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Research Article

Population dynamics, vertical distribution and damage characterization of burrower bug in peanut

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ABSTRACT: Although the burrower bug (*Cyrtomenus mirabilis*) is considered a vitalsoil pest in peanut crops, *Arachis hypogaea* L., in South America, there is little information on its occurrence and damage characterization. This study aimed to evaluate the vertical distribution and fluctuation of the burrower bug in the soil and the damage this species causes in peanuts. Two peanut cultivars (IAC OL3 and IAC 503) were evaluated in three locations in the state of São Paulo: Pindorama, Ribeirão Preto and Votuporanga, in the 2017/18 and 2018/19 harvests. Trenches were dug 0.5 m long, 0.3 m wide and 0.3 m deep, along plant lines at four spots on each sampling date, and stratified in layers 10 cm deep. Plant samples (0.5 m) were collected and evaluated for number of pods and percentage of kernels with symptoms of insect damage. More than 85 % of the *C. mirabilis* population was found in depths of up to 10 cm, especially after plant fructification, and an increase in nymphs from 100 days after sowing (DAS) was seen when an increased number of maturing pods was observed. The occurrence of nymphs and adults of *C. mirabilis* and their damage to peanut kernels was similar in both cultivars (IAC OL3 and IAC 503), when these were harvested according to their developmental cycles.

Keywords: *Arachis hypogaea* L., soil pest, kernel damage, pod maturation, monitoring

Introduction

The frequency of peanut (*Arachis hypogaea* L.) kernels with spots varying from white to dark brown has increased in recent years, impairing the quality of the product (Santos et al., 2016). This damage has been attributed to burrower bugs (Hemiptera: Cydnidae), as the Hemiptera insects are known, belonging to the Cydnidae family (Lis et al., 2000; Schwertner and Nardi, 2015). Several species with economic importance in the Neotropical region belong to the *Cyrtomenus* and *Pangaeus* Stål genera. Nymphs and adult individuals feed on roots, tubers (Riis et al., 2005) and soil fruits (Riis et al., 2005; Chapin et al., 2004; 2006). The damage reduces productivity and facilitates infections by soil pathogens, as observed in cassava (*Manihot esculenta* C.) and aflatoxins in peanuts.

The burrower bug, *Cyrtomenus mirabilis* (Perty, 1830), is a polyphagous species widely distributed in the Neotropical region and is considered a pest in several countries on account of the damage caused in several crops, with higher intensity in cassava and peanut (CIAT, 1989; Bellotti et al., 1999; Riis et al., 2005). It is noteworthy that the species *Cyrtomenus bergi* Froschner (1960) was recently defined as a junior synonym of *C. mirabilis* and all information published for the first species must now be considered as *C. mirabilis*.

The first report of *C. mirabilis* causing damage in peanuts in Brazil was in 1960 (Cruz et al., 1962; Calcagnolo and Tella, 1965). The damage caused by this bug in peanuts was attributed to attacks on pods containing developing kernels in which nymphs and adults inserted the stylet of their oral apparatus to reach the developing kernel (Chapin and Thomas, 2003).

C. mirabilis adults are black, with short legs and strong spines that help them move in the soil. Nymphs are dark brown to almost black with white to cream colored abdomens. All immature stages (five) and adults pierce the pods and feed on the kernels. A light attack will cause delimited yellow to brownish dry rot spots (approx. 1.5 mm diameter) and a severe attack can cause a complete decomposition of the harvest (Riis et al. 2005).

However, characterization of its occurrence in the soil profile and damage throughout the development of peanut plants are unknown. This knowledge is essential to the establishment of strategies for monitoring and control. Thus, this study aimed to evaluate the vertical distribution and fluctuation of burrower bug populations in different locations and the damage caused at distinct developmental stages of the pods of two peanut cultivars.

Material and Methods

The experiments were carried out at three locations in the state of São Paulo, Brazil, during the 2017/18 and 2018/19 harvests. In Pindorama (21°13′16.1″ S, 48°55′37.7″ W, altitude 527 m) the study was carried out on an Ultisol Udult Typic, sandy texture; in Votuporanga (20°27′24.3″ S, 50°03′55.5″ W, altitude 525 m), the study was carried out on an Alfisol Udult Eutroferric, sandy texture; while in Ribeirão Preto (21°12′57.7″ S, 47°52′17.4″ W, altitude 526 m) the soil was classified as an Oxisol Udox Eutrophic, clayey texture (Soil Survey Staff, 2014).

At each site, four plots of four sowing lines were sown, with a spacing of 0.9 m width and length of 20 m for cultivars IAC OL3 and IAC 503, arranged alternately. The sowing density was 20 seeds per meter. The seeds had been previously treated with Carboxin + Tiram at a dosage of 3.0 mL kg^{-1} per seed of the commercial product. These locations were chosen because they had a history of the occurrence of peanut kernels with insect damage in previous harvests.

IAC 503 and IAC OL3 cultivars are currently the most planted in Brazil. IAC 503 was the first high oleic runner cultivar in Brazil. However, due to their long cycle duration (145 days), the planting of these cultivars was limited under the sugarcane rotation system. To overcome this limitation, IAC OL3 cultivar was introduced which combines good yield performance with a shorter cropping time (130 days), and desirable technological traits (Godoy et al., 2014).

In the 2017/18 harvest, samplings were initiated when plants had a certain number of pods. Thus, seven samplings were carried out in Pindorama (sowing took place on 30 Oct 2017) at 58, 73, 87, 102, 116, 130 and 135 days after sowing (DAS). In Ribeirão Preto (sowing took place on 17 Nov 2017), five samplings were carried out at 70, 91, 106, 120 and 133 DAS, while in Votuporanga (sowing took place on 01 Dec 2017), five samplings were carried out as well, at 59, 81, 95, 111 and 121 DAS, due to the high incidence of fungal leaf diseases.

