

Incidence of *Spodoptera litura* (Lepidoptera: Noctuidae) and its feeding potential on various citrus (Sapindales: Rutaceae) cultivars in the Sargodha Region of Pakistan

Muhammad Irfan Ullah¹, Muhammad Arshad¹, Muhammad Afzal¹, Samina Khalid², Muqadas Saleem¹, Irfan Mustafa³, Yasir Iftikhar⁴, Jaime Molina-Ochoa^{5,6,*}, and John E. Foster⁶

Abstract

Spodoptera litura F. (Lepidoptera: Noctuidae) adversely affects important crops such as cotton, okra, tomato, potato, and pumpkin. For the first time, this species was observed in a citrus nursery in the Sargodha Region of Pakistan. The principal objectives of this study were to determine the effects of several citrus (Sapindales: Rutaceae) cultivars, including *Citrus reticulata* Blanco (Feutrell's Early, Seedless Kinnow, and Fairchild mandarin orange) and *Citrus paradisi* Macfad. (grapefruit), on feeding by *S. litura*, and the effects of these plants on the growth of the insect. Based on performance of 3rd instars, Feutrell's Early and Seedless Kinnow supported maximum relative growth rates, high efficiencies of conversion of ingested food, and high levels of leaf consumption and larval weight gain, relative to the other citrus cultivars. Values of these parameters were lowest on grapefruit. Thus, the mandarin oranges Feutrell's Early and Seedless Kinnow were more susceptible to damage than grapefruit.

Key Words: feeding; damage potential; relative growth rate; efficiency of conversion of ingested food; nutritional ecology

Resumen

Spodoptera litura F. (Lepidoptera: Noctuidae) es una plaga muy importante que ataca adversamente a muchos cultivos importantes tales como algodón, okra, jitomate, papa, y calabaza. Se reporta por primera vez a *S. litura* atacando en un vivero de cítricos (Sapindales: Rutaceae) en la región de Sargodha, Pakistán. Los objetivos principales de este estudio fueron determinar los efectos del potencial alimenticio de *S. litura* sobre diferentes variedades de cítricos incluyendo a *Citrus reticulata* Blanco (Feutrell's Early, Seedless Kinnow, Fairchild) and *Citrus paradisi* Macfad. (toronja). El potencial alimenticio de larvas de tercer estadio sugieren que Feutrell's Early y Seedless Kinnow soportan mayor tasa de crecimiento relativo (TCR) así como los valores más altos de eficiencia de conversión de alimento ingerido (ECI), y consumo de **área** foliar entre las variedades de cítricos. Los valores más bajos de estos parámetros fueron observados en la variedad de toronja. Los resultados indicaron que Feutrell's Early y Seedless Kinnow fueron las variedades más nutritivas y susceptibles entre las variedades cítricos ya que mostraron los valores más grandes de TCR, ECI, ganancia de peso larvario, consume de **área** foliar y pérdida de peso de follaje; por el contrario, en la variedad de toronja se corroboró como la menos nutritiva para *S. litura*, ya que produjo los valores más bajos de TCR and peso de excretas (PE).

Palabras Clave: alimentación; potencial de daño; tasa de crecimiento relativo; eficiencia de conversión de alimento ingerido; ecología nutricional

Citrus production is a significant component in the economy of Pakistan. Pakistan ranks 10th in the production of citrus in the world. Citrus crops cover an area of over 210,000 ha, with annual production of 2,237,000 tons (GOP 2013). The climatic conditions of Pakistan are appropriate for citrus cultivation (Syed 2007). Sargodha is the leading city for citrus production in Pakistan (Ashraf et al. 2014).

Unfortunately, citrus is threatened by a number of insect pests in Pakistan. The major pests of citrus in Pakistan include the Asian citrus psyllid, *Diaphorina citri* Kuwayama (Hemiptera: Liviidae); the fruit flies *Bactrocera zonata* (Saunders) and *B. dorsalis* (Hendel) (Diptera: Teph-

ritidae); the citrus blackfly, *Aleurocanthus woglumi* Ashby (Hemiptera: Aleyrodidae); the citrus leaf miner, *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillariidae); and the giant mealybug *Drosicha stebbingii* (Stebbing) (Hemiptera: Margarodidae) (Mahmood et al. 2014).

