ISSN: 1984-5529 Científica Jaboticabal v.44, n.3, p.326–332, 2016

http://dx.doi.org/10.15361/1984-5529.2016v44n3p326-332

Occurrence of thrips in lisianthus cultivation at different protected crop conditions

Ocorrência de tripes na cultura do lisianto em diferentes condições de cultivo protegido

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Recebido em: 03-09-2015; Aceito em: 28-01-2016

Abstract

Damages caused by thrips (Thysanoptera) are a constant concern in greenhouse. In lisianthus, *Eustoma grandiflorum* (Raf.) Shinners (Gentianaceae), it may cause direct and indirect damage. Thrips can affect flowers aesthetics through feeding on their petals. This study aimed to recording the thrips species associated with lisianthus under protected cultivation, assessing differences in species composition among lisianthus cultivars and among different growing conditions. The experiment was conducted in commercial greenhouses in the "Floricultura Florist" company, municipality of Dois Irmãos, RS state. The experiment was conducted from January to February 2013, comparing four cultivars with different colors (Marina, Orange Flash, Tu 668 and Ace White) and two greenhouses with structural differences. Each flower bed (25mx1m) containing one cultivar (treatment) was divided into 5 plots in which 10 plants were randomly selected and evaluated from the opening of the first floral bud until the cut of the flowers. The most common thrips species were *Frankliniella schultzei* and *F. occidentalis*. The different growing conditions influenced the amount and composition of thrips species collected. There was variation in the average number of thrips gathered from cultivated varieties. This paper presents the first record of thrips species on *E. grandiflorum* in greenhouse of Brazil.

Additional keywords: cut flower, *Eustoma grandiflorum,* thysanoptera.

Resumo

Os danos causados por tripes (Thysanoptera) são uma preocupação constante em cultivo protegido. Em lisianto, *Eustoma grandiflorum* (Raf.) Shinners (Gentianaceae), podem provocar danos diretos e indiretos, e prejudicar a estética das flores ao se alimentarem de suas pétalas. Este estudo objetivou registrar as espécies de tripes associadas ao lisianto sob cultivo protegido e avaliar diferenças na composição das espécies entre cultivares de lisianto e entre diferentes condições de cultivo. O experimento foi conduzido em cultivo comercial na empresa "Floricultura Florist", no município de Dois Irmãos-RS. As coletas ocorreram entre janeiro e fevereiro de 2013, em quatro cultivares com colorações distintas (Marina, Orange Flash, Tu 668 e Ace White) e em duas diferentes áreas de cultivos protegidos com distintas características estruturais. Foram realizadas amostragens semanais, durante o período de 24 de janeiro a 07 de fevereiro de 2013. Cada canteiro (25mx1m) contendo uma cultivar (tratamento) foi subdividido em 5 parcelas, nas quais 10 plantas foram sorteadas ao acaso e avaliadas desde a abertura dos botões até o corte das flores. As espécies de tripes mais frequentes foram *Frankliniella schultzei* e *F. occidentalis*. As diferentes condições de cultivo influenciaram na quantidade e na composição de espécies de tripes coletadas, e foi observada variação no número médio de tripes coletados entre as cultivares estudadas. Este artigo apresenta o primeiro registro de espécies de tripes em cultivo protegido de *E. grandiflorum* no Brasil.

Palavras-chave adicionais: *Eustoma grandiflorum;* flor de corte; thysanoptera.

Introduction

Thrips (Insecta: Thysanoptera) are among phytophagous species that cause more damages to ornamental plants (Monteiro et al., 1999). They are important vectors of virus (tospoviruses) for ornamental plants (Campos-Farinha, 2006) and since some species are polyphagous, it enhances the spread of this organism and extends disease transmission risk to plants (Monteiro et al., 2001). The most characteristic thrip damages are observed by the presence of white and silver spots on leaf surface that can injure

photosynthetic capacity and, moreover, aesthetic damages must be considered in floriculture commercial activities (Inoue et al., 2010).

Lisianthus, *Eustoma grandiflorum* (Raf.) Shinners (Gentianaceae), arouses the interest of producers and consumers due to its button durability, diversity and rare colors, and, flowers offer all over the year (Camargo et al., 2004). However, difficulties in growing lisianthus are evident. They were recently introduced in national market and for this reason there are little scientific and technical information on production and management of this crop (Backes et al., 2008). Furthermore, information concerned to pests associated to these plants is rare. Regarding the insects of Thysanoptera order, there is the record of some Terebrantia species related to lisianthus crop in Israel (Kritzman et al., 2000). However, no studies were found associating these two organisms in Brazil.