In the 2018/19 harvest, samplings started during sowing and ended according to the developmental cycle of each cultivar, i.e., 130 – 135 DAS for IAC OL3 and 145 – 150 DAS for IAC530. Thus, in Pindorama (sowing at 06 Nov 2018) eight samplings were completed at 0, 20, 42, 60, 80, 105, 126 and 133 (IAC OL3) or 148 (IAC 503) DAS. In Ribeirão Preto (sowing on 21 Nov 2017), eight samplings were carried out at 0, 20, 43, 61, 85, 110, 123 and 133 (IAC OL3) or 148 (IAC 503) DAS. Finally, in Votuporanga (sown late on 04 Dec 2018, seven samplings were carried out at 0, 20, 41, 58, 79, 100 and 120 DAS.

At each sampling event, trenches of 0.5 m long × 0.3 m wide × 0.3 m deep were dug along the lines of plants in each plot for each cultivar, following the methodology described in Nascimento et al. (2014). The excavation (removal of soil samples) was carried out manually, with hoes and a metal figure with the same dimensions as the trench. The trenches were stratified in 10 cm layers from the surface down to 30 cm of depth. For the sampling events carried out in Pindorama and Votuporanga, in each layer, the soil was carefully washed in a sieve (2.2 mm mesh) and the burrower bugs were separated and counted. In Ribeirão Preto, the soil was sieved using a 2.2 mm mesh, not washed as a function of the high clay content.

During counting, bugs were categorized into adults and nymphs above 2.5 mm in length, followed by the estimated totaling of nymphs and adults for each square meter of the plots. The identification of species was based on a comparison of morphological characteristics with previously identified specimens based on data in the literature (Avendaño et al., 2017; 2018). A representative sample of the collected specimens was dry assembled and deposited in the Hemiptera Collection of the Museum of Zoology of the University of São Paulo (MZ – USP).

In order to assess the damage caused by the bugs, five plants were manually collected in sequence in the sowing line on the same day the trenches were dug, placed in identified plastic bags and transported to the laboratory in Pindorama. Subsequently, the number of plants and pods per plant were counted.

Pods were removed from the plants, opened manually with a cutting blade and classified according to the degree of maturation, using a methodology adapted from Boote (1982). Next, the green film was removed and the percentage of damaged kernels evaluated according to the pods' degree of maturation.

The results were submitted to the Kruskal-Wallis test ($p < 0.05$), and later a multiple comparison test ($p < 0.05$), taken from the Agricolae package in R (Mendiburu and Simon, 2015; R Development Core Team, 2021), was also applied.

Results

Sampling of burrower bug

As regards the number of nymphs and adults of burrower bug sampled in trenches, no difference was observed between the IAC OL3 and IAC 503 cultivars. Thus, the results presented below refer only to those observed for the IAC OL3 cultivar.

A difference was observed in comparison to the evaluated locations, with the highest occurrence of the burrower bug in Pindorama, intermediate in Ribeirão Preto and the lowest in Votuporanga.

In Pindorama, during the 2017/18 harvest, from 58 to 116 DAS only adults of *C. mirabilis* were sampled. In the following samples, nymphs started to be observed and at 130 and 135 DAS, an increased number of nymphs were collected, reaching 100.0 nymphs m^2 in the 0 – 10 cm layer (Figure 1). Up to 116 DAS, 92.9% of adults were found in the 0 – 10 cm layer. No insects were found in the 21 – 30 cm layer, and in the samplings carried out at 130 and 135 DAS, 73.2 % of the insects were found in the 0 – 10 cm layer (Figure 1).

During the 2018/19 harvest, the evaluations based on the sowing revealed the occurrence of nymphs in