Spodoptera litura F. (Lepidoptera: Noctuidae) affects numerous crops including cotton, tomato, okra, potato, chili, cucumber, pumpkin, cabbage, pigeon pea, and gram. A wide host range is one potentially important evolutionary strategy to ensure survival (Lee et al. 2003). Generally, polyphagous insect pests like *S. litura* feed on multiple plant species and on various parts of these plants (Suomela 1996). Host se-

¹Department of Entomology, University of Sargodha, Sargodha 40100, Pakistan

²Department of Environmental Sciences, COMSATS Institute of Information Technology, Vehari, Pakistan

³Department of Zoology, University of Sargodha, Sargodha 40100, Pakistan

⁴Department of Plant Pathology, University of Sargodha, Sargodha 40100, Pakistan

⁵Universidad de Colima, Coordinación General de Investigación Científica, Centro Universitario de Investigación y Desarrollo Agropecuario, Facultad de Medicina Veterinaria y Zootecnia, Crucero de Tecmán, autopista Colima-Manzanillo, Km. 40, Tecmán, Colima 28930, México

⁶Department of Entomology, University of Nebraska-Lincoln, Lincoln, Nebraska 68583, USA

*Corresponding author; E-mail: jmolina@uclm.mx

lection may be associated with the presence of secondary metabolites found in these plants, but they also display variation in nutritional value for the insects (Simpson et al. 2002; Lee et al. 2003).

Very few studies have evaluated the influence of various host plant species or cultivars on the growth and development of *S. litura*. The principal objective of this study was to assess feeding by *S. litura* on various citrus cultivars. *Spodoptera litura* is a potentially serious threat to citrus crops, inflicting not only immediate damage but also damage to the subsequent crop because defoliation effects often carry over into the next season.

Materials and Methods

INSECT CULTURE

Spodoptera litura was initially observed in a citrus nursery of the Department of Horticulture, University College of Agriculture, University of Sargodha, Pakistan. Larvae of *S. litura* were collected from the mandarin orange cultivar Seedless Kinnow and brought to the Department of Entomology, University of Sargodha, Sargodha, Pakistan. The larvae were reared on artificial diet made of chickpea flour (Seth & Sharma 2002). The adults of *S. litura* were provided with a cotton boll saturated with 30% honey solution. The mature 3rd instars from the 1st laboratory-reared generation were used for further experiments.

CITRUS CULTIVARS

To evaluate the feeding potential of *S. litura* on various citrus (Sapindales: Rutaceae) cultivars, we selected the 3 cultivars Seedless Kinnow, Fairchild, and Feutrell's Early of *Citrus reticulata* Blanco (mandarin oranges) and 1 cultivar of *Citrus paradisi* Macfad. (grapefruit).

LEAF AREA CONSUMPTION

Third instars were individually placed in Petri dishes (9.0 × 1.5 cm). We used healthy, tender leaves, which were taken from the upper third of the plants. Leaves from the 4 host plants were collected daily and washed with distilled water. In each treatment, 5 leaves were placed in separate Petri dishes and 1 leaf of each cultivar served as a control (it was not offered to larvae). The experiment was replicated 3 times. The leaves were replaced after 2 d. The leaf area was measured with a leaf area meter (LI-COR model LI-3000, Lincoln, Nebraska, USA). Consumption of leaves by *S. litura* larvae was determined by subtracting the remaining leaf area from the initial leaf area after correcting by using the value obtained in the control.

LEAF WEIGHT LOSS

We placed 3rd instar larvae on a weighed leaf within a plastic Petri dish to test larval feeding on the 4 citrus cultivars. The ends of the petioles were wrapped in moistened cotton to prevent desiccation. After 24 and 48 h, leaves were weighed and feces were removed from the leaves and weighed. Petri dishes were cleaned, and new weighed leaves were supplied. This process was continued every second day for each replication. Temperature and humidity were 25 ± 1 °C and 65 ± 5%, respectively, during the experiment. Daily food consumption per larva was estimated by subtracting the weight of the remaining leaf tissue from the weight of the initial leaf provided.

RELATIVE CONSUMPTION RATE (RCR)

The relative consumption rate (RCR) was measured by the following formula (Mehrkhou 2013):

$$RCR = I / B \times T$$

Where *I* is the dry weight of food (mg) consumed, *T* is the duration of feeding period (d), and *B* is the insect dry weight gain (mg).

