LIFE TABLE STUDIES OF *RACHIPLUSIA NU* (GUENÉE) AND *CHRYSODEIXIS* (= *PSEUDOPLUSIA*) *INCLUDENS* (WALKER) (LEPIDOPTERA: NOCTUIDAE) ON ARTIFICIAL DIET

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ABSTRACT

Rachiplusia nu (Guenée) and *Chrysodeixis* (= *Pseudoplusia*) *includens* (Walker) (Lepidoptera: Noctuidae) are 2 economically important species in soybean in northern Argentina. Life cycle, reproductive and population parameters of *R. nu* and *C. includens* reared on artificial diet were determined under controlled environmental conditions. Fecundity increased from d 2 to 3 with values of 67.6 eggs per female for *R. nu* and 75.7 eggs for *C. includens*. An average of 79.9% (*R. nu*) and 74.4% (*C. includen*s) of individuals reached the larval stage and 71.1% (*R. nu*) and 71.4% (*C. includen*s) of individuals reached the adult stage. The life expectancy (*ex*) curve showed 4 and 3 periods of mortality. The survivorship curves of the 2 species resembled the theoretical type I. Life table analysis determined that *R. nu* and *C. includen*s have the potential to quickly increase their populations under controlled rearing conditions. These results provide important information that will be used to improve artificial rearing procedures contributing with biological studies towards to develop management programs of both species.

Key Words: loopers species, soybean pests, life cycle, reproductive and population parameters

Resumen

Rachiplusia nu (Guenée) y *Chrysodeixis* (= *Pseudoplusia*) *includens* (Walker) (Lepidoptera: Noctuidae) son dos especies de importancia económica de la soja en el noroeste argentino. Se determinaron parámetros del ciclo de vida, reproductivos y poblacionales de *R. nu* y *C. includens* criadas en dieta artificial bajo condiciones ambientales controladas. La fecundidad incrementó entre el segundo y tercer día con valores de 67,6 por hembra para *R. nu* y 75,7 huevos para *C. includens*. Un promedio de 71,1% (*R. nu*) y 71,4% (*C. includen*s) de individuos alcanzaron el estado adulto y la curva de expectativa de vida (*ex*) mostró cuatro y tres períodos de mortalidad. La curva de supervivencia de las dos especies fue comparable a la Curva de Tipo I. El análisis de tabla de vida reveló que *R. nu* y *C. includen*s tienen el potencial de incrementar rápidamente su población bajo condiciones controladas de cría. Estos resultados proporcionan información importante que podrá ser utilizada para mejorar los procedimientos de cría artificial contribuyendo con estudios biológicos orientados a desarrollar programas de manejo de ambas especies.

Palabras Clave: especies de oruga, plagas de la soja, ciclo de vida, parámetros poblacionales y reproductivos

Rachiplusia nu (Guenée) and *Chrysodeixis* (= *Pseudoplusia*) *includens* (Walker) usually known as semi-loopers or measuring worms (Eichlin & Cunningham 1978; Lafontaine & Poole 1991) are 2 species within the subfamily Plusiinae (Lepidoptera: Noctuidae) which are economically important pest. Larvae of these species feed on several high value crops including aromatic and oleraceous plants, plus many field and vegetable crops such as sunflower (*Helianthus annus* L.), soybean (*Glycine max* (L.) Merrill), alfalfa (*Medicago sativa* L.), cotton (*Gossypium hirsutum* L.),

bean (*Phaselous vulgaris* L.), flax (*Linum usitatissimum* L.), and tobacco (*Nicotiana tabacum* L.) (Artigas 1994; Betancourt & Scatoni 2006; Pastrana 2004; Pastrana et al. 2004; Pereyra 1995; Pereyra 1998; Sanchez & Pereyra 1995; Navarro et al. 2009).

