Infraspecific Taxon:		
Resident Species:		
Requestor Name and Affiliation:		
ITP Completed by:		
Date ITP started:	Date ITP completed:	

**INSTRUCTIONS** 

*Either check appropriate response or enter it in the designated space. Attach additional sheets with evidence as necessary using appropriate section numbers.* 

SUMMARY OF ITP RESULTS
Use Status Assessment
<ul> <li>Resident Species</li> <li>List independently of resident species</li> <li>Compare conclusions to resident species and use the most precautionary conclusions from the two assessments</li> </ul>
Use Predictive Tool
Infraspecific Taxon Conclusions
North:
Central:
South:
Resident Species Conclusions (from Status Assessment)
North:
Central:
South:

**Note1:** If the infraspecific taxon cannot be distinguished in the field from the resident species but it escapes and turns out to be more invasive than the resident species, it is assumed that the Conclusions for the resident species will become more precautionary over time as invasions of the infraspecific taxon are documented as new sites and impacts of the resident species. Because they must match those of the resident species, the Conclusions for the infraspecific taxon will also become more precautionary.

**Note2:** If the Conclusion is "Use of a predictive tool is recommended" then apply the predictive tool separately to the infraspecific taxon if possible. However, if this is not possible, apply the outcome of the predictive tool from the resident species to the infraspecific taxon.

## Section 1

(Only applies to infraspecific taxa that **can** be distinguished in the field from the resident species.)

<b>1.1.</b> Will botanists / field personnel typically be able to easily distinguish the infraspecific taxon fit the resident species or other infraspecific taxa? If no experts are given by requestor, select No.				
		YES	Provide information below, then Go to question 1.2	
		NO	Go to question 1.3	
Comr	nents: _			
<b>1.2</b> .	<b>1.2</b> . Is there evidence that the infraspecific taxon is likely to regress, revert, or produc would revert to the characteristics of the resident species? (If there is no evidence NO.)			
		YES	<i>Provide information below</i> ; Use the Status Assessment <i>and so indicate on</i> <i>Page 1</i> . For each zone, compare these conclusions to those of the resident species and use the most precautionary conclusions from these two assessments for the infraspecific taxon.	
		NO	Use the Status Assessment and select List independently of the resident Species on Page 1.	
Comme	ents:			

**1.3.** Has the resident species been assessed?



- YES Go to question 1.4
- NO Evaluate the resident species with the **Status Assessment** and indicate so on Page 1, then **Go to question 1.4**

**1.4.** Is the conclusion for the previously assessed, resident species "*Not a problem species; may be recommended*" or "*Use of a predictive tool is recommended*" for all three zones?

YES	Go to question	1.5
-----	----------------	-----

**NO Go to Section 2, question 2.1** 

**1.5.** Has the infraspecific taxon been in Florida (or in the U.S. if Florida data are not available) for > 10 years for herbaceous species or > 20 years for woody plants (if there is no evidence, then the answer is NO)?

YES Highlight attached distribution records that show presence in Florida before 10 or 20 years ago and enter a conclusion for infraspecific taxon on Page 1 of same per zone as the resident species

NO

Go to question 1.6

**1.6.** Are there *obvious* characteristics of the infraspecific taxon that make it likely to spread more quickly or have worse ecological impacts than the resident species?

YES Provide evidence below; Use Predictive Tool and indicate

so on Page 1

Examples for a YES answer include:

- Infraspecific taxon produces many more fruit/viable seeds than resident species.
- Infraspecific taxon hybridizes with Federal or Florida-listed Species of Special Concern, Threatened or Endangered plants or commercially-important species.
- Infraspecific taxon has been documented to be a problem elsewhere but the resident species has not been.

NO *Enter a conclusion for infraspecific taxon on Page 1 of* **same per zone as the resident species** 

\_\_\_\_\_

Comments: \_\_\_\_\_

#### Section 2

(Only applies to infraspecific taxa that **cannot** be distinguished in the field from the resident species and for which the previously assessed resident species has a conclusion of "*Caution; manage to prevent* escape" or "*Invasive; not recommended*" for at least one zone).

**2.1.** Is there evidence that the infraspecific taxon is likely to regress, revert, or produce hybrids that would revert to the characteristics of the resident species (if there is no evidence, the answer is NO)?

	YES	<i>Provide evidence below, enter a conclusion for infraspecific taxon on</i> <i>Page 1 of</i> <b>same per zone as the resident species</b>
	NO	Go to question 2.2
Comments: _		

**2.2.** Is there evidence that the combined characteristics that differ between the infraspecific taxon and the resident species will result in such <u>decreased</u> dispersal and spread compared to the resident species that the infraspecific taxon would be unlikely to become abundant in natural areas? Consider seed or vegetative propagules, spores, vegetative growth, etc. and the mechanism(s) by which the resident species has likely spread (including landscape waste material).

YES Provide evidence below then Go to question 2.3

Comments:

**2.3.** Is the primary negative ecological impact of the resident species linked to pollen-caused hybridization with natives or commercially important species, or another characteristic (e.g., host of pest/pathogen) that allows negative impacts in natural areas despite no or low spread <u>and</u> this characteristic is present in the infraspecific taxon?



YES Go to Section 3, question 3.1

□ NO

# Provide evidence below then enter a conclusion of "Not a problem infraspecific taxon; may be recommended"

Comments (If NO, provide evidence by listing the characteristics identified in questions 2.2 and 2.3):

**2.4.** Is there evidence that the combined characteristics that differ between the infraspecific taxon and the resident species will result in such <u>decreased</u> ecological impacts compared to the resident species that the infraspecific taxon would be unlikely to have negative ecological impacts in natural areas in any zones? If there is insufficient information about which traits in the resident species cause ecological impacts (see the IFAS Assessment of ecological impacts for the resident species), then answer NO.