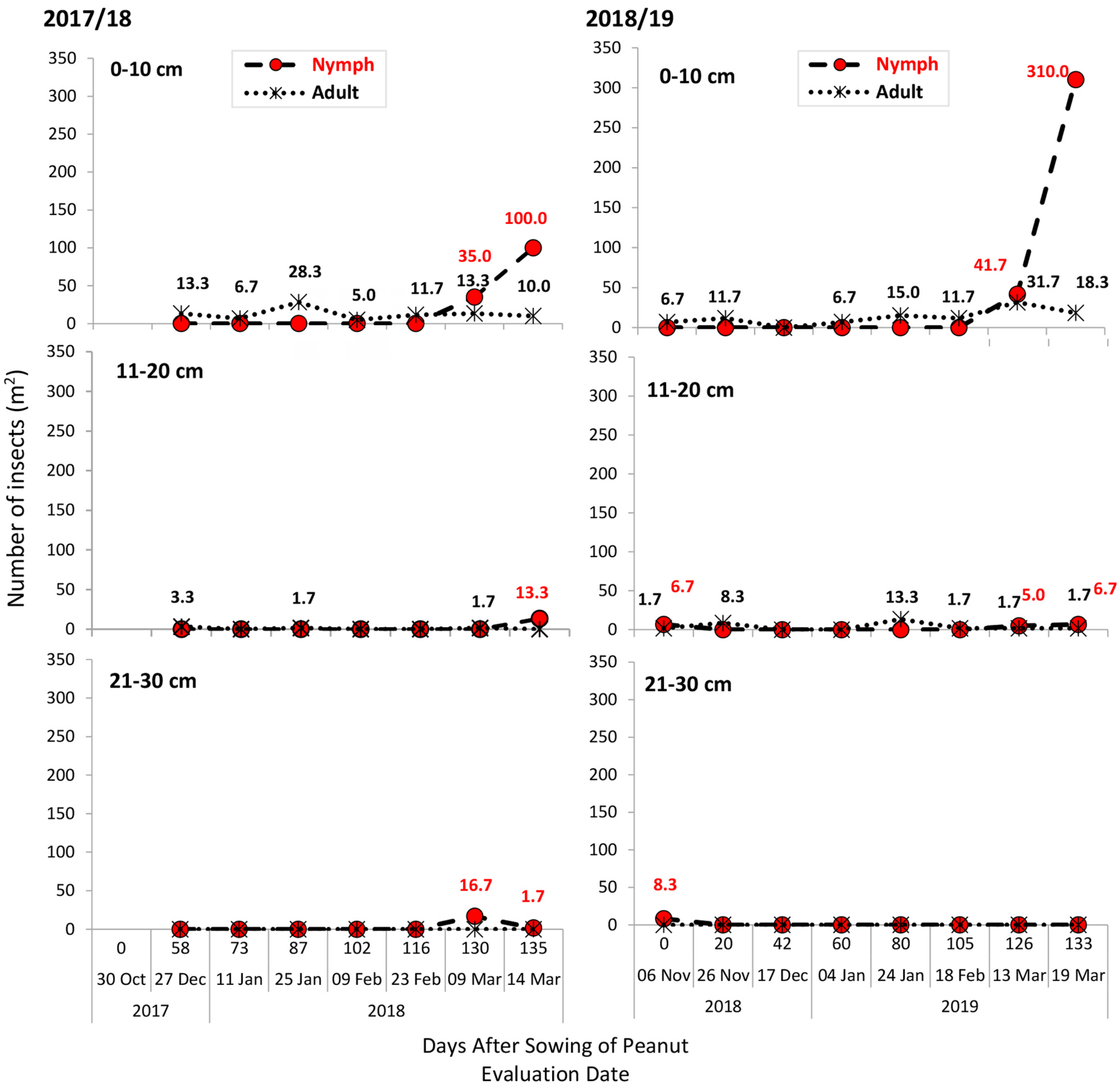

Figure 1 – Average number of nymphs and adults of *Cyrtomenus mirabilis* (m^2) at different soil depths during the development of peanut plants, IAC OL3 cultivar. Pindorama, SP; 2017/18 and 2018/19 harvests.

deeper layers (21 – 30 cm), indicating that in the absence of peanut plants or food (pods), nymphs seek shallower depths (Figure 1). From 40 to 105 DAS, no nymphs were observed at any depth, while at 133 DAS, a population increase occurred, especially in the surface layer (0 – 10 cm), with 94.4 % of the individuals composed of nymphs (Figure 1).

In Ribeirão Preto, during the 2017/18 harvest, only adult specimens were found from 70 to 120 DAS. At 133 DAS, an increased number of adults and mostly nymphs was observed, reaching 235.0 nymphs m^2 in the 0 – 10 cm layer (Figure 2). At 120 DAS, 87.3 % of adults were found in the superficial soil layer, while at 133 DAS, 96 % of the insects were found in the surface layer, as shown in Figure 2.

During the 2018/19 harvest, a low initial infestation was observed, but an increased number of insects was found at 110 DAS on the surface layer and only a few specimens in the deepest soil layers (Figure 2).

As regards the samplings carried out in Votuporanga, the total amount of insects observed in comparison to Pindorama and Ribeirão Preto was lower.