RELATIVE GROWTH RATE (RGR)

The relative growth rate (RGR) was calculated according to the following formula (Barrania 2013):

$$RGR = [\Delta B / BI] \times T$$

Where ΔB is the change in body weight of the insect (mg), *BI* is the initial larval weight, and *T* is the duration of the feeding period (d).

EFFICIENCY OF CONVERSION OF INGESTED FOOD (ECI)

The efficiency of conversion of ingested food (ECI) was calculated according to the following formula (Suwarno et al. 2010):

$$ECI = B / I \times 100$$

Where *B* is the insect dry weight gain (mg) and *I* is the dry weight of food (mg) consumed.

DATA ANALYSIS

Statistical analysis was conducted with a 2-factor analysis of variance (ANOVA) using the statistical software Statistix 8.1. If significant differences were detected, then means were compared using Tukey's honestly significant difference (HSD) test at $\alpha = 0.05$.

Results

The results of the feeding and performance analyses of *S. litura* 3rd instars are provided in Table 1. Feeding and performance parameters differed significantly among citrus cultivars, except for RCR. The maximum consumption of leaf area was observed on the orange cultivars Feutrell's Early (9.23 cm²) and Seedless Kinnow (4.71 cm²). Leaf area consumption was significantly lower on the Fairchild cultivar and the grapefruit, 3.30 and 2.70 cm², respectively (Fig. 1A). The maximum larval weight gain (larval biomass) was associated with Feutrell's Early (97.93 mg) and Seedless Kinnow (68.16 mg) cultivars, whereas the lowest weight gain was attained on grapefruit (7.02 mg) (Fig. 1E). The greatest production of feces was by larvae feeding on Feutrell's Early (22.80 mg) and Seedless Kinnow (15.84 mg) cultivars. The lowest value of feces production was by larvae feeding on grapefruit (2.60 mg) (Fig. 1F). The highest rate of relative growth (RGR) was observed on Seedless Kinnow (2.26 mg/mg/d), and the lowest was recorded on grapefruit (0.20 mg/mg/d) (Fig. 1B). In the case of relative consumption rate (RCR), the maximum value was observed on grapefruit (11.83 mg/mg/d) and the lowest was recorded on Seedless Kinnow (4.05 mg/

Table 1. Feeding and performance parameters of *Spodoptera litura* 3rd instars on 4 citrus cultivars as assessed by ANOVA.

Parameter	df	F	P
Leaf area consumption (cm ²)	3	29.55	<0.0001
Leaf weight loss (mg)	3	67.20	<0.0001
Larval weight (mg)	3	106.18	<0.0001
Feces weight (mg)	3	89.28	<0.0001
RGR (mg/mg/d)	3	26.57	<0.0001
RCR (mg/mg/d)	3	0.42	0.7377
ECI (%)	3	29.25	<0.0001