Thus, this work aimed to record thrips species in lisianthus crop in Rio Grande do Sul, Brazil, assessing the occurrence of thrips in protected environments with different structural and cropping features and comparing species densities in different plant cultivars.

Material and methods

Experiments were conducted on "Floricultura Florist" company, Dois Irmãos, State of Rio Grande do Sul (RS) (29º34' S; 51º05' W; 166 m altitude). Samples were collected from January 24 to February 7, 2013, including the opening of the first flower buds until the flower harvest. In summer conditions of Rio Grande do Sul, lisianthus cycle presents fast development and flowering period from two to three weeks (personal information of Sr. Yuuki Ban, owner), when the greatest damages by pests occur. When the market demand is great, harvest is performed with two weeks, there being full opening of two flowers per plant, which occurred in this work.

Two areas (A1 and A2) were used to record the occurrence of thrips in lisianthus crop and evaluate the composition in different environments of protected cultivation. A1 area had galvanized steel structures with arch type roof and 4.5m in height, tunnels longitudinally placed in the north-south direction covered by transparent plastic sheet of low density polyethylene (LDPE) 100 microns thick. In the lateral of greenhouse have a side curtains for the management of air circulation and shade cloth (50% shading). It was built in 2012 with a total area of 2180 m². It was only used for lisianthus cultivars of the colors white, pink, purple / white and orange.

Area A2 had 9000 m^2 total area and structure of wood covered with 100 and 150 microns thick LDPE with 3m in height. The lateral of greenhouse don't have side curtains from LDPE, only shade cloth (18% shading). It was built in 2005 and had cultivated Baby's breath (*Gypsophila paniculata* L.), goldenrod (*Solidago canadensis* L.) and snapdragon (*Antirrhinum majus* L.) besides of lisianthus crop in different development stages.

Lisianthus seedlings were transplanted into beds with approximately 60 days after germination when they had four true leaves at a density of 74 plants m^{-2} in 25 m-length beds and one meter width. Each bed had only one variety of lisianthus.

A bed with white flowers of Ace White cultivar (medium–sized semi double flower, late maturity, and heat sensitive) was selected in each of the areas for insects' collection. In area A1, beyond the cultivar described above, three more lisianthus cultivars were selected with different colors in order to collect thrips: Tu 668 (medium size double flower and pink color, medium to late cycle, tolerant to heat); Marina (larde size double flower and purple / white color, medium to late cycle, sensitive to heat); and Orange Flash (medium size double flower and orange color, medium to late cycle, tolerant to heat).

The experimental design had two treatments (about environments assessment) and four treatments (about cultivars assessment) with five plots and five sampling occasions (repetitions). Each 25 m bed was split into five plots of 5 m x 1 m. On each sampling occasion, 10 plants were randomly selected in each plot. Samples were performed on 01-24-2013, 01-28-2013, 01-31-2-2013, 02-05-2013, and 02-07-2013, period that included the opening of the first flowers until harvest on 02-08-2013.

A white container (18 cm x 25 cm x 6 cm) was used to collect insects by perform beat method on all the flowers and buds of each selected plant. Fallen thrips inside the container were placed in plastic tubes (2 mL) with 70% ethanol. After preparing microscope slides, specimens were identified and deposited in a reference collection at the Biological Control Laboratory of UFRGS (CBLab).

Two evaluations were carried out: comparison of thrips number collected on the same cultivar (Ace White) in two different environments (A1 and A2 areas) and comparison of thrips number collected in A1 cropping condition in different lisianthus cultivars.

Data analysis was performed by differences in the variations of the average number of collected insects per plant in each cropping condition each treatment. Data normality was assessed by Lilliefors test, and as data are nonparametric, means were compared using the Kruskal-Wallis test followed by Dunn using Biostat 5.3 program (Ayres et al., 2011).

For constancy analysis, species were classified as constant (W) when present in more than 50% of samples; accessory (y) when present at 25-50% of samples and accidental (z) when present in less than 25% of samples (Bodenheimer, 1955). Dominance (D) of species was defined according to Friebe (1983) [D% = (*i*/*t*)x100, where *i* is the total individuals of species and *t* is the total of collected individuals] according to the categories: eudominant: (EUD)>10%; dominant: 5% < (DOM) ≤ 10%; subdominant: 2% < (SUB) ≤ 5%; recessive: 1% < (REC) ≤ 2%; rare: (RAR) ≤ 1%.