While *C. includens* is widely distributed in the Western Hemisphere and is one of the most destructive insect pests of soybean in the southern United States (Betancourt & Scatoni 2006; Kitching & Rawlins 1987; Jost & Pitre 2002; Navarro et al. 2009), *R. nu* is a major defoliator restricted to South America (southern Brazil, Paraguay, Bolivia, Argentina, Chile, and Uruguay). During the last few yr, both species have emerged as major soybean pests in Argentina due to high levels of defoliation resulting in loss of photosynthetic area. One individual of *R. nu* can consume up to 100 cm^2 of soybean leaves whereas *C. includens* averages between 80 and 200 cm^2 (Pereyra 1994; Navarro et al. 2009; Casmuz et al. 2009; De Freitas et al. 2011). Given that soybean has become the most important cultivated crop in Argentina, reaching over 18 million ha planted with an average production of 30 million tons per year (Devani et al. 2006), the occurrence of both species inflict significant losses in the region.

Although various aspects of taxonomy, bioecology, crop damage, chemical control, natural enemies, host plants, attractiveness of floral odorant compounds, and oviposition preference for these species have been studied in different countries of America (Ruffinelli 1942; Angulo & Weigert 1975; Eichlin & Cunningham 1978; Rizzo & Saini 1990; Lafontaine & Poole 1991; Portillo et al. 1993; Igarzábal et al. 1994; Pereyra 1995; Luna & Greco 1998; Jost & Pitre 2002; Fichetti 2003; Pastrana 2004; Pastrana et al. 2004; Pogue 2005; Meagher & Landolt 2008; Navarro et al. 2009; Barrionuevo 2011), few studies about several specific parameters of the life cycle, especially in a controlled environment, were made. Life tables are powerful tools for analyzing and understanding the impact of abiotic factors on larval growth and survival on meridic diet, reproduction, and population rate of increase (Sandhu et al. 2010). In order to contribute to future studies regarding management strategies such as insecticide and *Bacillus thuringiensis* (Bt) susceptibility, the objective of this study was to provide information on life cycle, reproductive parameters of *R. nu* and *C. includens* reared on an artificial diet.

Material and Methods

Insect Collections

Adults of *R. nu* and *C. includens* were collected from Dec 2008 to Feb 2009 with a light trap in a commercial soybean field in La Virginia, Tucumán Province, Argentina. Collected adults were placed in wooden cages with metal mesh (30 × 30 cm) and transported to the laboratory. The adults were placed in chambers under controlled conditions (27 ± 2 °C, 70-75% RH, photoperiod 14:10 h L:D) to initiate the experimental colonies. Same insects from each species were deposited as voucher specimens in the collection of Instituto Fundación Miguel Lillo (IFML), Tucumán, Argentina.

Insect Rearing

From adult collected in the commercial soybean field, ten couples of each species were randomly selected to establish the experimental colony for each species. The couples were separately maintained in cylindrical polyethylene-terephthalate oviposition cages (25 cm high and 10 cm diam). For ventilation, the top of each cage was covered with a nylon mesh cloth. Adults were fed a 10% honey solution placed in 5 mL glass containers with a hydrophilic cotton pad. The glass containers were replaced daily. Eggs were collected with a moistened brush and deposited individually in Petri dishes lined with moistened filter paper. First instar larvae were placed individually in Petri dishes containing artificial diet that included bean flour (Grandiet®, Buenos Aires, Argentina), wheat germ (Grandiet®, Buenos Aires, Argentina), brewer's yeast (Calsa®, Tucumán, Argentina), vitamin C (Anedra®, Buenos Aires, Argentina), sorbic acid (Anedra®, Buenos Aires, Argentina), and methylparaben (Todo Droga®, Córdoba, Argentina) (Osores et al. 1982). This diet has largely been used to rear other polyphagous lepidopteran species as *Spodoptera frugiperda* and *Diatraea saccharalis* (Murúa et al. 2003; 2008; Prieto et al. 2008). Diet was replaced every 2 d and Petri dishes were lined with moistened filter paper to prevent desiccation and to allow the formation of the pupal chamber (Shourt & Sparks 1981; Navarro et al. 2009*;* Igarzabal et al. 2011). Pupae were then kept individually in 100 mL glass containers on filter paper moistened daily until adult emergence. After establishing the colony for each species, individuals from the 2nd to the 5th generation were used. Three groups of approximately 160 eggs of each species (total of 513 eggs of *R. nu* and 476 for *C. includens*) were used to analyze life cycle duration and sex proportion. Furthermore, a survival analysis was performed for these individuals. From the adults obtained, a set of 45 females for each species were randomly selected to determine longevity, reproductive and population parameters.