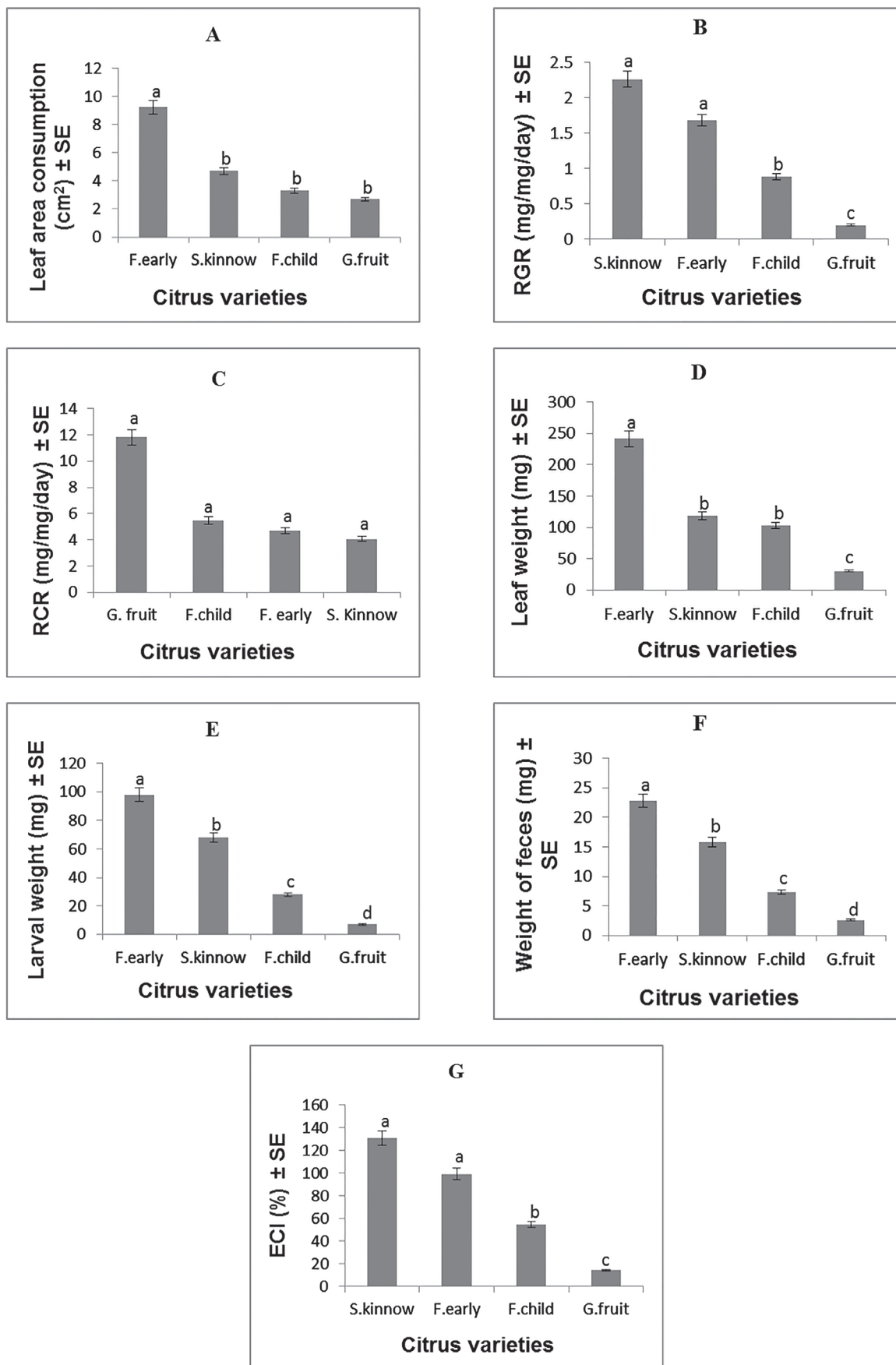


Fig. 1. Food consumption and performance of *Spodoptera litura* 3rd instars on 4 citrus cultivars: (A) leaf area consumption (cm²); (B) relative growth rate (RGR); (C) relative consumption rate (RCR); (D) leaf weight consumed (mg); (E) larval weight (mg); (F) weight of feces produced (mg); (G) efficiency of conversion of ingested food (ECI).

mg/d) (Fig. 1C). The efficiency of conversion of ingested food (ECI) by larvae of *S. litura* was highest on Seedless Kinnow (130.75%) and on Feutrell's Early (99.26%). The minimum ECI value was observed on grapefruit (14.47%) (Fig. 1G).

Discussion

Efficiency of conversion of ingested food (ECI) and relative growth rate (RGR) are the most important parameters related to feeding potential (Reese 1978). ECI and RGR measure the suitability of food, specifically the efficiency with which food is transformed into insect biomass (Waldbauer 1968), which assures the growth and development of insects (Nathan et al. 2005). In this study, significant differences were observed among the ECI and RGR values when *S. litura* was reared on various citrus cultivars. The highest values of ECI and RGR were related to feeding on Seedless Kinnow and Feutrell's Early cultivars of citrus, suggesting that the insects feeding on these hosts were most effective at the conversion of ingested food to biomass and larval growth. As can be seen in Fig. 1B and G, the maximum utilization of leaves was observed on Feutrell's Early and Seedless Kinnow orange.

This research showed that the Feutrell's Early and the Seedless Kinnow cultivars were fed upon more readily, and were more suitable for growth of *S. litura*, than the Fairchild orange and the grapefruit. Previous research also documented that larval development of *S. litura* varied greatly among various host plants (Zhu et al. 2000; Chen et al. 2002; Seema et al. 2004). As noted above, variation in consumption and growth might be due to either the variability of nutritional quality and quantity of the host plant or the presence of allelochemicals (Bernays & Chapman 1994).