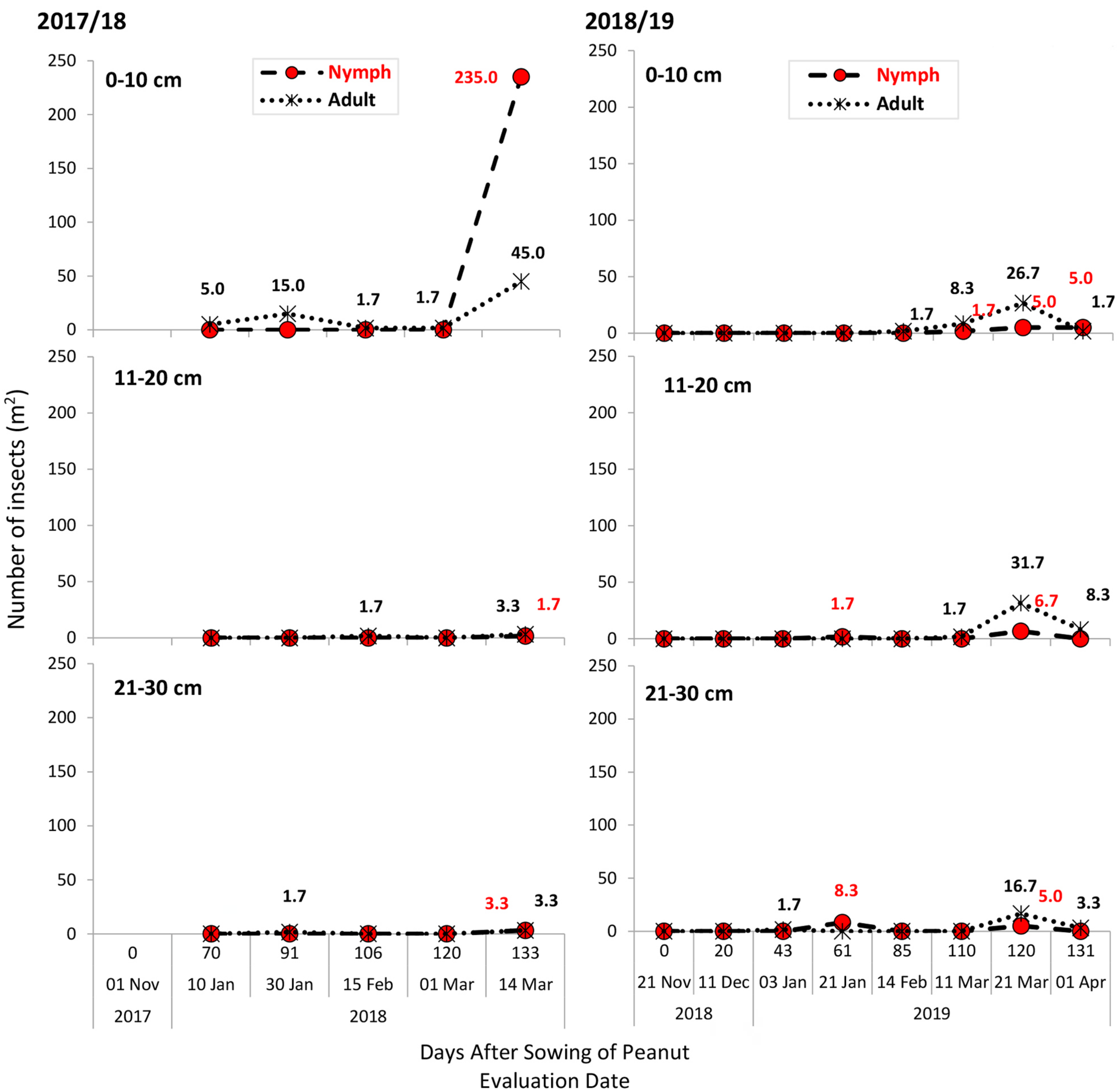

Figure 2 – Average number of nymphs and adults of *Cyrtomenus mirabilis* (m^2) at different soil depths during the development of peanut plants, IAC OL3 cultivar. Ribeirão Preto, SP; 2017/18 and 2018/19 harvests.

During the 2017/18 harvest, from 59 to 111 DAS, only a few individuals were collected from the 0 – 10 cm soil layer. During the 2018/19 harvest, practically no insects were found in the experimental area, with only a few adult specimens in the surface layer (Figure 3).

Quantification of damage caused by the burrower bug

During the 2017/18 harvest, kernel damage was first observed in the samplings carried out 73 DAS in Pindorama. However, from 102 DAS onwards, there was an increased number of observations, reaching 15.2 % at 130 DAS (Figure 4).

The first kernels with visual damage caused by the burrower bug at 73, 78 and 102 DAS were observed in pods at the R6 stage, when these were completely formed and at the R7 stage when the pods had kernels at the beginning of maturation. At 116 and 130 DAS, a higher percentage of damaged kernels was observed in pods at stages R7 and R8, representing together more than 90 % of the damaged kernels (Figure 5).

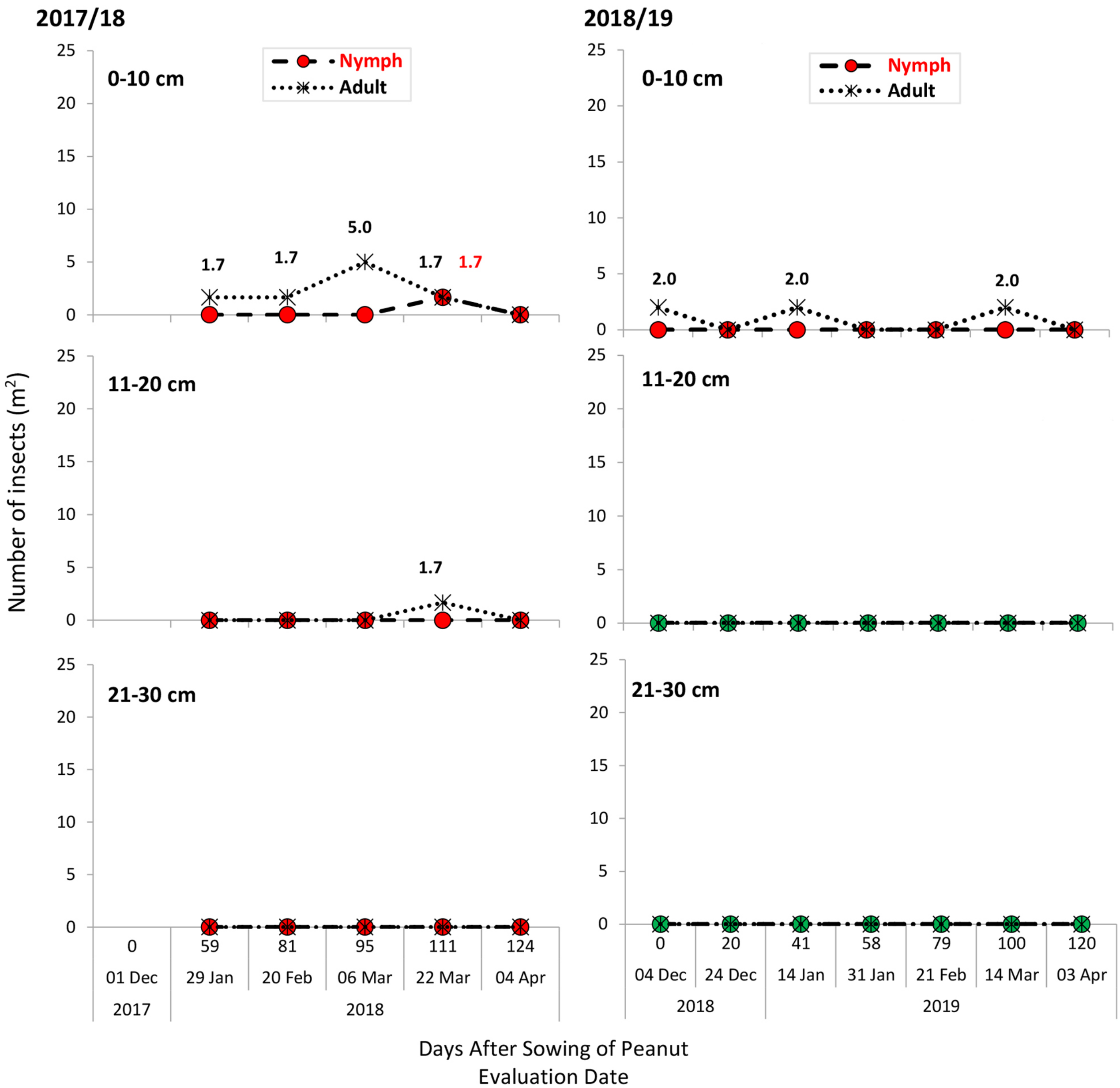

Figure 3 – Average number of nymphs and adults of *Cyrtomenus mirabilis* (m^2) at different soil depths during the development of peanut plants, IAC OL3 cultivar, in Votuporanga, SP; 2017/18 and 2018/19 harvests.

