



Interference of plant fixed oils on predation and reproduction of *Neoseiulus baraki* (Acari: Phytoseiidae) feeding on *Aceria guerreronis* (Acari: Eriophyidae)

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ABSTRACT

The coconut mite, *Aceria guerreronis* (Acari: Eriophyidae), is a major pest in coconut plantations in America, Africa and Asia. The management of this pest usually relies on scheduled applications of synthetic pesticides. Recent research demonstrated that degummed soybean and cottonseed oils may potentially control the mite. Although these fixed oils have considerable selectivity for predatory mites, little is known about their sublethal effects on these key natural enemies. This study aimed to assess whether degummed soybean and cottonseed oils modify the functional and numerical responses of *Neoseiulus baraki* (Acari: Phytoseiidae), a predatory mite frequently associated with *A. guerreronis*. The exposure of *N. baraki* to the dosage required to kill 99% of the population (LC₉₉) of the plant oils, estimated for *A. guerreronis*, changed the functional response of the predator from type III to type II. Attack rate and the change in consumption were not influenced by the fixed oils. However, handling time and consumption peak were adversely affected by the exposure of the predator to degummed soybean oil. Additionally, exposure to this oil reduced *N. baraki* female oviposition. Our results suggest that degummed soybean oil can be more detrimental to the control of *A. guerreronis* exerted by *N. baraki*. Although both oils altered the curve of the functional response, the predator could consume high proportions of *A. guerreronis*, mainly at low prey densities.

1. Introduction

The coconut mite *Aceria guerreronis* Keifer (Acari: Eriophyidae) is a serious pest in coconut plantations in America, Asia and Africa (Lawson-Balagbo et al., 2008; Oliveira et al., 2012; Navia et al., 2013). Colonies develop protected beneath the bracts of the perianth, where mites feed on the content of meristematic cells. Mite feeding results in the development of triangular-shaped yellowish-white spots on the fruit epidermis that turn necrotic as the attack progresses (Navia et al., 2013). In severe infestations, fruit can fall prematurely or exhibit distortion, smaller size, lower weight and reduced water and endosperm content (Nair, 2002; Navia et al., 2013). The coconut mite migrates walking from one fruit to another when exposed to food shortage or lack of space (Sabelis and Bruin, 1996; Galvão et al., 2011), when predation risk increases (Lesna et al., 2004) or when the host plant quality declines (Boczeck and Schevchenko, 1996). *Aceria guerreronis*

moves among plants or migrates longer distances via the wind (Galvão et al., 2012).

In Brazil, control of the coconut mite relies on prophylactic acaricidal sprays at monthly or shorter intervals. This treatment potentially leads to environmental and ecological problems such as pollution, pest resistance, pest resurgence and mortality of non-target organisms (Desneux et al., 2007; Geiger et al., 2010; Guedes et al., 2016). Therefore, alternatives to synthetic pesticides are urgently needed to manage this pest. Plant fixed oils, which are non-volatile compounds, are bioactive against a variety of arthropod pests (Bakkali et al., 2008; Isman, 2008; Khater, 2012; Lin et al., 2017). A few were recently demonstrated to hold the potential to control *A. guerreronis* (Oliveira et al., 2017; Teodoro et al., 2017; Freitas et al., 2019). In addition to their toxicity against the coconut mite, degummed soybean oil and cottonseed oil are highly selective for the predatory mites *Neoseiulus baraki* (Athias-Henriot) and *Typhlodromus ornatus* Denmark & Muma

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(Acari: Phytoseiidae; Oliveira et al., 2017; Teodoro et al., 2017). Although these findings support the potential use of these oils for controlling *A. guerreronis*, further ecological studies are needed to assess sublethal effects on natural enemies.

The functional response of a predator to a prey i.e. the intake rate as a function of prey density (Holling, 1959), and the numerical response i.e. the reproduction rate as a function of prey density (Solomon, 1949) are two useful ecological concepts of predator-prey interactions that assess potential biological control agents for pest management purposes (Sandness and McMurtry, 1970; Xiao and Fadamiro, 2010; Fathipour and Maleknia, 2016). Increasing evidence suggests that the functional and numerical responses of predators can be negatively affected by spraying with chemical or natural pesticides (Li et al., 2006; Lima et al., 2015a, 2015b). The present study aimed to determine the effect of degummed soybean and cottonseed oils on the functional and numerical responses of *N. baraki* to *A. guerreronis*. *Neoseiulus baraki* was chosen as model predator because this species is frequently associated with *A. guerreronis* colonies (Aratchige et al., 2007; Lawson-Balagbo et al., 2008; Negloh et al., 2011; Lima et al., 2012). Its morphological features allow individuals to move into the perianth region of the fruit (Lima et al., 2012), its development can be completed by feeding exclusively on the coconut mite (Domingos et al., 2010) and it possesses a high predation capacity (Lima et al., 2012).