YES *Provide evidence below, then enter a conclusion of* "Caution; may be recommended but manage to prevent escape"

NO Go to Section 3, question 3.1

Comments:

# Section 3

**3.1.** Does the infraspecific taxon have any characteristics that would shift its response per zone (e.g., changed tolerance to temperature)?

		YES	Provide evidence below then Go to question 3.2	
		NO	Enter a conclusion for infraspecific taxon on Page 1 of same per zone as the resident species	
Comr	nents: _			
3.2.	2. Does the shift in response per zone make the infraspecific taxon <u>more</u> likely to survive and ca ecological impacts in zones in which the resident species does not survive?			
		YES	Evaluate in which additional zones the infraspecific taxon would be able to survive compared to the resident species. For these zones, give the infraspecific taxon the most precautionary conclusion that was assigned to any zone of the resident species. For all other zones, the conclusions for the infraspecific taxon must be the same as for the resident species.	
		NO	Evaluate in which zones the infraspecific taxon would not be able to survive compared to the resident species. For those zones, the conclusion can be "Caution; manage to prevent escape". For all other zones, the conclusions for the infraspecific taxon must be the same as for the resident species.	

#### **Infraspecific Taxon Protocol Request**

Infraspecific taxon name:	
Resident species:	
Requestor name:	
Phone number:	E-mail:
Organization:	
Address:	

*Include any published documents and/or name and contact information of individual(s) who can provide information.* 

1. Provide publication or other appropriate documentation that the infraspecific taxon is a distinct entity (can be consistently and verifiably labeled). Include photographs if appropriate.

2. If this infraspecific taxon can be easily distinguished in the field, provide the names of three botanist/field experts who can verify this:

### Expert 1

Name:	Organization:	
Phone number:	E-mail:	
Expert 2		
Name:	Organization:	
Phone number:	E-mail:	
Expert 3		
Name:	Organization:	
Phone number:	E-mail:	

**3.** Is this infraspecific taxon is likely to regress, revert, or produce hybrids that would revert to the characteristics of the resident species, please attach.

#### **Infraspecific Taxon Protocol Request**

Date:
-------

**4.** Provide the date and information regarding the first introduction of the infraspecific taxon to Florida (or to the US if Florida data are not available).

 Describe how the plant traits of the infraspecific taxon differ from the resident species in relation to: life history, propagules production (seed & vegetative), dispersal mechanisms, hybridizations, plant hardiness, host to pests/pathogens.

6. Please provide other locations this infraspecific taxon occurs.

7. Additional information may be needed to complete the assessment on the taxon. Please include the names of three specialists (botanists, horticulturalists, plant breeders, etc.) who are familiar with this infraspecific taxon (these may or may not be the same experts as listed in item 2).

Expert 1		
Name:	Organization:	
Phone number:	E-mail:	
Expert 2		
Name:	Organization:	
Phone number:	E-mail:	
Expert 3		
Name:	Organization:	
Phone number:	E-mail:	

**Additional Information** 

# Responses to Questions on 'UF-1013-1' Lantana

Zhanao Deng and Sandra B. Wilson

**Question 1:** Specifically, could you provide a complete description of your statistical analysis. You referred to a simple ANOVA with mean separation as your analysis, but with 2 sites and multiple blocks, site and block should be accounted for in the model.

**Response:** We re-did the statistical analyses in JMP Pro in two ways: Simple ANOVA of the data (pollen stainability and fruit production for each site and ANOVA of combined data from the two sites. ANOVA outputs are provided in the document "Supplemental Materials – Statistical Analysis Outputs". The table below is a summary of the statistical analyses.

When the pollen stainability data from two sites were analyzed together, we observed significant differences among lantana cultivars (as we observed in simple ANOVA) and also significant differences between the two sites, which is new. When the fruit production data from two sites and four evaluations were analyzed in one model, we observed significant differences among lantana cultivars (as we observed significant differences among lantana cultivars (as we observed in simple ANOVA) as well as significant differences between two sites and among four evaluations. Overall, ANOVA of pollen stainability and fruit production from two sites (and four evaluations) provided new insights. Nevertheless, lantana cultivar difference in pollen stainability and fruit production remained significant, as observed in simple ANOVA. Based on the combined analysis, we have updated the main document.

B	Brief summary of statistical analysis of pollen stainability and fruit production										
		Expt. Or Sites	Blocks (or replicates)	Cultivars	Evalua- tions or harvests	Reference in Main document	Reference to Supplemental Materials				
Pollen stainability	Simple ANOVA – Expt. 1 (Balm)	NA	Not significant (P = 0.333)	Significant ( <i>P</i> < 0.0001)	NA	Table 3	Page 1, 1.1				
	Simple ANOVA – Expt. 2 (Ft. Pierce)	NA	Not significant (P = 0.3859)	Significant ( <i>P</i> < 0.0001)	NA	Table 3	Page 1, 1.2				
	ANOVA – Expt. 1 and 2 combined	Significa nt (P = 0.0027)	Not significant (P = 0.2210)	Significant (P <0.0001)	NA	Page 4	Page 1, 1.3				
Fruit production - Balm	Simple ANOVA – Balm, 1 <sup>st</sup> evaluation	NA	Not significant (P = 0.4932)	Significant (P <0.0001)	NA	Table 4	Page 2, 2.1.1				
	Simple ANOVA – Balm, 2 <sup>nd</sup> evaluation	NA	Not significant (P = 0.3833)	Significant (P <0.0001)	NA	Table 4	Page 2, 2.1.2				
	Simple ANOVA – Balm, 3 <sup>rd</sup> evaluation	NA	Not significant	Significant (P <0.0001)	NA	Table 4	Page 2, 2.1.3				



g) Lantana camara Pink Caprice in Florida



**Question 3:** One thing I have always wondered is how much of a reduction in pollen stainability is considered infertile? You indicate male pollen stainability was reduced by 95% from Pink caprice. How much was it reduced from the invasive *Lantana camara*? And generally, how different were the rest of the variables (germination rates, fruit production, etc.) measured for UF-1013-1 from the invasive *Lantana camara*?