In the 2018/19 harvest, the evaluations of damage in kernels initiated at 20 DAS and ended according to the development cycle of each cultivar, i.e. at 135 DAS for IAC OL3 and 145 DAS for IAC 503. In Pindorama, kernels with visible damages increased from 105 DAS onwards, reaching 27.9 % (Figure 4).

The first damaged kernels belonged to the pods at stages R4, R5 and R6 at 80 DAS. In the evaluations performed at harvest point, damage was observed at all stages of maturation in pods, though with a higher percentage in pods at R6, R7 and R8 (Figure 5).

During the 2017/18 harvest, in the first sampling event carried out in Ribeirão Preto at 70 DAS, a few kernels were found with visual damage (0.6 %). At 133 DAS, the plants presented 39.2 % of their kernels containing some kind of damage (Figure 4).

Damage was initiated in pods at stage R5 (seed formation) in the sampling carried out at 70 DAS. In subsequent evaluations, pods at stages R5, R6, R7 and R8 also presented kernels with symptoms. However, in the final evaluation (133 DAS), a predominance of pods at stages R7 and R8 was observed, which together

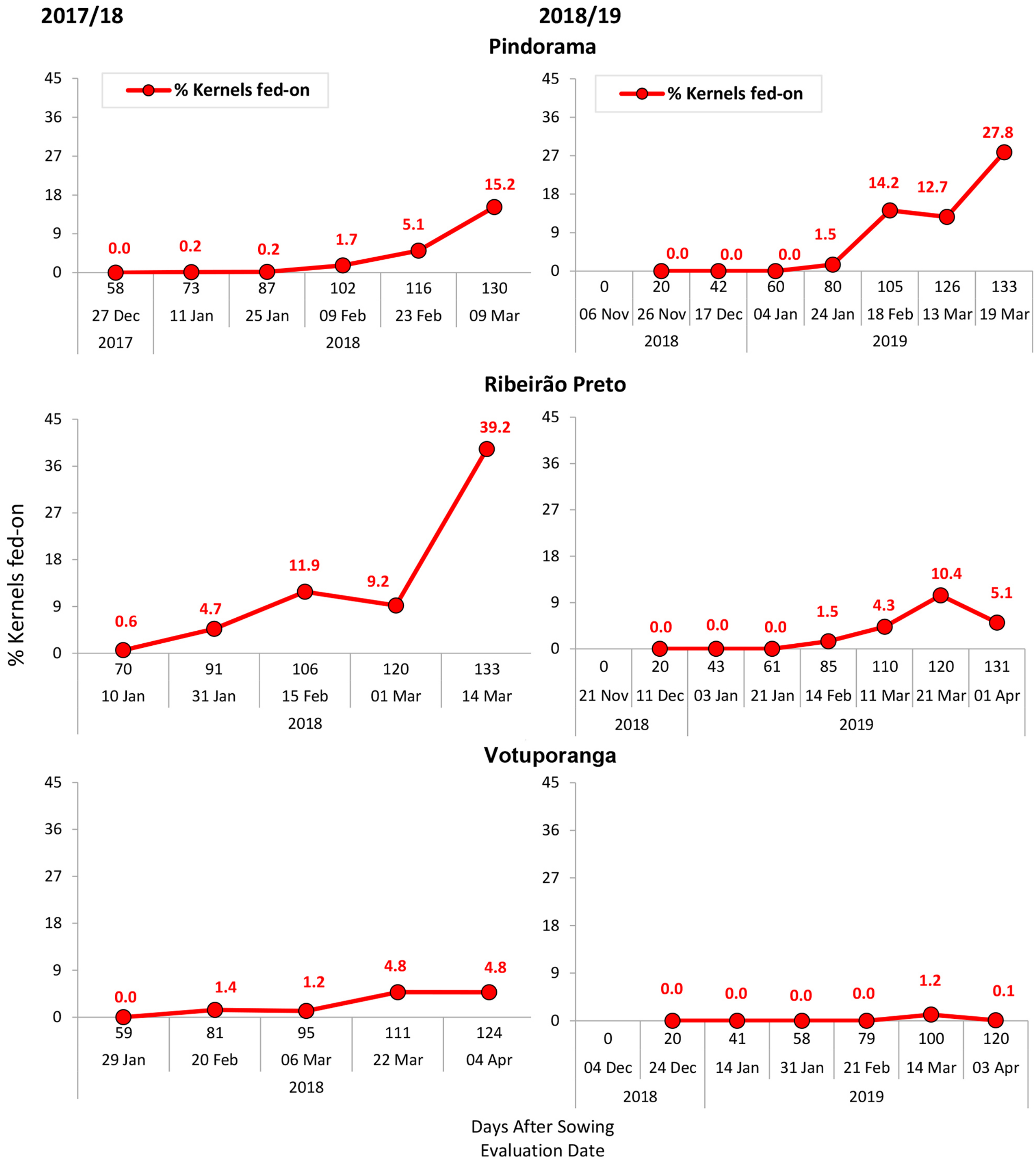

Figure 4 – Kernel feeding (%) by *Cyrtomenus mirabilis* during the development of plants from IAC OL3 peanut cultivars in Pindorama, Ribeirão Preto, and Votuporanga, SP; 2017/18 and 2018/19 harvests.

reached 84.3 % of the kernels having some kind of damage (Figure 5).

