other countries abiotic factors such as temperature and rainfall distribution can lead species of Curculionidae to concentrate the adult population only during favorable periods of the year as it occurs, for example, with *Sternechus subsignatus* Boheman in Argentina (Socias *et al.*, 2014) and *Curculio elephas* Gyllenhal in France (Menu, 1993).

In this study no correlation was found between climate variables and the population dynamics of *C. notaticeps*. In fact, many studies have failed to demonstrate this direct relationship, especially with rainfall data. However, rain has been

identified as the 'trigger' for the restart of development of many insect groups after periods of inactivity (i.e. diapause) and, also, for their population growth, especially in regions where there is a clear alternation between dry and rainy seasons (Wolda, 1988; Oliveira & Frizzas, 2008; Silva *et al.*, 2011).

Representatives of *Curculionidae*, except those of the *Entiminae* subfamily, usually have adults and larvae with restricted hosts (monophagous or oligophagous). Adults usually cause very little damage to host plants and larvae have cryptic habits, feeding inside roots, stems or reproductive organs (Casari & Ide, 2012). That seems to be the case for *C. notaticeps*. Unlike other insect species, whose juveniles and adults are free living and are subject to direct influence of abiotic factors and natural enemies, the larvae of *C. notaticeps*, protected inside the stem of the plants, seem to be able to develop throughout the year, allowing the emergence of adults even during the dry season, when environmental conditions are less favorable.

Baited traps of all kinds have been widely used for monitoring species of *Curculionidae*, especially those of economic interest, such as *R. palmarum* in palms (Aldryhim & Ayedh, 2015; Correia *et al.*, 2015; Löhr *et al.*, 2015), *Hypothenemus hampei* Ferrari in coffee (Pereira *et al.*, 2012; Aristizábal *et al.*, 2015; Fernandes *et al.*, 2015) and species of *Scolitinae* and *Platypodinae* in forest trees (Carvalho & Trevisan, 2015; Steining *et al.*, 2015; Iidzuka *et al.*, 2016). Although there are no other studies using traps for monitoring populations of *C. notaticeps* currently available for comparison, the CPAC16 trap seems to be suitable for this purpose because it allowed the capture of adults in all sampling dates, trapping up to 20 adults per trap in just 1 week during the population peak. Therefore, the trap CPAC16 model can be a viable alternative for monitoring this species in commercial plantations of *jatropha*.

Among the factors that can influence capturing a specific insect species, the color of the trap used can play an important role (Hoback *et al.*, 1999). In studies with *Rhabdoscelus obscurus* (Boisduval) (Coleoptera: Curculionidae), it was observed that the brown color was the most efficient, and all other tested colors (black, red, gray, blue, yellow, white and green) showed no significant differences among themselves in the number of captured adults (Reddy *et al.*, 2011). We did not observe statistically significant differences in the number of captured adults, so any of these colors can be used for monitoring *C. notaticeps*.

In addition to the visual stimulus (color), the olfactory cues are one of the most important factors for locating the host plant. The adults have antennal sensilla that function as olfactory receptors. These receptors are capable of recognizing host plant volatiles allowing the insects to reach their target (Leskey *et al.*, 2001; Prokopy *et al.*, 2001). Several compounds, such as alcohols and esters from the fermentation of plant tissues, have proved attractive to many species of *Curculionidae* (Vacas *et al.*, 2013; Miller *et al.*, 2015; Flechtmann & Atkinson, 2016) such as *Cosmopolites sordidus* (Germar) (Budenberg *et al.*, 1993), *Ceutorhynchus assimilis* Payk. (Smart & Blight, 1997), *Curculio caryae* (Horn) (Collins *et al.*, 1997), *Sitona lineatus* L. (Landon *et al.*, 1997), *Rhynchophorus ferrugineus* (Olivier) (Gunawardena *et al.*, 1998), and *Metamasius hemipterus* L. (Perez *et al.*, 1995).

Molasses has a variable chemical composition (Veana *et al.*, 2014), and studies of its volatile components have revealed the presence of 35 compounds, among which are aliphatic and aromatic esters, aldehydes, alcohols and various derivatives

of furan. The acidic fraction contains at least 11 compounds in which five are simple aliphatic acids (Yokota & Fagerson, 1971). Although it is not certain that volatile compounds are involved, it was observed during the trial, that the fermentation of sugarcane molasses proved to be an efficient lure, capturing higher numbers of *C. notaticeps* adults than ethanol, which is widely used for monitoring *Scolytinae* and *Platypodinae* (Aristizábal *et al.*, 2015; Fernandes *et al.*, 2015), and pineapple, an efficient bait for capturing *R. palmarum* (Duarte *et al.*, 2003). These results support the use of sugarcane molasses for monitoring *C. notaticeps*.

The initial survey of plants with signs of *jatropha* stem-borer infestation showed a very high incidence rate of 66.4%. There was a sharp increase in the period of a year, with incidence reaching 100% of the plants, in a 6-year old *jatropha* plantation. These results indicate that, although the Cerrado presents potential for planting *jatropha*, the cultivation of this crop on a large scale may be impractical due to the occurrence of high populations of *C. notaticeps*, an insect pest with no efficient control recommendations, to this date (Oliveira *et al.*, 2011).

Our results suggest that the flow of *C. notaticeps* adults arriving or moving from attacked plants to healthy ones in the experimental area was continuous, which would hinder management strategies targeting the adult stage. However, since the greatest population incidence of adults occurs between November and January, it is suggested that any management measures focusing on adult control should be adopted during this period, especially in the month of December in order to reduce the pest population density in the cultivated areas.

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