2. Materials and methods

2.1. Rearing of *N. baraki*

Adults of the predatory mite *N. baraki* were collected on Green Dwarf coconuts in São Luís city (02°35'03.46"S, 44°12'32.14"W) located in the state of Maranhão, in the Brazilian Northeast. Approximately, 100 females were removed from *A. guerreronis* infested perianths. Mites were transferred onto a 13-cm plastic disc that sat on a water-soaked polyurethane foam disc (1 cm high × 15 cm diameter) placed inside a polyethylene plastic tray (3 cm high × 16 cm diameter). The PVC plastic arena was surrounded by a layer of moistened filter paper and hydrophilic cotton wool to prevent the mites from escaping. Pieces of bract and perianth (3 × 3 cm) infested with ca. 300–400 *A. guerreronis* individuals from different developmental stages and pollen of castor oil (*Ricinus communis* L.) were offered to the predatory mites every other day (Oliveira et al., 2017).

2.2. Functional and numerical responses

The bioassays were conducted under controlled temperature (27 ± 1 °C), relative humidity (70 ± 10%) and photoperiod (12 h light:12 h dark). PVC discs (7 cm diameter) were immersed in a mixture of agar (5%), the fungicide methylparaben (Nipagim®; 0.3%) and distilled water placed on Petri dishes (9 cm diameter, 1.5 cm deep). Once solidified, 5-cm diameter agar discs were removed with the help of a mould in order to expose the PVC discs used as arenas. The discs were surrounded by a layer of moistened cotton wool to prevent mites from escaping.

Cottonseed oil and degummed soybean oil were purchased from ABOISSA (São Paulo, Brazil). A 1.7-mL aliquot at a pressure of 5 psi/pol², which corresponds to a residue of 1.8 ± 0.1 mg/cm², was used to spray the arenas through a Potter spray tower (Burkard, Rickmansworth, UK). The utilised concentration corresponded to the dosage required to kill 99% of the population (LC₉₉) of cottonseed oil (1.60 µL/cm²; Teodoro et al., 2017) and degummed soybean oil (1.39 µL/cm²; Oliveira et al., 2017), previously estimated for *A. guerreronis*. Ten µL of a commercial neutral liquid detergent based on sodium lauryl ether sulphate was added as a coadjuvant. The sprayed discs were left to dry at ambient conditions for 30 min. Subsequently, the predatory mites and prey were transferred into the arenas. Seven-to-eight-day-old *N. baraki* females (at the beginning of their reproductive

period) were individually transferred onto the arenas treated with cottonseed oil, degummed soybean oil or distilled water (as control) that also contained 0.5-cm pieces of perianth with unsexed adult coconut mites at the following densities: 10, 30, 60, 120, 240 and 300 individuals. The bioassay was replicated 15 and 10 times for the lower (10, 30 and 60) and higher (120, 240 and 300) densities, respectively. The number of attacked prey and the number of eggs laid by females were counted 24 and 48 h, respectively, after transferring the predatory mites onto the arenas. After the first observation, the number of prey was replaced with the initial densities. Oviposition data of the first day were excluded from the analysis to minimise the effect of previous food ingestion.

2.3. Statistical analyses

Data analysis for functional responses was conducted in two steps following the protocol of Juliano (2001). First, the shape of the functional response curve for each treatment was estimated with logistic regression analysis of the proportion of the prey consumed as a function of prey density using Proc CATMOD of SAS software (SAS Institute, 2002). The significance and sign of the linear coefficient was used to classify the type of functional response (Holling, 1959, 1961). If it is not significant, the linear coefficient denotes a type I functional response, if it is significant and negative, it indicates a type II functional response and if it is significant and positive, the linear coefficient indicates a type III functional response. In the second step, handling time (*Th*; the time spent killing and ingesting the prey) and attack rate (*a'*; the efficiency in prey searching) were estimated using nonlinear regression with the method of least squares (PROC NLIN SAS; Juliano, 2001).