**Response:** A former graduate student from our research program David Czarnecki conducted a pretty extensive study about 10 years to address the question related to pollen stainability. He used 10 *Lantana camara* cultivars and 15 breeding lines representing a range of ploidy level and pollen stainability (from 1.5% to 79.1%) and hand-pollinated *Lantana depressa* (Pro Native Consulting, Miami) reciprocally. His conclusion was that triploid *Lantana camara* with pollen stainability below 10%, even 15% in some cases, had little potential to cross-pollinate *Lantana depressa*. The graph below was copied from Dr. Czarnecki's dissertation. His dissertation was cited in the main document entitled "Main Characteristics, Fertility and Hybridization Potential of *Lantana camara* Cultivar 'UF-1013-1'. The pollen stainability of 'UF-1013-1' ranged from 2.0% to 2.5%, which is far below 10%. Also in hand pollination studies, 'UF-1013-1' did not cause any fruit set on *Lantana depressa*, indicating that 'UF-1013-1' is highly infertile.



Pink Caprice is a good representative of invasive *Lantana camara*. Compared to this invasive *lantana* plant, 'UF-1013-1' has greater than 95% reduction in pollen stainability and greater than 99% reduction in fruit production.

Regarding the level of fertility reduction needed, we contacted three plant breeding professors (Thomas Ranny at North Carolina State University, Neal Anderson at the University of Minnesota, and Ryan Contreras at the Oregon State University) in the U.S. universities who have been working on genetic sterilization of the invasive ornamental plants. Their replies indicated that the Oregon Department of Agriculture had developed a protocol for evaluating sterility and set the required level of fertility reduction for Buddleja. The required level of fertility reduction in Oregon for this genus is that candidate cultivars produces less than 2% viable seeds compared to fertile cultivars, that is, 98% reduction in female fertility. The contacted professors considered that this is reasonable. These breeders are not aware of any set requirements for male fertility or pollen stainability. The Oregon Department of Agriculture Protocol can be found at

<https://www.oregon.gov/ODA/shared/Documents/Publications/NurseryChristmasTree/BuddlejaScreenin g.pdf>. Also, I attached the Oregon Department of Agriculture protocol for your information.

**Question 4:** Generally, how long does it typically take invasive *Lantana camara* or Pink caprice to germinate? Also, what do you mean on line 145 by "abnormal"? How would that abnormality affect a germination trial?

**Response:** Typically when Pink Caprice seeds are germinated in Petri dishes, they will begin to germinate in seven days and reach the maximum germination in six to seven weeks (41 to 50 days). Below is a graph showing the germination of Pink Caprice seeds in Petri dishes. The germination study was conducted at the Indian River Research and Education Center in February through April 2016 using Pink Caprice seeds collected in 2015.

The abnormal lantana seeds were floating in the water and did not contain any developed embryos. Abnormal seeds did not germination.



**Question:** What do you mean by the experimental unit "two containerized plants"? Are these paired plants for the hand pollination (one experimental unit = depressa/ UF-1013-1 cross)? Also, did trials include UF-1013-1 crossed with other cultivars including Pink caprice?

**Response:** Two containerized plants of Lantana camara cultivars ('UF-1013-1', 'Bloomify Red', or 'Pink Caprice). Yes, the two plants were paired with two plants of *L. depressa*.

The concerns as expressed by Hammer (2004) (https://plants.ifas.ufl.edu/wp-

<u>content/uploads/files/caip/pdfs/TheLantanaMess.pdf</u>) has been hybridization of *L. camara* with *L. depressa*. So the hybridization potential of *L. camara* cultivars with other *L. camara* cultivars including Pink Caprice was not determined. However, based on four pieces of information (triploidy, very low pollen stainability, very little fruit production in the field where hundreds of other *L. camara* plants were grown side by side, and lack of hybridization potential with *L. depressa*), we expect that 'UF-1013-1' would not hybridize with other *L. camara* cultivars including Pink Caprice.

1	Main Characteristics, Fertility and Hybridization Potential of Lantana camara Cultivar
2	'UF-1013-1'
3	
4	Zhanao Deng <sup>1</sup> and Sandra B. Wilson <sup>2</sup>
5	
6	<sup>1</sup> University of Florida/IFAS, Department of Environmental Horticulture, Gulf Coast Research
7	and Education Center, 14625 County Road 672, Wimauma, FL 33598. Tel: 813-419-6605; E-
8	mail: zdeng@ufl.edu.
9	<sup>2</sup> University of Florida/IFAS, Department of Environmental Horticulture, Gainesville, FL 32611.
10	Tel: 352-273-4576; E-mail: sbwilson@ufl.edu.
11	
12	
13	1. Set-up of replicated field trials
14	Two replicated field trials were conducted simultaneously in Florida in 2015, one at the
15	UF/IFAS GCREC in Balm, FL (southwest Florida, USDA hardiness zone 9A, and AHS heat
16	zone 10), and one at the UF/IFAS Indian River Research and Education Center (IRREC) in Ft.
17	Pierce, FL (southeast Florida, USDA hardiness zone 9B, and AHS heat zone 9-10). The
18	experimental design used in Balm was a randomized complete block with three blocks and two
19	plants per plot (Fig. 2). Raised ground beds at the GCREC were fumigated with a multi-purpose
20	liquid fumigant (Pic-Clor 60®; active ingredients 1,3-Dichloropropene and chloropicrin) at 448
21	kilograms per hectare in Feb. 2015 and covered with white-on-black plastic. The experimental
22	design used in Ft. Pierce was also a randomized complete block, but with four blocks and a
23	single plant per plot. Ground beds at the IRREC were not fumigated but herbicided, disked and