Biological Parameters

Development and survivorship of the different stages (egg, larvae instars, pupa and adult) and the resulting sex ratio were recorded. Larvae

were checked daily to determine the transition (molt) to the next instar by visual observation of the cephalic capsules or mortality. Pupae were sexed according to the methodology described by Angulo & Weigert (1975).

Reproductive Parameters

The oviposition period, total fecundity (number of eggs deposited by a female during her entire life period) and total fertility (percentage of eggs hatching) were estimated for each of the 45 females mentioned before. Each female represented a replicate. One virgin female and one virgin male (less than 24 h old) were paired in the cylindrical oviposition cages described above. Moths were maintained in their cages until the female died. Daily mortality and oviposition were recorded. Eggs collected per female were deposited individually in Petri dishes and total fertility was calculated.

Age-specific female survival (l_x, percentage of females alive at specific age x) and age-specific fecundity (m_x, number of female offspring produced by females at age x) were determined. These parameters were recorded for each d (x) the females were alive.

Life Tables

To construct life tables and to estimate population parameters of *R. nu* and C. *includens*, age-specific female survival (l_x) and age-specific fecundity (m_x) obtained from the 45 females described above were used. The methodology described by Rabinovich (1978), Sedlacek et al. (1986), Carey (1993, 1995) and Bellows & Van Driesche (1999) were used. The net reproductive rate [($R_o = \sum (l_x m_x)$], time interval between generations [$T = \sum (x\, l_x m_x) / \sum (l_x m_x)$], intrinsic rate of increase (r = ln Ro/

T), population doubling time (DT= ln2/r) and finite rate of increase ($\lambda = e^r$) were estimated. Considering that both populations have overlapping generations, time interval between generations (T) was interpreted as the age at which, if all the reproductive effort was concentrated on female, the replacement rate would be the same that with the effort shared among several ages.

The survival analysis was performed following the methodology described by Rabinovich (1978) and Carey (1993, 1995).

Data Analysis

Duration of each stage, reproductive and population parameters between *R. nu* and *C. includens* were compared by a T test ($P < 0.05$). All data were analyzed with InfoStat (2006).

RESULTS

Biological and Reproductive Parameters

The egg stage duration for *R. nu* ranged from 2 to 5 d (3.48 ± 0.02). This species had 6 larval instars and the duration of the larval stage was 25.3 ± 0.11 d. First and sixth instars had the longest duration. Duration of the pupal stage was 9.9 ± 0.06 d, while adult lived for 8.31 ± 0.11 d. Of the 365 individuals that reached the adult stage, 215 were females resulting in a female-biased sex ratio (1.43:1) (Table 1). Considering the duration of different stages, the life cycle (egg to adult) of *R. nu* lasted 39 d.

The oviposition period was 4.9 ± 0.13 d ($N = 45$) and during this period a female laid an average of 433.7 ± 11.67 eggs ($N = 45$), with a range of 184 to 613 eggs. Total fertility was 93.6 ± 0.79% and ranged from 71.92 ± 99.27%.

TABLE 1. AVERAGE DURATION IN DAYS (MEAN ± SE) OF THE EGG, LARVAL (L1 - L6), AND PUPAL STAGES, AND ADULT LONGEVITY AND SURVIVORSHIP (PERCENTAGE) OF *RACHIPLUSIA NU* AND *CHRYSODEIXIS INCLUDENS* REARED WITH ARTIFICIAL DIET AT 27 ± 2 °C, 70-75% RH AND 14:10 H L:D. THREE GROUPS OF APPROXIMATELY 200 INDIVIDUALS WERE USED FOR EACH SPECIES.