The consumption peak was calculated for each treatment based on the reciprocal of *Th* (1/*Th*) and compared based on the confidence intervals. The change in prey consumption for each treatment at each density (ΔNa) was calculated according to the following equation: $\Delta Na = [(NaN_{max} - NaN_{min}) / (N_{max} - N_{min})]$, where NaN_{min} and NaN_{max} are the minimum and maximum numbers of prey consumed, respectively, and N_{min} and N_{max} are the minimum and maximum prey densities, respectively (Poletti et al., 2007). The change in prey consumption was further analysed by a one-way analysis of variance (ANOVA) followed by the Tukey test.

Oviposition rates of *N. baraki*, as a function of *A. guerreronis* densities, were submitted to regression analyses using PROC REG of SAS Software (SAS Institute, 2002).

3. Results

The linear coefficients of the logistic regression for predators exposed to the control were positive and significant. These data denoted a type III functional response for *N. baraki* to *A. guerreronis*. In contrast, when predators were exposed to degummed soybean oil or cottonseed oil, the linear coefficients were negative and significant. These results indicated a type II functional response (Table 1).

The functional response curves of the predator to the coconut mite were similar, with a linear trend of increasing prey consumption as density increased and irrespective of treatment. The maximum consumption was observed at 300 prey. At this density, *N. baraki* females consumed on average 175 ± 9 (control), 162.6 ± 9.9 (degummed soybean oil) and 171.2 ± 8.8 (cottonseed oil) prey (Fig. 1). At densities between 10 and 120 individuals, the proportion of prey consumed in the control was close to 1. This proportion decreased for oil-exposed predators. For densities above 120 *A. guerreronis*, there was a greater reduction in the proportion of prey consumed for oil-exposed predators (0.83 ± 0.02 for cottonseed oil and 0.77 ± 0.01 for degummed soybean oil) compared to control predators (0.96 ± 0.01). Nevertheless, even at the highest density, the proportion of prey consumed was greater than 0.5 in all the treatments (Fig. 2).

The *N. baraki* attack rate (*a'*) was not affected by exposure to the

Table 1

Estimated parameters of the logistic regression of the proportion of *Aceria guerreronis* consumed by females of the predatory mite *Neoseiulus baraki*, exposed to distilled water (control), degummed soybean oil and cottonseed oil. The concentrations of the oils correspond to the lethal dosage required to kill 99% of the population (LC₉₉) estimated for *A. guerreronis*.

Treatment	Parameters	Value (± SE)	Df	χ^2	P	Functional response type
Control	Intercept	3.0728 ± 0.3925	1	61.29	< 0.0001	III
	Linear	0.0409 ± 0.00944	1	20.66	< 0.0001	
	Quadratic	-0.00040 ± 0.000057	1	49.07	< 0.0001	
	Cubic	0.0007598 ± 0.00009979	1	57.98	< 0.0001	
Degummed soybean oil	Intercept	5.9081 ± 0.4258	1	192.5	< 0.0001	II
	Linear	-0.0610 ± 0.00778	1	61.59	< 0.0001	
	Quadratic	0.000210 ± 0.000042	1	24.70	< 0.0001	
	Cubic	-0.0000234 ± 0.00007085	1	10.95	< 0.0001	
Cottonseed oil	Intercept	7.2198 ± 0.6037	1	143.0	< 0.0001	II
	Linear	-0.0750 ± 0.0105	1	50.87	< 0.0001	
	Quadratic	0.000269 ± 0.000055	1	23.89	< 0.0001	
	Cubic	-0.0000032 ± 0.00008952	1	12.79	< 0.0001	