During the 2018/19 harvest, in Ribeirão Preto, kernels with visual damage were first observed at 85 DAS. A decrease was observed in the percentage of damaged kernels at harvest point at 133 DAS (5.1 %) (Figure 4). The same pattern of damage in pods, as a function of the degree of maturation, was observed, with damage at all stages and higher percentages at stages R6, R7 and R8 (Figure 6).

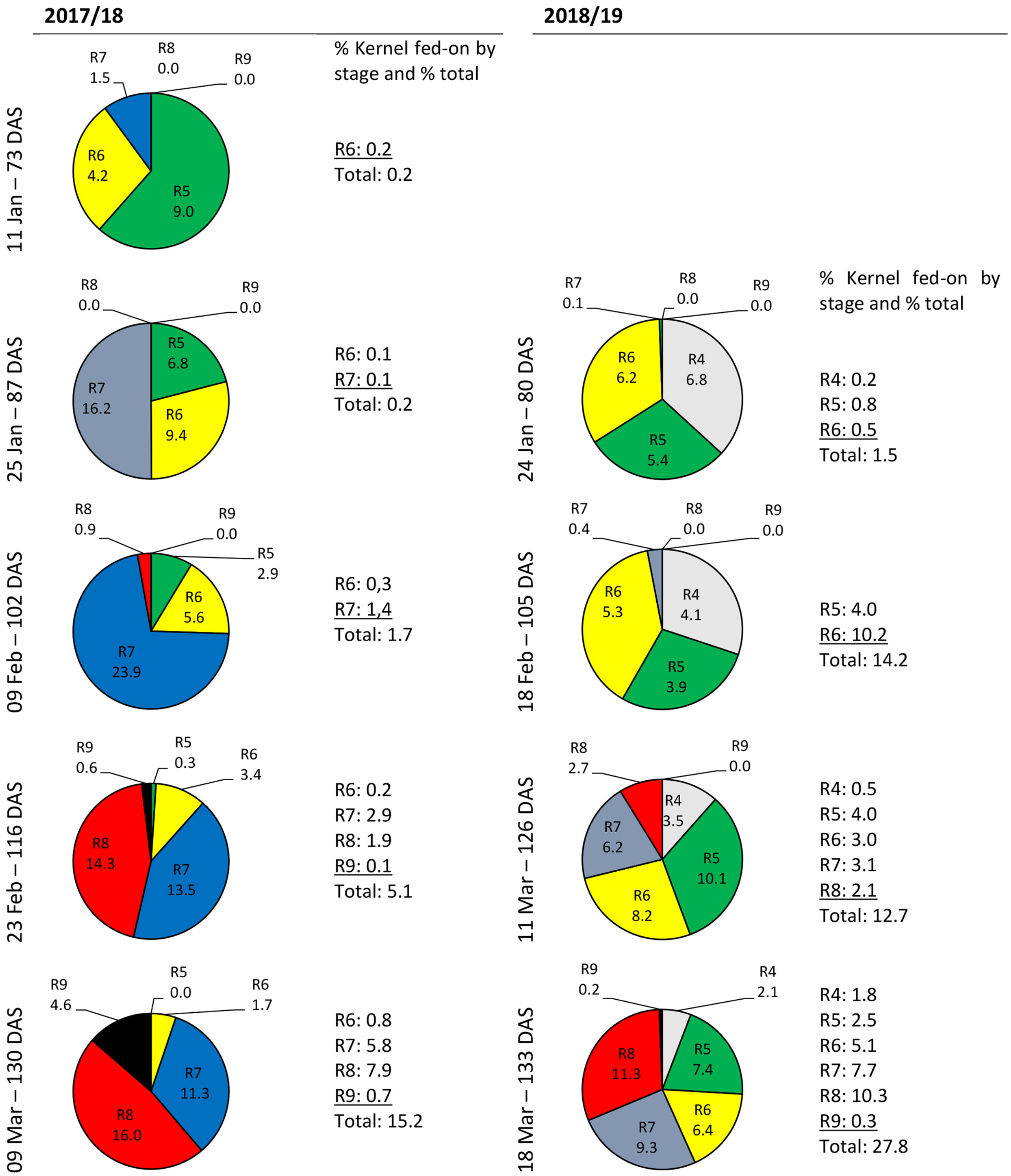

Figure 5 – Average number of reproductive stages by plant and kernel feeding (%) by burrower bug of peanut plants, IAC OL3 cultivar, in Pindorama, SP; R5 = Beginning seed; R6 = Full seed; R7 = Beginning maturity; R8 = Harvest maturity and R9 = Over-mature pod. (Adapted of Boote, 1982). DAS = Days after sowing; 2017/18 and 2018/19 harvests.

In Votuporanga, during the 2017/18 harvest, the damage was first found at 81 DAS and reached 4.8 % of damaged kernels at 124 DAS (Figure 3). Initially, kernels with pod damage at stages R5 and R6 were observed; however, in the evaluations done at 111 and 124 DAS, a higher percentage of kernels with attack