Life cycle stages	*Rachiplusia nu*		*Chrysodeixis includens*	
	Duration	Survivorship (%)	Duration	Survivorship (%)
Egg	3.48 ± 0.02 a	100.0	3.73 ± 0.02 b	100.0
L1	6.62 ± 0.06 a	89.7	4.79 ± 0.05 b	85.1
L2	4.52 ± 0.06 a	84.8	3.1 ± 0.05 b	78.9
L3	2.77 ± 0.05 a	81.9	2.6 ± 0.05 b	76.5
L4	2.67 ± 0.03 a	81.7	2.61 ± 0.04 a	75.6
L5	2.8 ± 0.05 a	81.1	3.88 ± 0.06 b	74.6
L6	5.84 ± 0.08 a	81.1	6.33 ± 0.06 b	74.4
Overall larval stage	25.34 ± 0.11 a	79.9	23.3 ± 0.11 b	74.4
Pupa	9.94 ± 0.06 a	78.5	8.59 ± 0.06 b	72.3
Adult**	8.31 ± 0.11 a	71.1	7.47 ± 0.09 b	71.4

Values followed by same letters within a column are not significantly different according to Student's t-test (P > 0.05)
**Longevity was estimated from 45 females for each species.

The egg stage duration for *C. includens* ranged from 3 to 5 d (3.73 ± 0.02). Larval development was completed in 6 instars during a period of 23.3 ± 0.11 d. As with *R. nu,* the first and sixth larval instars had the longest duration. The pupal stage duration was 8.59 ± 0.06 d and longevity was 7.47 ± 0.09 d. Of the 340 individuals that reached the adult stage, 206 were females resulting in a female-biased sex ratio (1.5:1) (Table 1). Taking into account our results, the life cycle (egg to adult) of *C. includens* fed on artificial diet was completed in 43 d.

The oviposition period was 4.5 ± 0.14 d and total fecundity during this period was 318.4 ± 9.88 eggs per female ranging from 155 to 449 eggs per female (*N* = 45). Total fertility was 84.9 ± 2.46% and ranged from 32.9 to 98.83%.

All biological (Table 1) and reproductive parameters varied significantly between *R. nu* and *C. includens* [egg (t-test, t = -7.3; df = 987; P < 0.0001), larva (t-test, t = 13.03; df = 762; P < 0.0001), pupa (t-test, t = 16.29; df = 745; P < 0.0001), longevity (t-test, t = 6.03; df = 88; P = 0.0001), oviposition period (t-test, t = 1.98; df = 88; P = 0.05), total fecundity (t-test, t = 3.36; df = 88; P = 0.001), total fertility (t-test, t = 7.69; df = 88; P < 0.0001)]. Differences were also significant when comparing duration of each larval instar between species. Duration of L1 (t-test, t = 22.6; df = 861; P < 0.0001), L2 (t-test, t = 17.65; df = 810; P < 0.0001) and L3 (t-test, t = 2.54; df = 783; P = 0.01) was higher for *R. nu* than for *C. includens*, while duration of L5 (t-test, t = -14; df = 700; P < 0.0001) and L6 (t-test, t = -5.08; df = 755; P < 0.0001) was higher for *C. includens*. Differences were not significant when duration of the fourth larval instar was compared between species (t-test, t = 1.32; df = 777; P = 0.18).

Life Tables

Population parameters of both species are shown in Table 2. The only parameter that differ between *R. nu* and *C. includens* was " R_o" (t-test, t = 6.3; df = 88; P < 0.0001). No differences were detected when T (t-test, t = -0.1; df = 88; P =0.89), r (t-test, t = 1.55; df = 88; P = 0.12), DT (t-test, t = -1.33; df = 88; P = 0.18) and λ (t-test, t = 1.71; df = 88; P =0.09) were compared between species. Because " R_o" values were greater than 1.0 for both

species, the 2 populations under laboratory conditions increased in size. The number of times that a population of *R. nu* and *C. includens* multiplies per generation was 252.69 and 192.71 respectively, which indicates that a female of these 2 species can produce on average other 253 and 193 new females respectively in each generation (Table 2). For both species, reproductive effort (T) was concentrated in the third d, when females put the most amounts of eggs showing that *R. nu* and *C. includens* continually multiply within the limits of their breeding season.

Age-specific survival (l_x) and age-specific fecundity (m_x) of *R. nu* and *C. includens* females are illustrated in Fig. 1. Fecundity was higher for *R. nu* and increased from d 2 to 3 with values of 66.1 and 77.2 eggs per female, respectively (Fig. 1a). *C. includens* showed one peak of high fecundity from 2 to 3 d with 61.4 eggs per female (Fig. 1b).