Results and discussions

Total of thrips species recorded and associated to lisianthus plants

Four species of Thripidae family and one Phlaeothripidae specie were identified (Table 1), both families are previously registered in Brazil (Monteiro & Lima, 2011). The Thripidae family is diverse and cosmopolitan and includes most of the pest species found. Generally, species live in flowers or feed on leaves, although including some predators (Mound & Marullo, 1996). *Haplothrips* is one of the Phlaeothripidae genera and can also be associated to flowers (Mound & Zapater, 2003). Since collecting method did not favour thrips capture in immature stage, only a few unidentified immatures were collected. Observation of adults and immatures presence at same plant suggests that some of the collected species might be using lisianthus cultivars as host.

Frankliniella schultzei (Trybom) (Thysanoptera: Thripidae) species had the highest number of individuals collected (268), higher frequency (87.58%) and was classified as constant and eudominant. *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae), even with a lower number of individuals collected (27), was present in all samples (constant) with a frequency of 8.82%. *Thrips tabaci* (Lindeman) (Thysanoptera: Thripidae), *T. australis* (Bagnall) (Thysanoptera: Thripidae) and *Haplothrips gowdeyi* (Franklin) (Thysanoptera: Phlaeothripidae) species had low number of individuals collected and were classified as accessory.

It is important to note that species found in flowers do not indicate, necessarily, that lisianthus is their host (Mound, 2013). For the author, the simple presence of species in plant may be related to dispersal behaviour of species and many of them can land on a wide range of substrates, without, however, feeding or cause actual damage to them.

Table 1 - Family, species, relative frequency (F), constancy (C) and dominance (D) of thrips species collected in cutting lisianthus crop in greenhouse from January to February 2013 in Dois Irmãos, RS.

Family	Species	F (%)	C	D
Thripidae	*Frankliniella schultzei*	87.58	w	EUD
	Frankliniella occidentalis	8.82	w	DOM
	Thrips tabaci	1.31	y	REC
	Thrips australis	0.65	y	RAR
Phlaeothripidae	*Haplothrips gowdeyi*	1.63	y	REC
Total		100		

F: Relative frequency (%). C (constancy): w (constant), y (accessory). D (dominance): EUD (eudominant) > 10%; 5% < DOM (dominant) ≤ 10%; 2% < SUB (subdominant) ≤ 5 %; 1% < REC (recessive) ≤ 2%; RAR (rare) ≤ 1%.

This work is the first record of *F. schultzei* in lisianthus, in Brazil. This specie, attacks plants belonging to 35 families and 83 species (Milne & Walter, 2000). In Brazil, it was recorded in ornamental plants, vegetables, oleaginous and tobacco (Pavan et al., 1993; Monteiro & Lima, 2011). The first record of this specie in Brazil was made in 1944 in Rio Grande do Sul and it is currently described in association to 25 plant species in several states (Lima, 2015).

The second species with highest frequency, *F. occidentalis*, is recorded attacking a large number of species of ornamental plants due to its high reproductive and dispersal ability, being able to explore ephemeral resources such as cut flowers (Demirozer et al., 2012).

In recent work, Manners et al. (2013) describe the importance of these organisms in chrysanthemums, roses and gerberas crops, associating them to different plant stages and propose its biological control using predators such as *Mallada signata* (Schneider) (Neuroptera: Chrysopidae) and *Neoseiulus cucumeris* (Oudemans) (Acari: Phytoseiidae).

Thrips tabaci can infest plants belonging to 25 families, being considered as important pest on to onions, garlic, ornamental plants, micropropagation herbs, cotton, tomato, tobacco and wheat (Mound, 2007).

The presence of these three species referred as virus transmitters in our study indicates the risk of virus transmission to lisianthus crop, considering that 14 thrips species are tospoviruses vectors wordlwide (Riley et al., 2011). In addition, Srinivasan et al. (2012) concluded that *T. tabaci* and *F. fusca* thrip species are vectors of Iris Yellow Spot Virus (IYSV) (*Bunyaviridae* family, *Tospovirus* genus) (mechanical inoculation) using the *E. russellianum* lisianthus species as indicator plant. Kritzman et al. (2000) indicated the presence of this virus in lisianthus grown in Israel. According to the authors, this phytovirus causes "Lisianthus leaf necrosis", whose symptoms are systemic necrosis, necrotic and curled spots in lisianthus leaves. Doi et al. (2003) observed the same symptoms in lisianthus grown in protected environment of Shizuoka and Saga provinces in Japan. The authors reported the virus transmission by mechanical inoculation of *T. tabaci* and excluded transmission by *F. schultzei*. Assessing virus population dynamics in Japan, Fuji et al. (2015)

suggest that the incidence of IYSV in lisianthus was triggered by invasion of infected *T. tabaci* individuals that have acquired the virus from nearby onion fields. Thus, although the disease has not been detected in the cropping areas of this experiment, the presence of thrips species may represent a risk to the crop.