24	covered with black ground cover. At each site, 'Pink Caprice' (Fig. 3) was included as a
25	"resident species" taxon. It is commercially produced and very prolific in fruit (and seed)
26	production (Fig. 4; Czarnecki et al., 2009). Although a named cultivar, it is most similar to the
27	escaped plants found along ditches and pastures (i.e. excessive fruiting, multicolored flowers,
28	and vigorous plants). Also at each site, 'Bloomify Red' was included as a sterile check.
29	Released under 'UF-1013A-2A', 'Bloomify Red' was determined not to be a problem species by
30	the IFAS Assessment's Infraspecific Taxon Protocol evaluation in 2016
31	(https://assessment.ifas.ufl.edu/assessments/lantana-camara-bloomify-red/). Characteristics of
32	'Bloomify Red' were described by Deng et al. (2017). In addition, 21 commercial cultivars with
33	various levels of male and female fertility were randomly placed in each block at both sites
34	where 'UF-1013-1' (Figs. 5 and 6) and 'Bloomify Red' were evaluated.
35	Prior to installation at the two sites, plants were propagated at GCREC. Cuttings were
36	taken on 9 Feb. 2015 and rooted in the greenhouse in 128-cell Speedling trays filled with a
37	customized potting substrate. The bottom ends of cuttings were treated with a rooting hormone
38	(Dip'n Grow, 1:10 dilution, final concentration 0.1% indole-3-butyric acid and 0.05% 1-
39	naphthaleneacetic acid) (Dip'n Grow Inc., Clackamas, OR). Rooted cuttings were pinched on 13
40	Mar. 2015, and then transplanted on 5 May 2015 to 10.2-cm plastic containers filled with a
41	commercial potting mix (Fafard 3B) and grown in the greenhouse at GCREC (at 15°C /night to
42	33°C /day). The container-grown plants were distributed to experimental sites in early June,
43	2015, and then transplanted to the ground beds. Transplanting was completed in the week of 12
44	June 2015. Each plant was top-dressed with approximately 15 grams of a controlled-release
45	fertilizer (Osmocote®: 15N-9P2Os-12K2O 5-6 months Scotts Marysville OH) and irrigated

through a seep system at the GCREC site and through drip tapes, twice a week and two hours perirrigation event, at the IRREC site.

48

#### 49 2. Main characteristics compared to the 'resident' taxon ('Pink Caprice')

'UF-1013-1' resulted from a cross between breeding line DROP-25 (L. camara) and 50 51 'Landmark Flame Improved' (L. camara). The cross was made in fall 2010 at the GCREC, and 'UF-1013-1' was initially selected as an individual plant in April 2012. 'UF-1013-1' is distinctly 52 different from 'Pink Caprice' morphologically (plant and flower), cytologically (nuclear DNA 53 54 content and ploidy level), and molecularly (simple sequence repeat or SSR marker profile). Table 1 summarizes the main differences between 'UF-1013-1' and 'Pink Caprice'. 55 'UF-1013-1' is a sibling of the approved and released cultivar 'Bloomify Red' ('UF-56 1013A-2A'). They are different in several characteristics. 'UF-1013-1' is shorter and its flower 57 clusters are smaller. 'UF-1013-1' contains approximately 6% higher nuclear DNA and has a 58 different DNA fingerprinting profile when analyzed with molecular (SSR) markers (Table 2). 59

60

#### 61 **3.** Pollen stainability

Previous studies have shown that pollen stainability is a good indicator of lantana's male fertility (or sterility) and hybridization potential with *Lantana depressa*, the Florida's native lantana species (Czarnecki, 2011; Czarnecki et al., 2012; Czarnecki et al., 2014; Dehgan and Guy, 2004; Hammer, 2004). Czarnecki (2011) showed that triploid *L. camara* with low pollen stainability (<15%) had little potential to cross-pollinate *L. depressa*.

Two pollen staining experiments were conducted using fresh anthers collected from theabove-described field-grown plants. In Expt. 1, newly opened flowers were collected from plants

69	grown in Balm, FL in late July 2015, and anthers were extracted from the flowers and collected
70	into a 1.5-mL Eppendorf tube. The collected anthers were stained with 10 <sup>-6</sup> M fluorescein
71	diacetate (FDA) (Sigma-Aldrich, St. Louis, MO) in 0.22 M sucrose at room temperature in the
72	dark for 1 hour (Czarnecki et al., 2014). Stained anthers were transferred onto a microscope slide
73	and covered with a coverslip. Pollen grains in the anthers were released by gently tapping and
74	pressing the coverslip and then examined under a fluorescent microscope. Plump, round pollen
75	grains fluorescing bright yellowish green light were considered stainable, while misshaped, non-
76	fluorescing, or unevenly, lightly fluorescing pollen grains were counted as non-stainable. In
77	Expt. 2, flowers were collected from lantana plants grown in Ft. Pierce, FL in mid-August 2015.
78	Anther staining and pollen examination were performed as above described.
79	The number of pollen grains examined for each lantana cultivar in each staining
80	experiment was between 1,094 and 2,122 (Table 3). An analysis of variance (ANOVA) and
81	mean separation were conducted using JMP Pro 14.1.0 (SAS Institute, Cary, NC) to compare the
82	pollen stainability of 'UF-10131-1' with that of 'Bloomify Red' and 'Pink Caprice' and to
83	compare the pollen stainability data of the three lantana varieties from two Experiments or two
84	sites (Balm and Ft. Pierce). Results indicated that the two experiments (two sites) had a
85	significant difference, with a higher pollen stainability in Expt. 2 (Ft. Pierce) than in Expt. 1
86	(Balm). The mean pollen stainability of 'UF-1013-1' was 2.2% (Table 3), comparable to the
87	mean pollen stainability of the previously released sterile cultivar 'Bloomify Red' and indicating
88	little potential to hybridize with other lantana plants (Czarnecki, 2011). The mean pollen
89	stainability of 'Pink Caprice' was 73.1%. These results indicate that the pollen stainability (or
90	male fertility) of 'UF-1013-1' was reduced substantially by 95% from that of 'Pink Caprice'.
91	

92 **4. Female fertility** 

Previous studies have indicated that fruit (seed) production per peduncle and seed
germination or seedling emergence are the primary factors determining lantana's female fertility
(or sterility) and that it is possible to factor these two characteristics into a female fertility index
(FFI) by multiplying fruit production per peduncle and seed germination (Czarnecki, 2011;
Czarnecki et al., 2012).