Considering the survival analysis, an average of 71.1% of the *R. nu* individuals reached the adult stage (Table 1). The life expectancy (*ex*) curve showed 4 periods of mortality (Fig. 2). The highest one occurred from egg stage to first larval instar; the second and third periods occurred from the first to second instar and from second instar to third instar, respectively, and the fourth one occurred before adult emergence. Similar to *R. nu*, 71.4% of *C. includens* individuals reached the adult stage (Table 1). The life expectancy curve showed 3 periods of high mortality for this species (Fig. 2). The first period was from egg stage to first larval instar, the second period occurred from the first to second instar and the third one was from second to third instar. The survivorship curves of the 2 species (Fig. 3) resemble the theoretical type I (Rabinovich, 1978).

DISCUSSION

This study assessed biological and reproductive parameters of 2 species of polyphagous Plusiinae that have been recognized as serious pests in soybean crops in Argentina. *Rachiplusia nu* and *C. includens* reared on artificial diet did not show similar biological characteristics. Duration of life cycle, longevity, oviposition period, total fecundity, total fertility and R_o varied between species. However, T, r , DT and λ were similar for *R. nu* and *C. includens*.

TABLE 2. POPULATION PARAMETERS OF *RACHIPLUSIA NU* AND *CHRYSODEIXIS INCLUDENS* REARED WITH ARTIFICIAL DIET AT 27 ± 2 °C, 70-75% RH AND 14:10 H L:D.

Species	R_o	T	r	DT	λ
Rachiplusia nu	252.69 ± 7.39 a	3.01 ± 0.1 a	1.93 ± 0.07 a	0.38 ± 0.01 a	7.77 ± 0.67 a
Chrysodeixis includens	192.71 ± 5.99 b	3.03 ± 0.09 a	1.79 ± 0.05 a	0.4 ± 0.01 a	6.44 ± 0.4 a

Values followed by same letters within a column are not significantly different according to Student's t-test (P > 0.05)

R_o: net reproductive rate, T: time interval between generations (days), r: intrinsic rate of increase (females/female/day), DT: population doubling time (days), l: finite rate of increase (days).
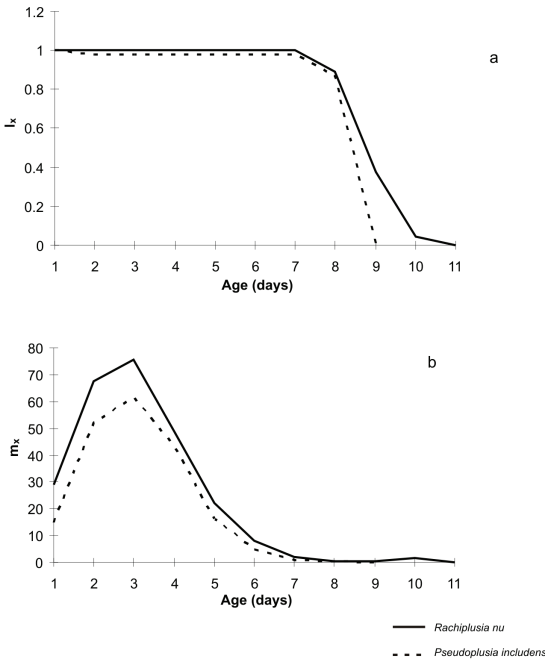
Fig. 1: Survival l_x (a) and fecundity m_x (b) of females of *Rachiplusia nu* (solid line) and *Chrysodeixis includens* (dashed line) reared with artificial diet at 27 ± 2 °C, 70-75% RH and 14:10 h L:D.