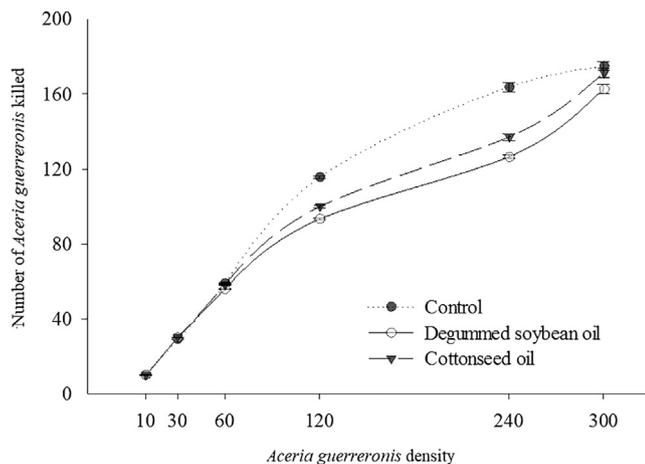


Fig. 1. Number of adult *Aceria guerreronis* individuals consumed by *Neoseiulus baraki* females exposed to degummed soybean oil, cottonseed oil or distilled water (control) as a function of prey density. The concentrations of the oils correspond to the lethal dosage required to kill 99% of the population (LC₉₉) estimated for *Aceria guerreronis*.

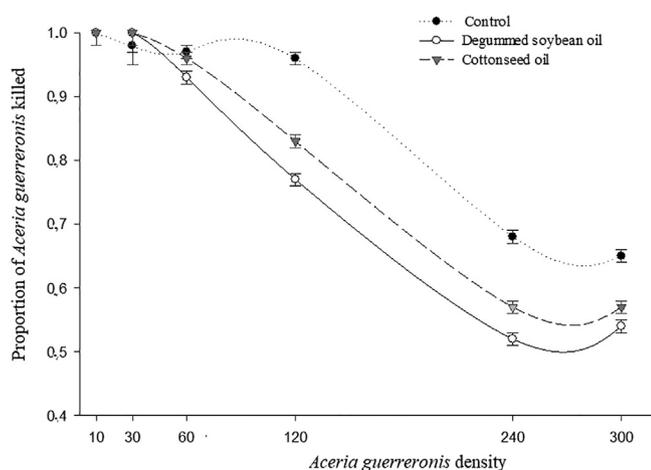


Fig. 2. Proportion of adult *Aceria guerreronis* individuals consumed by *Neoseiulus baraki* females exposed to degummed soybean oil, cottonseed oil or distilled water (control). The concentrations of the oils correspond to the lethal dosage required to kill 99% of the population (LC₉₉) estimated for *Aceria guerreronis*.

oils. In contrast, the predatory mites displayed a greater handling time (T_h) when exposed to the degummed soybean oil compared to

predators in the control treatment. The consumption peak for predators exposed to water was significantly greater (7.57 prey/h) when compared to predators exposed to degummed soybean oil (6.38 prey/h; Table 2). The variation in prey consumption was not significantly modified ($F_{2,27} = 0.45$; $P = 0.64$; Fig. 3).

The number of eggs laid by the predator in relation to prey density adjusted to a quadratic regression model irrespective of the treatment. Oviposition by water-exposed and oil-exposed predators peaked at the density of 120 prey (Fig. 4).

4. Discussion

Our results demonstrated that *N. baraki* females exhibited a type III functional response to *A. guerreronis*. This type of response is characterised by an accelerated rise in prey consumption up to an inflection point, followed by a reduced rate up to a plateau (sigmoidal response; Holling, 1959). Although the type II functional response, which denotes a decelerating prey consumption as prey density increases up to a plateau (predator saturation), is commonly observed in phytoseiid mites (Sabelis, 1985; Poletti et al., 2007; Lima et al., 2015b; Song et al., 2016; Freitas et al., 2018; Mendes et al., 2018), our results are consistent with those obtained by Lima et al. (2012). When *N. baraki* was exposed to either degummed soybean oil or cottonseed oil, the functional response changed from type III to type II. The functional response of phytoseiid predatory mites can be influenced by several factors, including feeding history, temperature, prey species and physical traits of the host plant (Castagnoli and Simoni, 1999; Gotoh et al., 2004; Escudero and Ferragut, 2005; Ahn et al., 2010). Studies that assessed the effect of natural and synthetic compounds on the functional response of arthropod predators demonstrated that the type of functional response may or may not change. For example, the type of functional responses of *T. ornatus* and *N. baraki*, both of which prey on *A. guerreronis*, does not change when exposed to coconut oil (Freitas et al., 2018) and various acaricides (Lima et al., 2015b), respectively. A shift from type II to type III in the functional response of *Scolothrips takahashii* Priesner (Thysanoptera: Thripidae) to *Tetranychus viennensis* (Acari: Tetranychidae) eggs also occurs when the predatory thrips are exposed to abamectin and fenproprathrin (Li et al., 2006). Furthermore, the functional response of *Habrobracon hebetor* (Hymenoptera: Braconidae) to *Ephesthia kuehniella* Zeller (Lepidoptera: Pyralidae) changes from type II to type III when parasitoids are exposed to garlic (*Allium sativum*) and rosemary (*Rosmarinus officinalis*) (Asadi et al., 2018).