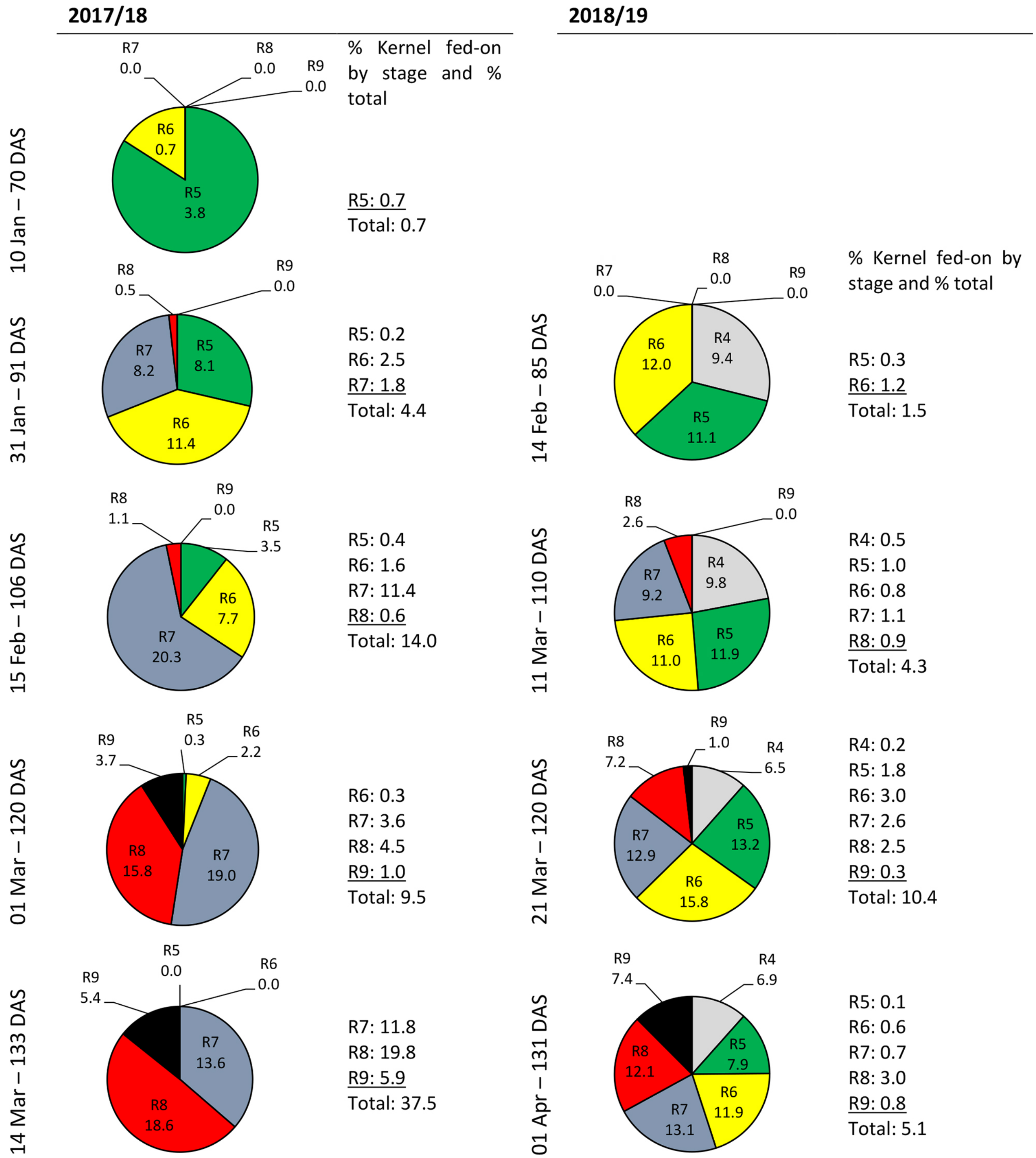

Figure 6 – Average number of reproductive stages by plant and kernel feeding (%) by burrower bug of peanut plants, IAC OL3 cultivar, in Ribeirão Preto, SP; R5 = Beginning seed; R6 = Full seed; R7 = Beginning maturity; R8 = Harvest maturity and R9 = Over-mature pod. (Adapted of Boote, 1982). DAS = Days after sowing; 2017/18 and 2018/19 harvests.

signs by the burrower bug in R8 pods was verified. During the 2018/19 harvest, despite the need to end evaluations prematurely as a result of the high incidence of late leaf spot (*Nothopassalora personata* (Berk. & M.A.Curtis) U.Braun, C.Nakash., Videira & Crous), which is a leaf fungi disease, almost no signs of damaged kernels were observed attributable to the burrower bug.

Discussion

In the samplings of developing plants (harvest 2017/18), nymphs and adults of the burrower bug were primarily found in the surface soil layer, regardless of the cultivar and location. When the samplings were initiated (during the sowing in the 2018/19 harvest), more insects were found in sub-superficial layers for both nymphs and adults, especially in Pindorama (Figure 1).

The movement of this species in the soil is influenced by the soil and air humidity. Higher mortality rates of *C. bergi* were observed in dry soils, but at depths below 10 cm, the insect populations were not affected (Riis and Esbjerg, 1998). These authors also reported that this species presents positive geotaxis when the soil surface is moistened. On the other hand, the relative air humidity stimulates the activity of both adults and nymphs in their final developmental stages above the soil surface (Riis and Esbjerg, 1998). However, in the experiments conducted in the present study, no correlation between accumulated rainfall and the evaluated parameters was found throughout the development cycle of plants, possibly due to the small number of sites evaluated.

Under laboratory conditions, Riis et al. (2005) verified that *C. bergi* did not tolerate extremely dried soils, which reduced the longevity of females and prevented their oviposition. On the other hand, in moist soils close to field capacity, the fecundity of females was augmented. Usually, peanut cultivation in Brazil is carried out between spring and summer, seasons characterized by high rates of rainfall, which in turn might favor this form of proliferation of the pest insect. A practical implication of this movement in relation to soil moisture is the use of entomopathogenic fungi and nematodes that can be contaminated, and those that bury themselves in the soil after reaching the site by immigration (Riis and Esbjerg, 1998). In Colombia, applications of specific strains of *Metarhizium anisopliae* (Metsch.) Sorokin (Deuteromycotina: Hyphomycetes) in association with low dosages of imidacloprid resulted in high mortality of *C. bergi* nymphs, when applied to greenhouse soils (Jaramillo et al., 2005).