Results of our study show that *R. nu* and *C. includens* takes approximately 39 and 43 d, respectively to complete a single generation (from egg to adult) under laboratory conditions. Larval developmental time varied from 23 to 26 d according to the species, and was longer than reported by others (Boldt et al. 1975; Herzog 1980; Shourt & Sparks 1981; Kidd & Orr 2001; Navarro et al.
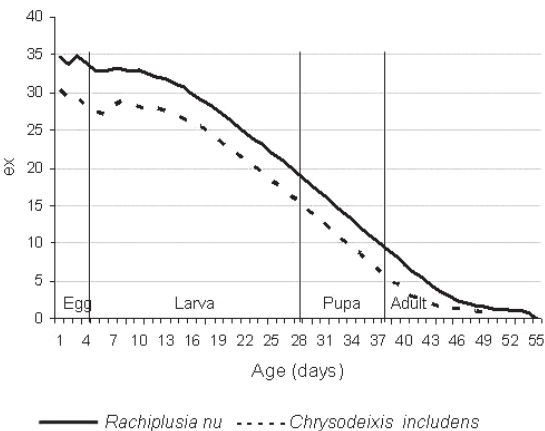

Fig. 2: Life expectancy (*ex*) curves age-specific of *Rachiplusia nu* (solid line) and *Chrysodeixis includens* (dotted line) reared with artificial diet at 27 ± 2 °C, 70-75% RH and 14:10 h L:D.
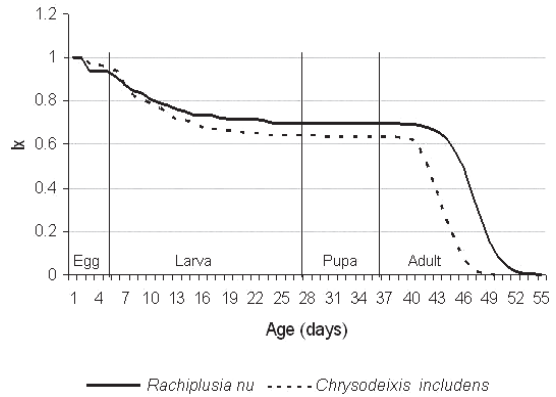

Fig. 3: Age-specific survivorship (*lx*) curves of *Rachiplusia nu* (solid line) and *Chrysodeixis includens* (dashed line) reared with artificial diet at 27 ± 2 °C, 70-75% RH and 14:10 h L:D.

2009). Larval developmental can vary widely depending on rearing conditions (temperature, humidity, etc.), host diet, or host phenology. For example, larvae of other Plusiinae species as *Trichoplusia ni* (Noctuidae) held at 25 °C consumed 23% more leaf area than those held at 20 ºC and 32% more than those held at 30 °C (Boldt et al. 1975). Temperature and humidity have been recognized as important factors affecting life history of lepidopterans (Boldt et al. 1975; Sandhu et al. 2010; Tamiru et al. 2012). Although some studies of the species considered here did not specify the environmental conditions considered (Herzog 1980; Navarro et al. 2009), others were performed under similar temperatures (25-27 °C) but humidity ranged from 40 to 80% (Boldt et al. 1975; Shourt & Sparks 1981; Kidd & Orr, 2001). However, host-plant nutritional value has also been proposed among other factors affecting survivorship of development stages (Pereyra & Sanchez 2006). Ruffinelli (1942) found that when *R. nu* was reared on sunflower, the egg duration period was longer but the larval and pupae periods were shorter compared with individuals of *R. nu* reared on artificial diet. The use of fresh soybean leaves rather than the artificial diet used in this study appears to shorten the larval and pupal periods (Sanchez & Pereyra 1995). Fresh plant material also appears to affect *C. includens* development time, as our results suggest longer larval development times than reported by others (Boldt et al. 1975; Herzog 1980; Shourt & Sparks 1981; Kidd & Orr, 2001; Navarro et al. 2009). Supernumerary larval stages for *C. includens* have been reported depending on diet used (Navarro et al. 2009) and represent a disadvantage for experimental colonies reared in the laboratory because extra larval stages tend to prolong the life cycle. The artificial diet evaluated here, which includes bean flour, wheat germ and brewer's yeast, has been widely

used to rear other polyphagous lepidopterans like *Spodoptera frugiperda* (Noctuidae) and *Diatraea saccharalis* (Crambidae) (Murúa et al. 2003, 2008; Prieto et al. 2008). However, some critical nutrients may be lacking for a better performance of *R. nu* and *C. includens* since many authors reported the importance of nutritional value of the diet for the performance of lepidopteran species. Therefore, the identification of critical nutrients for incorporation into the diet to shorten the life cycle may be the next important step.