Although *S. litura* damages numerous crops in the Sargodha Region of Pakistan, we believe this to be the first documented occurrence of this species affecting commercial citrus in a nursery. Although citrus proved to be susceptible to *S. litura*, we identified significant differences in susceptibility (amount of leaf consumption) and suitability (RGR and ECI) among 4 cultivars of citrus.

Acknowledgment

We are thankful to the biological control laboratory for providing us space and necessary support for the completion of this research.

References Cited

- Ashraf S, Khan GA, Ali S, Iftikhar M, Mehmood N. 2014. Managing insect pests and diseases of citrus: on farm analysis from Pakistan. *Pakistan Journal of Phytopathology* 26: 301–307.
- Barrania AA. 2013. Antifeedant, growth inhibitory and toxicity effects of chlorantraniliprole, thiamethoxam and novaluron against the cotton leaf worm, *Spodoptera littoralis* (Boisd.) (Lepidoptera: Noctuidae) in cotton fields. *Egyptian Journal of Agricultural Research* 91: 903–911.
- Bernays EA, Chapman RF. 1994. *Host-Plant Selection by Phytophagous Insects*. Chapman and Hall, New York, New York.
- Chen QJ, Yang JQ, Zhang JZ, Zhang YZ, Chen JH. 2002. Effect of temperature on laboratory population of *Spodoptera litura* (Fabricius) in tobacco fields. *Tobacco Science and Technology* 2: 42–45.
- GOP (Government of Pakistan). 2013. *Pakistan Statistical Year Book*. Federal Bureau of Statistics, Statistics Division, Islamabad, Pakistan.
- Lee KP, Raubenheimer D, Behmer ST, Simpson SJ. 2003. A correlation between macronutrient balancing and insect host-plant range: evidence from the specialist caterpillar *Spodoptera exempta* (Walker). *Journal of Insect Physiology* 49: 1161–1171.
- Mahmood R, Rehman A, Ahmad M. 2014. Prospects of biological control of citrus insect pests in Pakistan. *Journal of Agricultural Research* 52: 229–244.
- Mehrkhou F. 2013. Effect of soyabean varieties on nutritional indices of beet armyworm *Spodoptera exigua* (Lepidoptera: Noctuidae). *African Journal of Agricultural Research* 8: 1528–1533.
- Nathan SS, Chung PG, Murugan K. 2005. Effect of biopesticides applied separately or together on nutritional indices of the rice leaf folder, *Cnaphalocrocis medinalis*. *Phytoparasitica* 33: 187–195.
- Reese JC. 1978. Chronic effects of plant allelochemicals on insect nutritional physiology. *Entomologia Experimentalis et Applicata* 24: 625–626.
- Seema R, Goel BB, Gupta GP. 2004. Effects of temperature on the development and reproduction of *Spodoptera litura*. *Annals of Plant Protection Science* 12: 205–206.
- Seth RK, Sharma VP. 2002. Growth, development, reproductive competence and adult behavior of *Spodoptera litura* (Lepidoptera: Noctuidae) reared on different diets, pp. 15–28 In Bloem S, Carpenter JE, Hendrichs J [eds.], *Evaluation of Lepidoptera Population Suppression by Radiation Induced Sterility*. IAEA-TEC-DOC-1283, April 2002. Vienna, Austria.
- Simpson SJ, Raubenheimer D, Behmer ST, Whitworth A, Wright GA. 2002. A comparison of nutritional regulation in solitary and gregarious phase nymphs of the desert locust, *Schistocerca gregaria*. *Journal of Experimental Biology* 205: 121–129.
- Suomela J. 1996. Within-tree variability of mountain birch leaves causes variation in performance for *Epirrita autumnata* larvae. *Vegetation* 127: 77–83.
- Suwarno, Salmah MRC, Ali A, Hassan AA. 2010. Oviposition preference and nutritional indices of *Papilio polytes* L. (Papilionidae) larvae on four rutaceous (Sapindales: Rutaceae) host plants. *Journal of the Lepidopterists Society* 64: 203–210.
- Syed R. 2007. Searching new avenues to enhance citrus fruit export. *Daily Times Pakistan*, May 5, 2007.
- Waldbauer GP. 1968. The consumption and utilization of food by insects. *Advances in Insect Physiology* 5: 229–288.
- Zhu SD, Lu ZQ, Chen LF, Yu W, Zhang SJ. 2000. Effect of temperature and food on *Spodoptera litura* population. *Chinese Journal of Applied Ecology* 11: 111–114.