Fruit production per peduncle in replicated field trials: Fruit production data were 98 regularly collected from field-grown plants in Balm and Ft. Pierce. In each round of fruit harvest, 99 100 20 peduncles were randomly sampled from each plant in the replicated field trials (see above). and drupes on these peduncles were counted, regardless of maturity. A total of four harvests 101 were made for each plant at each experimental site. Thus, in each fruit harvest, approximately 102 103 120 peduncles were sampled for each cultivar grown in Balm, and approximately 80 peduncles were sampled for each cultivar grown in Ft. Pierce. The four harvests in Balm were done on 17 104 Aug., 14 Sept., 16 Oct., and 18 Nov., 2015, respectively. The four harvests in Ft. Pierce were 105 done on 12 Aug., 10 Sept., 14 Oct., and 11 Nov. 2015, respectively. 106

An analysis of variance and separation of mean fruit production values were conducted 107 108 using JMP Pro 14.1.0 (SAS Institute) to compare the fruit production of 'UF-1013-1' with that of 'Bloomify Red' and 'Pink Caprice' and to compare fruit production from the two sites in four 109 evaluations. Results indicated no significant differences between the two sites (3.257 and 2.651) 110 111 or between blocks, but highly significant differences among lantana varieties and four harvests (or evaluations). The mean fruit production of three lantana varieties in the first harvest (4.212) 112 113 was significantly higher than the mean fruit production of these varieties in the second (2.821), 114 third (2.611), or fourth harvest (2.173).

115	As previously reported by Deng et al. (2017), 'Pink Caprice' produced the largest number
116	of drupes among all the entries in the two replicated trials (Fig. 4; Table 4). Each peduncle bore
117	an average of 7.941 drupes in Ft. Pierce and 10.313 drupes in Balm, with an overall average of
118	9.127 across the two sites and four harvests. The number of drupes per peduncle for the sterile
119	cultivar 'Bloomify Red' ranged from 0 to 0.050 and averaged to 0.015 across the two sites over
120	the 4 months. The number of drupes 'UF-1013-1' produced per peduncle ranged from 0 to 0.038
121	and averaged to 0.009 across two experimental sites and over 4 months (Table 4 and 5). This
122	level of fruit production in 'UF-1013-1' represented greater than 99% reduction from the fruit
123	production of 'Pink Caprice'.
124	Fruit production in field trials in Citra: In spring 2018, 'UF-1013-1' was included in
125	an independent field trial run by Dr. R. Freyre, flower breeder at the UF/IFAS Environmental
126	Horticulture Department, and graduate student Mr. A. Moseley. 'UF-1013-1' and 'Bloomify
127	Red' didn't produce fruit while a commercial variety produced an average of 1.2 fruit per
128	peduncle (data not shown).
129	Seed germination: This was conducted as previously reported by Deng et al. (2017).
130	Mature drupes were collected from each plant in the above described experiments. Seeds were
131	extracted, cleaned, and air-dried at each test site and germinated at the IRREC. Due to having
132	few fruit for 'UF-1013-1' and 'Bloomify Red', fruit from four harvests at each site were
133	combined before seed extraction. Seeds were germinated in a 10.9-cm $\times$ 10.9-cm transparent
134	polystyrene germination boxes (Hoffman Manufacturing, Corvallis, OR) containing 2 sheets of
135	germination paper (Anchor Paper Company, St. Paul, MN) moistened with 15 mL of water.
136	Germination boxes were placed in temperature and light-controlled chambers equipped with
137	cool-white fluorescent lamps (Model 818; Precision Scientific, Winchester, VA). The

germination condition was 12 h light at 25°C (photosynthetic photon flux was 22 to 30 µmol m<sup>-2</sup>
s<sup>-1</sup> at shelf level) followed by 12 h dark at 15°C. Germination of seeds was monitored every other
day for a period of 60 days. An additional 5-10 mL of nanopure water was added to the
germination boxes as needed. A seed was considered germinated when radicle emergence was
2.0 mm or greater. Seeds were removed once germination occurred to prevent inaccurate data
collection.

144 Seeds of 'Pink Caprice' were also sent to a commercial seed testing laboratory (Midwest 145 Seed Services, Brookings, SD) for seed viability tests. The distal end of the cotyledon of each 146 seed was cut off and seeds were stained overnight at 30°C in 1.0% tetrazolium (2, 3, 5-triphenyl 147 chloride). Seeds were considered viable if the entire embryo stained evenly. 'UF-1013-1' and 148 'Bloomify Red' ('UF-1013A-2A') produced very few or no seeds at either site and were 149 therefore not subjected to viability tests.

As previously reported by Deng et al. (2017), seeds of 'Pink Caprice' showed an average of 65.0% viability, germinated readily, with an average germination percentage of 45.0% in 60 days (Table 5). For 'Bloomify Red', no drupes or no mature drupes were collected from Balm or Ft. Pierce, thus no seeds were available for seed viability test or germination. As for 'UF-1013-1', three mature drupes were collected at Ft. Pierce trials over 4 months. Three seeds were extracted, but all were abnormal. Thus, no seeds were available for 'UF-1013-1' and 'Bloomify Red' to conduct seed viability or germination tests.