The type III functional response displayed by *N. baraki* females in the control treatment was the only response where the proportion of prey mortality increased with prey density, given a constant predator density (Holling, 1959). Predators that exhibit a type III response are considered the most efficient biological control agents (Song et al.,

Table 2

Estimates (means \pm SE) and confidence intervals for attack rate (a'), handling time (Th) and consumption peak ($\frac{1}{Th}$) of unexposed (control) and exposed predatory mite *N. baraki* preying on adults of *Aceria guerrerensis*. Exposed predators had contact with either degummed soybean oil or cottonseed oil. The concentrations of the oils correspond to the lethal dosage required to kill 99% of the population (LC_{99}) estimated for *A. guerrerensis*.

Treatment	$a' \pm$ SE	(95% CI)	$Th \pm$ SE	(95% CI)	Consumption peak	(95% CI)
Control	0.00467 \pm 0.00219a	(0.000321–0.00902)	0.1367 \pm 0.00385a	(0.1290–0.1444)	7.569 \pm 0.034a	(6.925–7.752)
Degummed soybean oil	0.00206 \pm 0.000386a	(0.00130–0.00283)	0.1567 \pm 0.00488b	(0.1470–0.1664)	6.382 \pm 0.031b	(6.009 – 6.802)
Cottonseed oil	0.00237 \pm 0.000481a	(0.00141–0.00332)	0.1469 \pm 0.00438ab	(0.1381–0.1556)	6.807 \pm 0.034ab	(6.427 – 7.241)

SE = Standard error of the mean.

Attack rate a' (ratio of units of preys caught by the predator per unit of searching time), handling time Th (ratio of units for a 24-h exposure period) and consumption peak $1/Th$ (exposure period per handling time). Means followed by the same letter in the column do not differ. Significantly different from control at 5% level when 95% confidence intervals of estimates did not overlap.

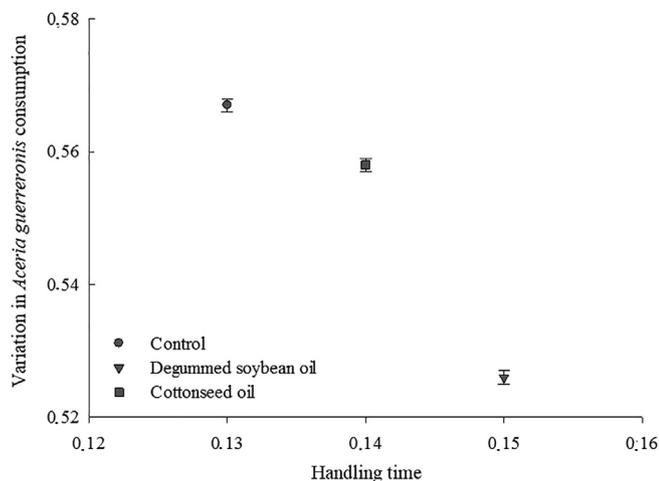


Fig. 3. Consumption variation of *Aceria guerrerensis* (mean \pm standard error of the mean) as a function of *Neoseiulus baraki* handling, exposed to degummed soybean oil, cottonseed oil or distilled water (control). The concentrations of the oils correspond to the lethal dosage required to kill 99% of the population (LC_{99}) estimated for *Aceria guerrerensis*.