Occurrence of thrips in different crop environments

In Ace White cultivar, the number of collected adult thrips per plant for sample occasion was significant higher in area A2 (0.664 ± 0.086 thrips/plant/occasion) compared to area A1 (0.192 ± 0.038 thrips/plant/occasion) ($H=6.9018$; $df= 1$; $p<0.05$). Regarding the number of captured thrips per plant, this also varied according to species and among environments (Table 2).

F. schultzei, F. occidentalis and H. gowdeyi species were collected in the two protected environments, while T. australis was only collected in area A1 and T. tabaci only in area A2. The diversity observed may be related to the crop type remained inside and outside the studied environments. Thrips often migrate from adventitious plants located in cropping fields or in their neighborhood to cropping plants (Northfield et al., 2008). However, these neighborhood plant species have not been studied to determine and assess the abundance and sources of thrips migration in this work. In area A2, the polyculture (lisianthus, baby's breath, goldenrod and snapdragon) at different development stages and the type of phytosanitary management adopted for each culture may have promoted the migration of thrips among bed plants, resulting in a major presence of thrips in flowers. This characteristic dispersion for different substrates is addressed by Mound (2013), who says it can be used in cropping management with the use of adhesive traps of different colors in order to monitor or control the populations of this insects group.

Table 2 - Species, number of individuals (N), mean (± standard error) of thrips species collected in cutting lisianthus crop (Ace White) in greenhouse in different areas of protected cultivation (A1 and A2) from January to February 2013 in Dois Irmãos, RS.

Specie	N		Mean of thrips/plant/occasion (±EP)	
	A1	A2	A1	A2
Frankliniella schultzei	39	152	0.156±0.04[b]	0.608±0.104[a]
Frankliniella occidentalis	5	11	0.02±0.01[a]	0.044±0.017[a]
Thrips tabaci	0	2	0	0.008±0.004
Thrips australis	1	0	0.004±0.004	0
Haplothrips gowdeyi	3	1	0.012±0.008[a]	0.004±0.004[a]
Total	48	166	0.192±0.038[b]	0.664±0.086[a]

N: number of individuals captured.
Row with different letters differ significantly by Kruskal -Wallis test and Dunn ($p<0.05$).

The size of cropping area and other structural differences such as the presence of side cloth in area A1 may have contributed to the lowest number of thrips/plant compared to A2 area during the assessment period. This result reinforces the findings of Berlinger et al. (2002) that tested the effectiveness of insect exclusion screens to prevent transmission of Tomato Yellow Leaf Curl Virus by *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) in commercial cultivation of tomatoes in Israel. The authors reported that virtually all table tomatoes in Israel had grown under exclusion screens from the year 2000 and their use proved to be an economically viable pest management method.

Our results corroborate Teitel (2006) who emphasized that screens act as a mechanical barrier in order to prevent migratory insects reach plants. Furthermore, the authors indicate that physical exclusion of insects in protected environment reduces the incidence of direct damage to crops and also viruses transmitted by them.

Comparison of thrips number collected in different lisianthus cultivars

Mean capture of thrips in area A1 differs among lisianthus cultivars (Figure 1). Marina and Ace White cultivars have higher average thrips captured by plant and differs from Orange Flash and Tu 668 cultivars (Figure 1).

The white colour of flower petal or the white kernel in mixed flower may have influenced higher presence of thrips in these cultivars, since polyphagous thrips that feed on a variety of plant tissues usually respond to white (ultraviolet low - UV), yellow and blue colours, while thrips with specific floral hosts are often attracted by the host flower colour (Czenz, 1987).

The colour seems to attract thrips species in different ways. Hoddle et al. (2002) reported high attraction to white by *F. occidentalis* in avocado flowers in California and blueberries in Florida (Liburd et al., 2009). Rodriguez-Saona et al. (2010) observed that white sticky traps were more effective than

coloured traps for thrips tracking in blueberry crop in New Jersey and explained that thrips are attracted to blueberry crop during flowering due to white colour of flowers. However, Gharekhani et al. (2014) observed that yellow sticky traps were more efficient than the white and blue ones. In the present study, despite coloured traps for catching insects have not been used, the greater presence of thrips in Marina and Ace White cultivars suggests the preference of these insects by inflorescence cultivars of white coloration rather than coloured cultivars Orange Flash and Tu 668 (Figure 1).