# 157 Female Fertility Index (FFI): The FFI for 'Pink Caprice' was 4.107 (Table 5), similar to 158 previously reported values (Czarnecki et al., 2014) and indicating an extremely high level of 159 female fertility. Because of the lack of seed germination data, it was not possible to calculate the

FFI for 'UF-1013-1'. However, based on its triploidy and extremely low fruit production, it was
expected that the FFI for 'UF-1013-1' would be close to 0 and similar to that of 'Bloomify Red'.

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#### 163 5. Hybridization potential with *L. depressa* after hand pollinations

Hand pollination experiments were performed in the greenhouse at GCREC in June and 164 165 July 2015 to assess the hybridization potential of 'UF-1013-1', as a male or female patent, with L. depressa. 'Bloomify Red' and 'Pink Caprice' were included in the hand pollination 166 167 experiments as a sterile and a fertile lantana check, respectively (Deng et al., 2017). Stock plants of all lantana cultivars and L. depressa were grown on metal benches in 1-gallon plastic 168 containers filled with a commercial soilless mix (Fafard 3B) amended with a controlled release 169 170 fertilizer (Osmocote®,15N-3.9P-10K, 5-6 months release at 21 °C; The Scotts Company) at 7.12 kg  $\cdot$  m<sup>-3</sup>. The stock plants were arranged into three blocks and in each block, they were randomly 171 placed on the benches. The experimental unit was two containerized plants for each L. camara 172 cultivar and two containerized plants of L. depressa. Temperatures inside the greenhouse ranged 173 from a low of 21 °C at night to a high of 33 °C during day. No supplemental lighting was 174 175 provided. Plants were drip-irrigated twice a day. Fresh anthers were collected from mature unopened flowers of male parents and applied immediately to emasculated flowers of female 176 parents. At maturity, fruit produced by the pollinated flowers were collected and counted, and 177 seeds were extracted and germinated to determine seed germination. 178 As previously described by Deng et al. (2017), 'Pink Caprice', as a male parent, caused 179

an average of 8.6% fruit set on *L. depressa* flowers (Table 6). When pollinated with *L. depressa*,
'Pink Caprice' flowers showed 19.7% fruit set (Table 6). Seeds from crosses between 'Pink
Caprice' and *L. depressa* or vice versa showed 11.1% or 19.7% seedling emergence (Table 6).

As a male parent, 'Bloomify Red' did not cause any fruit set on *L. depressa* flowers. Nor did it
set any fruit after having been hand-pollinated with *L. depressa*.

A total of 389 *L. depressa* flowers were pollinated with 'UF-1013-1', and none of the pollinated flowers set fruit, resulting in 0% fruit set (Table 6). When 'UF-1013-1' was used as the female parent, it did not set any fruit after having been pollinated with *L. depressa*. Thus, 'UF-1013-1' did not hybridize with *L. depressa* when they were hand pollinated (Table 6). These data confirm the high level of male and female infertility in 'UF-1013-1'.

190

#### 191 **6.** Conclusion

Compared to 'Pink Caprice', a cultivar of *L. camara* that is the closest to the species' 192 resident taxon (wild or naturalized type), the pollen stainability of 'UF-1013-1' has been reduced 193 by more than 95%. This new triploid cultivar did not cause fruit set or set any fruit when used as 194 a male or female parent in hand-pollination with L. depressa. Fruit production of this triploid has 195 been reduced by greater than 99% and it did not produce normal, viable seeds in replicated field 196 trials. The high level of male and female infertility of this triploid was stable in Balm, Ft. Pierce 197 and Citra. These results indicate that 'UF-1013-1' has little potential to hybridize with L. 198 199 *depressa* to produce viable interspecific progeny.

200

#### 201 7. Acknowledgement

The development and evaluation of 'UF-1013-1' were funded in part by USDA hatch projects (Project no. FLA-GCR-005065 and FLA-GCC-005507), the former USDA/Tropical and Subtropical Agriculture Research (TSTAR) program, and the Florida Department of Agriculture Consumer Service (FDACS) Specialty Crop Block Grant program (Project no. 021747). Dr. X.

206	Ying stained lantana pollen, counted stainable pollen grains, and performed hand pollination. G.
207	Bowman, M. Derrick, P. Frey, and J. Jones provided extensive technical assistance. Dr. C. Chen
208	did SSR marker analysis. Dr. R. Freyre and A. Moseley collected data from their trials in Citra,
209	FL in 2018. Ball Horticultural Company, Proven Winners North America, LLC, and Riverview
210	Flower Farms, Inc., trialed 'UF-1013-1' in West Chicago, California, or Florida and shared their
211	observations, fruit count data, and/or photos.
212	
213	8. Literature Cited
214	Chen, C., C.H. Bock, W.R. Okie, F.G. Gmitter Jr., S. Jung, D. Main, T.G. Beckman, and B.W.
215	Wood. 2014. Genome-wide characterization and selection of expressed sequence tag
216	simple sequence repeat primers for optimized marker distribution and reliability in peach.
217	Tree Genetics and Genomes 10:1271-1279.
218	Czarnecki II, D.M. 2011. Genetic sterilization and reproductive biology of Lantana camara. PhD
219	Diss., Univ. of Fla., Gainesville.
220	Czarnecki II, D.M. and Z. Deng. 2009. Occurrence of unreduced female gametes leads to sexual
221	polyploidization in lantana. J. Amer. Soc. Hort. Sci. 134(5):560-566.
222	Czarnecki II, D.M., A.J. Hershberger, C.D. Robacker, D.G. Clark, and Z. Deng. 2014. Ploidy
223	level and pollen stainability of Lantana camara cultivars and breeding lines. HortScience
224	49:1271-1276.
225	Czarnecki II, D.M., S.B. Wilson, G.W. Knox, R. Freyre, and Z. Deng. 2012. UF-T3 and UF-T4:
226	Two sterile Lantana camara cultivars. HortScience 47:132-137.
227	Dehgan, B. and C.L. Guy. 2004. Reproductive biology and invasive potential of Lantana
228	camara. 1 July 2016. < http://www.reeis.usda.gov/web/crisprojectpages/191420.html>.