Another aspect that can influence the movement of this species is the soil's texture and compaction. Carballo and Saunders (1990) found that as the soil compaction increased, the occurrence of insects in maize crops, *Zea mays* L., decreased, possibly due to the greater difficulty of excavation. In the present study, soil compaction was not evaluated, but the experiments were installed in two soil textures, sandy (Pindorama and Votuporanga) and clayey textures (Ribeirão Preto). Except for Votuporanga, where the occurrence of insects was low in both years, the infestation was similar in both soils in alternate years. This fact does not support the hypothesis that the occurrence of this insect is related to sandy soils of low fertility only (Calcagnolo and Tella, 1965).

The low occurrence of *C. mirabilis* in trenches and consequently the minor damage observed in kernels collected in Votuporanga may have been influenced by the presence of *Crotalaria spectabilis* in the surroundings of the experiment in both years, which was not present at other sites. Castaño et al. (1985) reported that the presence of *Crotalaria* sp. plants in consortium with cassava reduced the damage from 61.2 % (single planting) to 3.7 % (intercropping). It is noteworthy that with light traps, the occurrence of migrating *C. mirabilis* was observed in the area close to the experiment in Votuporanga in 2019 (Michelotto et al., 2019); thus, these bugs were migrating in the area. Nevertheless, specific studies are necessary to prove the repellency and control of this species, and the possibilities of its use in peanut plants.

The soil tillage system also influences the occurrence of this insect species. Chapin et al. (2001) observed a higher occurrence of *Pangaeus bilineatus* (Say, 1825) in peanut plants under no-tillage in maize and wheat (*Triticum aestivum* L.) straw. Chapin and Thomas (2003) reported a higher number of insects captured in soil traps in no-tillage systems under maize straw in comparison to planting over rye straw or conventional planting. A possible explanation of this fact is the species's ability to feed on maize plants while maintaining its population in the area throughout the planting carried out during the autumn.

The presence of pods influences the distribution of the burrower bug along with the soil profile. In this study, from 100 DAS onwards (when a higher frequency of R7 pods, i.e. in maturity), a higher percentage of adults and especially nymphs was found, i.e., where these are located in the pods. According to Chapin and Thomas (2003), in pods at stage R6, higher concentrations of nutrients are available for the insects, and both nymphs and adults can reach the kernels with their mouthparts in developing pods. Michelotto et al. (2019) observed that the capture of adults in soil traps is efficient while there are no mature pods, which happens from 100 DAS onwards.

In cassava, an important characteristic has shown resistance to *C. bergi* in Colombia. Cultivars with high carcinogenic potential, i.e, a high capacity of liberating hydrocyanic acid (HCN) – a highly toxic substance – were strongly neglected compared to cultivars with low carcinogenic potential by nymphs and adults of this bug (Riis et al., 2003a; 2003b). Cassava cultivars with higher levels of cyanogenic compounds presented almost no damage caused by the bug. However, in peanut plants this component is not present and, to date, no chemical compound with this characteristic is known.

Kernel damage was initially observed at approximately 70 DAS in pods at stages R4 and R5, but the damage was greater from 100 DAS, when a higher percentage of pods at the R7 stage (developing pods) were found. Smith Jr. and Pitts (1974) observed adults of *P. bilineatus* in the soil only when a few mature pods existed in peanut plants. In addition, these authors verified kernels damaged by the bug at all stages of maturation, though with greater frequency in mature pods. In our experiments, a positive correlation was observed between kernel damage and the number of nymphs, while no correlation was found between damaged kernels and adults, indicating that the control should focus on nymphs, as these were found in higher numbers.

As for the damage caused in kernels as a function of the cultivars selected for this study, during the 2017/18 harvest (when the last evaluation was performed at 124 and 133 DAS), the highest percentages of kernels with symptoms of attack were found in the IAC OL3 cultivar. However, when the last evaluation was carried out in the crop's cycle in the 2018/19 harvest, this difference was not seen, or it was higher in the IAC 503 cultivar. Unlike what happens in certain other countries, Brazilian farmers are no longer well paid for the lower incidence of damaged kernels caused by bugs, largely due to the absence of an established management plant for its control. In the United States, the application of chlorpyrifos at pegging is recommended to reduce damage in peanut kernels caused by bugs (Chapin and Thomas, 2003). Mbata and Shapiro-Ilan (2013) reported that the combination between chlorpyrifos and the entomatogenic nematode *Heterorhabditis bacteriophora* Poinar was synergistic resulting in higher mortality of *P. bilineatus*, and can thus be considered an option to control this species and other soil bugs.

Conclusions

More than 85 % of the population of the burrower bug, *C. mirabilis* is found in depths up to 10 cm when plants are in the fruiting period. The presence of adults is more common throughout the development cycle of peanut plants, with an increase in nymphs occurring from 100 DAS, when the number of maturing pods is observed, i.e. at stage R7. The occurrence of nymphs and adults of *C. mirabilis* and the damage they inflict on peanut kernels was similar in cultivars IAC OL3 and IAC 503, when these were harvested in accordance with their own development cycle.

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Authors' Contributions

Conceptualization: Michelotto, M.D.; Godoy, I.J. **Design of methodology:** Michelotto, M.D.; Godoy, I.J. **Data acquisition:** Michelotto, M.D.; Godoy, I.J.; Bolonhezi, D.; Freitas, R.S.; Schwertner, C.F. **Data analysis:** Michelotto, M.D.; Santos, J.F.; Godoy, I.J. **Writing and editing:** Michelotto, M.D.; Bolonhezi, D.; Freitas, R.S.; Santos, J.F.; Godoy, I.J.; Schwertner, C.F.

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