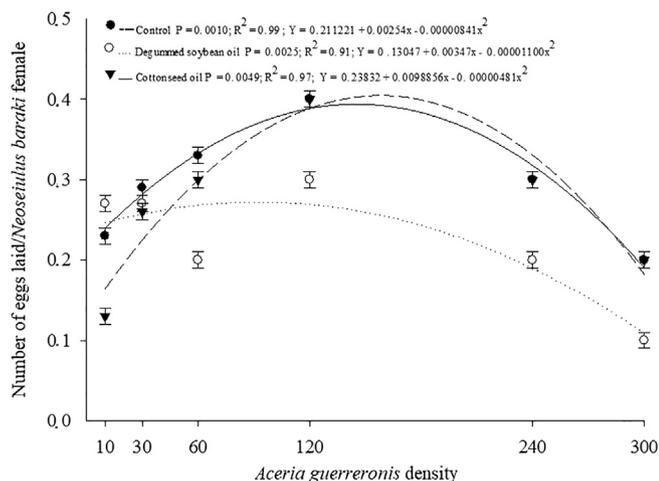


Fig. 4. Mean number of eggs laid by *Neoseiulus baraki* females exposed to degummed soybean oil, cottonseed oil or distilled water (control) as a function of prey density. The concentrations of the oils correspond to the lethal dosage required to kill 99% of the population (LC_{99}) estimated for *Aceria guerrerensis*.

2016). On the other hand, the type II functional response, presented by females in the oil treatments, indicates an increase in prey consumption with prey density. However, the searching rate is not constant, and the proportion of prey consumed declines and tends to stabilise with increasing prey density (Holling, 1959; Pervez, 2005). In this type of response, the predator tends to cause the highest mortality at low prey

densities (Ali et al., 2011; Khanamani et al., 2014), and species that display this type of functional response are also successful predators (Fathipour and Maleknia, 2016). Regulation of *A. guerrerensis* populations occurs when prey density does not overpass the consumption capacity of the predator; that phenomenon can result in a decline in the prey mortality ratio (Mori and Chant, 1966; Sandness and McMurtry, 1970; Mills, 1982). Therefore, our findings suggest that *N. baraki* exhibited a high predation potential when exposed to the fixed oils tested in this study, mainly at low prey densities.

Neither degummed soybean oil nor cottonseed oil altered the *N. baraki* attack rate. Predators exposed to degummed soybean oil, however, exhibited increased handling time and significantly reduced consumption peak. These results may be related to physical or chemical characteristics of this oil perhaps impairing prey identification or capture. Degummed soybean oil and cottonseed oil contain large quantities of linoleic acid (58.65% and 49.61%, respectively), a fatty acid with proven repellent activity against insects (Dani et al., 1996; Nilsson and Bengtsson, 2004). Degummed soybean oil also contains large portions of palmitic acid (16.15%) whereas cottonseed oil presents oleic (23.50%), palmitic (10.04%), linolenic (6.64%) and stearic (4.48%) acids (Oliveira et al., 2017; Teodoro et al., 2017), some of which can modify insect behaviour (Marshall et al., 1979; Mukherjee et al., 2014). For instance, oleic acid can repel the ants *Crematogaster scutellaris*, *Formica cunicularia* and *Lasius* sp. (Insecta: Formicidae; Dani et al., 1996). Additionally, oleic, linolenic and linoleic acids present repellent activities against *Liposcelis bostrychophila* (Psocoptera Liposcelididae; Green, 2011), and palmitic acid can repel *Protaphorura armata* (Collembola; Nilsson and Bengtsson, 2004). Furthermore, there is evidence that cottonseed oil repels *T. ornatus* females after 1 and 24 h exposure (Teodoro et al., 2017).

The exposure to degummed soybean oil reduces the number of eggs laid by *N. baraki* females. This change in the numerical response may stem from the increased handling time and lower consumption observed in individuals exposed to this oil. Alterations in these factors may have reduced protein ingestion. The results suggest that the oil treatment decreased fertility, which, in turn, potentially compromised population growth and negatively affected predator-prey interactions and the biological control of the coconut mite by *N. baraki*.

Taken together, our results suggest that cottonseed oil, and to a lesser extent degummed soybean oil, are compatible with the predatory mite *N. baraki*, a key biological control agent of *A. guerrerensis* in coconut crops.

CRediT authorship contribution statement

Adenir Vieira Teodoro: Conceptualization, Formal analysis, Funding acquisition, Methodology, Project administration, Supervision, Writing - original draft, Writing - review & editing. **Natália Nicolle Furtado Costa de Oliveira:** Investigation, Formal analysis, Writing - original draft. **Andreia Serra Galvão:** Methodology, Formal analysis. **Jose Guedes de Sena Filho:** Writing - original draft, Writing - review &

editing. Delia M. Pinto-Zevallos: Writing - original draft, Writing - review & editing.

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