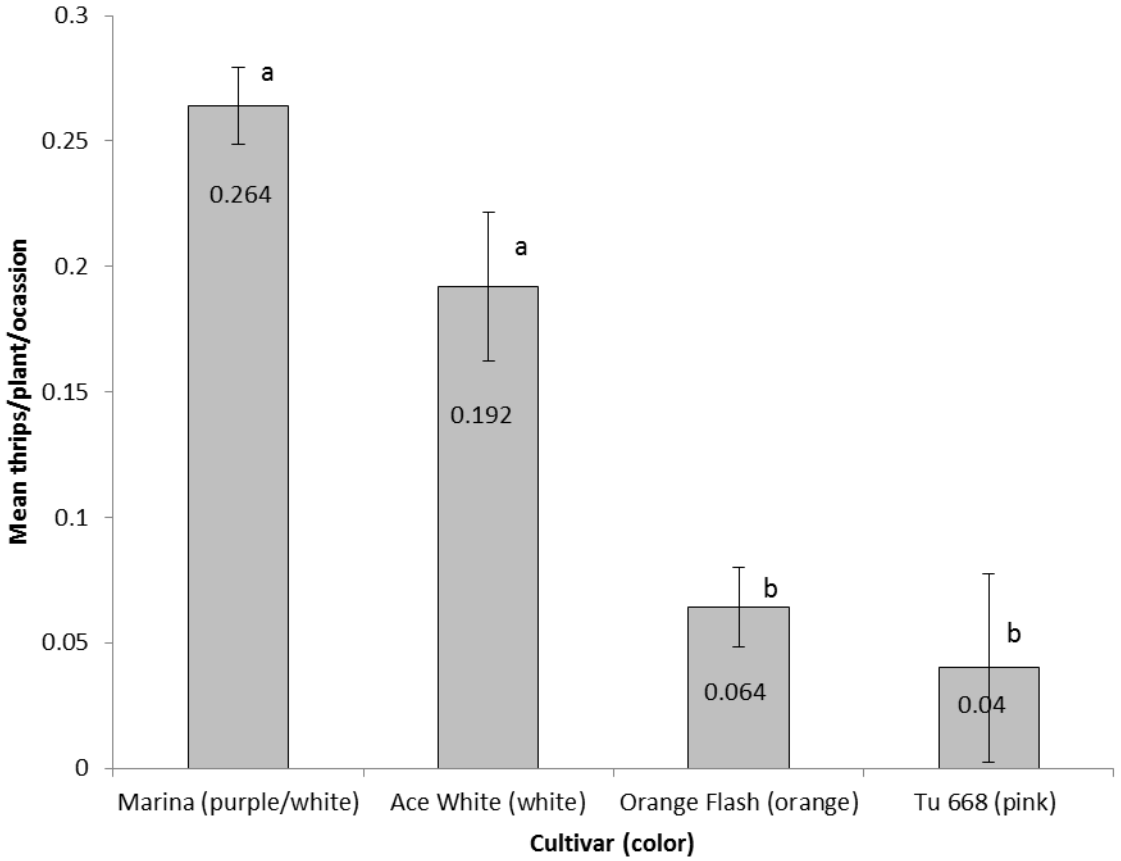

Figure 1 - Mean (± standard error) of thrips collected per lisianthus stem in Marina, Ace White, Orange Flash and Tu 668 cultivars in a greenhouse (A1 area) at each sampling occasion from January to February 2013. Dois Irmãos, RS, 2013. Columns with different letters differ significantly by Kruskal -Wallis test and Dunn ($p < 0.05$).

However other stimuli can also act in the attractiveness of these insects. De Kogel & Koschier (2001) evaluated the responses of thrips to odorous compounds of plants with olfactometer with Y tube. Several compounds were attractive to *F. occidentalis* adult females; while some odours were not attractive to *T. tabaci*. However, in other tests on these two species, the authors suggest that colour is a dominant factor for thrips on orientation for flowers. Thus, the possibility of management strategy would be the implementation of beds with colourful cultivars as borders of beds of white colouring cultivars aimed at protecting petals from the direct damage caused by the taste bite of these insects.

Conclusions

Different species of thrips are associated to cut lisianthus. *Frankliniella schultzei* and *F. occidentalis* are continuous species in lisianthus crop in protected environment and this is their first record in Brazil.

Structural differences in protected cultivation such as the presence of side protection screens seem to influence in quantity and composition of thrips species collected in lisianthus crop.

The greater presence of thrips in Marina and Ace White cultivars suggests the preference of thrips for white coloration cultivars rather than Orange Flash and Tu 668 coloured cultivars.

Acknowledgments

To Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) for providing financial support to the research and to Dr. Adriano Cavalleri (UFRGS) for identifying thrips species.

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