229	Deng, Z., S.B. Wilson, X. Ying, and D.M. Czarnecki. 2017. Infertile Lantana camara cultivars
230	UF-1011-2 and UF-1013A-2A. HortScience 52(4):652-657.
231	Hammer, R.L. 2004. The lantana mess: A critical look at the genus in Florida. The
232	Palmetto, 23(1): 21-23.
233	Langeland, K.A., H.M. Cherry, C.M. McCormick, and K.A.C. Burks. 2008. Lantana camara L.,
234	p126. In: K.A. Langeland, H.M. Cherry, C.M. McCormick, and K.A.C. Burks (second
235	edition). Identification and biology of nonnative plants in Florida's natural areas.
236	University of Florida IFAS Communication Services, Gainesville, FL.
237	
238	
239	

- 240 Table 1. Summary of major characteristics of 'UF-1013-1' as compared to 'Pink Caprice', a
- cultivar close to the resident taxon.

	Resident taxon	'UF-1013-1'
Plant vigor, size	Very vigorous, much larger	Moderate vigor, much smaller
Plant branching	Erratic branching, stems of	Mounding growth habit, round form
habit, and form	various lengths, open plant	(Fig. 5)
	center, irregular form (Fig. 3)	
Flower color	Yellow center and light pink	Yellow and red (Fig. 6)
	(Fig. 4)	
Fruit and seed	Very high fruit set; produce lots	Extremely low or absent fruit set
production	of fruit and seed (Table 4; Fig. 4)	(Table 4)
Pollen staining	Round, fully developed grains;	Misshaped, aborted; much fewer
	the majority of the pollen grains	pollen grains; the great majority of
	are deeply stained (Table 3)	pollen grains do not stain or stain
		very lightly (Table 3)
Nuclear DNA	$6.25 \pm 0.17$ (Table 3)	4.82 ± 0.11(Table 3)
content (pg/2C)		
Ploidy level	Tetraploid (Table 3)	Triploid (Table 3)
DNA marker profile	Different marker profile at three	Different marker profile at three SSR
	SSR markers (Lantana11, 12 and	markers (Lantana11, 12 and 20) (Fig.
	20) (Fig. 1; Table 2)	1; Table 3)

	Alle	les am	plifie	d by SSR	markers	s (size	of alle	les in	base	pairs) <sup>y</sup>		
Lantana	Mar	ker La	intana	11	Mark	er Lar	itana 12	2			Lant	ana20
cultivars <sup>z</sup>	150	152	156	160	135	143	145	147	150	152	93	109
UF-1013-1		+		+	+	+		+			+	
Bloomify Red	+	+		+		+	+				+	
DROP-25	+	+		+	+	+	+	+			+	+
Pink Caprice		+	+	+		+	+		+	+	+	+

<sup>z</sup> The other parent of 'UF-1013-1' ('Landmark Flame Improved') was not available for analysis.

<sup>y</sup> Lantana genomic DNA was isolated from lantana leaves at the GCREC, Balm, FL. SSR marker

analysis (PCR-based DNA amplification, capillary electrophoresis, and allele scoring) was

conducted by Dr. Chunxian Chen at the USDA/ARS Fruit and Tree Nut Research Laboratory,

**Table 2.** Results from simple sequence repeat (SSR) marker analysis of 'UF-1013-1' and

'Broomify Red' ('UF-1013A-2A') breeding line DROP-25 and 'Pink Caprice' a highly prolific

Byron, GA, using a procedure previously described by Chen et al. (2014), with minor

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259

261 modifications. PCR was performed on a C1000 Touch Thermal Cycler with a CFX384 block

module (Bio-Rad, Hercules, CA) in a 5- $\mu$ L volume consisting of 1× PCR buffer, 0.2 mM dNTPs,

263 2 mM MgCl<sub>2</sub>, 0.3  $\mu$ M of the dye-labeled forward and regular reverse primers, 0.5U Taq DNA

264 polymerase (BioExpress, Kaysville, UT), and ~10 ng lantana genomic DNA template. A

touchdown PCR program was used, with an initial step of 94°C for 3 min, followed by 10 cycles

of denaturation at 94°C for 30 sec, annealing at 68°C for 30 sec with a 0.5°C decrement each

267	cycle, and extension at 72 °C for 45 sec, followed by 25 more cycles with a constant annealing
268	temperature of 63°C (other parameters were the same), plus a final extension at 72°C for 15 min.
269	The dye-labeled PCR products were separated on a 3500 Genetic Analyzer (Life Technologies,
270	Carlsbad, CA) to generate the chromatographic trace files. The SSR allele table and peak
271	chromatograms were generated using GeneMarker 2.4 (SoftGenetics, State College, PA).
272	Primers used in the above PCR reactions were Lantana-specific primers developed from
273	SSR-enriched lantana genomic sequences (L. Gong and Z. Deng, unpublished). Primer
274	sequences are as follows.
275	Lantana11F: (M13 tail sequence)-TGCAATTGGAGGCTTTTTCT, and Lantana11R:
276	AAAGCAGCTTCAAGTTTGTGC.
277	Lantana12F: (M13 tail sequence)- GGATGAGATGATAAGGTAGGGTGT, and Lantana12R:
278	TTGGTGGTGATGACTTTGATTC.
279	Lantana20F: (M13 tail sequence)-AGAATCAGGGTTTGGGGGTTG, and Lantana20R:
280	TCGTAGCCACCACTCCTCAC.

281 M13 tail sequence = CCCAGTCACGACGTTG.

Table 3. Nuclear DNA content, ploidy level, and pollen stainability of lantana cultivar 'UF-1013-1' and two checks, 'Bloomify Red' and 'Pink
Caprice', grown in Balm and Ft. Pierce, FL in full sun in 2015.