Adults of semi-loopers had an average survival time of 7-8 d with maximum progeny production of females occurring from d 2 to 3. Longevity of *R. nu* and *C. includens* were similar to those reported by Pereyra (1994) and Jensen et al. (1974) under laboratory conditions. However, no records of adult longevity are known for these species under field conditions. Adult longevity and resulting reproductive qualities may vary according to the nutritional quality of the larval diet and the adult food. Pereyra (1994) found that soybean leaf age (vegetative or reproductive stage) affected female longevity and the oviposition period of *R. nu*. Jensen et al. (1974), mentioned that different sources of adult food (10% honey solution, 10% sugar solution or nectar of cotton blossoms) affected longevity, mating and the oviposition period of *C. includens*. Female longevity and the sex ratio for *R. nu* were similar but, the oviposition period and fecundity differed from those reported by others (Popich 1992; Pereyra 1994; Sanchez & Pereyra 1995; Kidd & Orr 2001; Navarro et al. 2009). Also Navarro et al. (2009) reported higher fecundity of *C. includens* than recorded in our study. Different laboratory conditions where such studies were conducted (temperature or humidity) may explain the differences with our results.

Life tables are an important tool to describe the growth potential of a species, and our results suggest that both *R. nu* and *C. includens* have a high capacity to increase populations under laboratory. These 2 species compared here show, in general, similar population characteristics. For *R. nu*, Sanchez & Pereyra (1995) reported a higher net reproductive rate but lower values for the rate of population increase, and the finite rate of increase. For all the variables registered, the different environmental conditions may satisfactorily explain the differences in results. Life table parameters are usually affected by temperature and it's been shown with other lepidopteran species that as the temperature rises, so does the rate of population increase until a maximum temperature is achieved (Chi 1988; Murúa & Virla 2004; Sandhu et al. 2010). Also, rearing temperature affects the number of female offspring produced (Sandhu et al. 2010).

The life table analysis determined that *R. nu* and *C. includens* have the potential to quickly increase their populations under controlled rearing conditions. The high proportion of individuals surviving to the adult stage with a high fecundity and a short population doubling time give *R. nu* and *C. includens* a considerable biotic potential. Results of this study suggest that overlapping stages added to the extended oviposition period, would confound the precise prediction of seasonal occurrence of life stages, optimal sample scheduling and timing of control measures in natural populations of both species.

The life expectancy curves (*ex*) indicated the critical ages of mortality. Individuals of *R. nu* and *C. includens* reared on artificial diet revealed higher mortality at early stages (egg and first instar larvae); however mortality was lower than found by Sanchez & Pereyra (1995). These authors reported that mortality ranged between 25 and 58% in larvae and between 33 and 69% in adults of *R. nu*. Mortality in the egg stage was lower than 10% and no mortality was found in the pupal stage. For *C. includens*, Ashfaq et al. (2001) reported that the mortality during the larval stage was significantly affected by the length of time that first and third instars, but not fifth instar, fed on Bt-cotton. On the other hand, survival to pupation of *C. includens* larvae obtained in this study (72.3%) was intermediate to that reported by Kidd & Orr (2001) who found survival to range between 61.5 and 94.5%. The survivorship curve (*lx*) represents the proportion of living individuals at a certain age in relation to the initial number of individuals. The Theoretical type I curve was also described for *R. nu* reared on fresh soybean leaves (Sanchez & Pereyra 1995).

This study is the first record of rearing *R. nu* and *C. includens* on an artificial diet. The use of artificial diet for mass rearing is practical because it provides the easiest and most consistent food source, and eliminates most problems involved with the production and maintenance of host plants. Because of the agricultural importance of *R. nu* and *C. includens* and their simultaneous presence in soybean fields causing crop damage, the maintenance of both colonies under laboratory conditions and following the same procedures, is imperative to achieve mass rearing of these pests. Establishment of experimental colonies following the rearing procedure described here may facilitate different biological studies as well as help to develop new management programs. In these sense, improved rearing procedures will help in diet incorporation studies for host plant resistance and assessment of transgenic genotypes.

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