284									
285 286		Nuclear DNA content ± SD	Ploidy level	Pollen grair	ns examined (no.)	Pollen stainability (%) <sup>z</sup>			
287	Cultivars	(pg/2C)		Expt. 1	Expt. 2	Expt. 1	Expt. 2	Mean	
288									
289	UF-1013-1	$4.82 \pm 0.11$	<i>3</i> ×	1464	1840	<mark>2.0</mark> b	<mark>2.4</mark> b	2.2 b	
290	Bloomify Red	$4.54 \pm 0.08$	<i>3</i> ×	2122	1466	1.5 b	4.5 b	3.0 b	
291	Pink Caprice	$6.25 \pm 0.17$	$4 \times$	<mark>1271</mark>	1094	70.8 a	75.3 a	73.1 a	
292									

<sup>2</sup> Lantana anthers used in Expt. 1 were collected from plants grown in ground beds in full sun in Balm, FL and stained on 22 July 2015; lantana

anthers used in Expt. 2 were collected from plants grown in the replicated field trials in Ft. Pierce, FL and stained on 13 Aug. 2015. Pollen

stainability data were arcsine-transformed before analysis of variance was performed. Means with the same letter within the column are not

significantly different by the LSD procedure at P < 0.05.

**Table 4.** Fruit production of 'UF-1013-1' and the two check cultivars 'Bloomify Red' ('UF-1013A-2A') and 'Pink Caprice' grown outdoors in

Expt. Site	Cultivars	Fruit per peduncle at 8 to 21 weeks post transplanting (WPT)				Total peduncles	Total fruit collected	Total mature fruit collected	Mean fruit per peduncle
		8	12	16	21	examined	(no.)	(no.)	r r
		Aug. 12	Sept. 9	Oct. 7	Nov. 11	(no.)			
Balm <sup>z</sup>	UF-1013-1	0.008 b	0.008 b	0 b	0 b	480	2	0	0.004 b
	Bloomify Red	0.012 b	0.050 b	0.025 b	0 b	481	11	0	0.023 b
	Pink Caprice	14.258 a	8.850 a	10.117 a	8.025 a	480	4,950	1,416	10.313 a
		8	12	17	21				
		Aug. 12	Sept. 10	Oct. 14	Nov. 11				
Ft.	UF-1013-1	0.013 b	0 b	0.038 b	0 b	320	4	3	0.013 b
Pierce <sup>y</sup>									
	Bloomify Red	0 b	0 b	0 b	0 b	320	0	0	0 b
	Pink Caprice	11.590 a	8.325 a	6.263 a	5.588 a	320	2,541	1,832	7.941 a

ground beds in full sun at two sites in Florida (2015).

299

 $^{2}20$  peduncles randomly sampled on each of the two plants in three blocks in Balm, FL over 4 months in 2015.

301 <sup>y</sup>20 peduncles randomly sampled on each plant in four blocks in Ft. Pierce, FL over 4 months in 2015.

Table 5. Fruit production, seed viability, seed germination, and female fertility of lantana cultivars 'UF1013-1' and two checks ('Bloomify Red' and 'Pink Caprice') grown outdoors in ground beds in full sun
at two sites in Florida (2015).

305	
306	

306		Mean	Seeds	Seed	Seeds	Seed	Female
307	Lantana	fruit	examined	viability	planted	germination	fertility
308	cultivars	production <sup>z</sup>	(no.)	(%) <sup>y</sup>	(no.) <sup>x</sup>	(%) <sup>x</sup>	(FFI) <sup>w</sup>
309							
310	UF-1013-1	0.009					close to 0
311	Bloomify Red	0.012					close to 0
312	Pink Caprice	9.127	100	65.0	100	45.0	4.107
313							

<sup>z</sup>Average of fruit production per peduncle of two sites (Balm and Ft. Pierce).

<sup>y</sup> Average seed viability of 100 seeds (2 replicates of 50 seeds from Balm and Ft. Pierce sites) determined

by the Midwest Seed Services (Brookings, SD). Seed viability tests were not performed for 'UF-1013-1'

and 'Bloomify Red because seeds were not available.

318 <sup>x</sup>Seed germination was conducted at the Indian River Research and Education Center in Ft. Pierce, FL

beginning 2 Feb. 2016 and for 60 days. There were no seeds available for 'UF-1031-1' and 'Bloomify

320 Red'. One hundred seeds of 'Pink Caprice' (50 from Ft. Pierce and 50 from Balm) were tested for seed

321 germination.

322 <sup>w</sup>Female fertility index = average fruit production per peduncle  $\times$  seed germination (%) /100. The female

- fertility index of 'UF-1013-1' and 'Bloomify Red' could not be calculated because they did not produce
- mature seeds. It was expected that their female fertility index would be close to 0.

Table 6. Hybridization potential of 'UF-1013-1' with *L. depressa* after hand pollinations, as compared to 'Bloomify Red' (sterile) and 'Pink
Caprice' (fertile).

327								
328		L. depressa as the female parent			L. depressa as the male parent			
329								
330		Flowers	Fruit set	Seedling	Flowers	Fruit set	Seedling	
331	Cultivar	pollinated (no.)	(%)	emergence (%)	pollinated (no.)	(%)	emergence (%)	References
332								
333	UF-1013-1	389	0 b <sup>z</sup>		496	0 b <sup>z</sup>		
334	Bloomify Red	353	0 b		558	0 b		Deng et al. (2017)
335	Pink Caprice	388	8.6 a	11.1	452	19.9 a	15.8	Deng et al. (2017)
336								

<sup>2</sup>Fruit set data were arcsine-transformed before analysis of variance was performed in JMP Pro 12.0.1. Means with the same letter within the

column are not significantly different by the LSD procedure at P < 0.05.



Figure 1. SSR marker profile of 'UF-1013-1' (first row), 'Bloomify Red' (second row), breeding



342 Caprice' with SSR marker Lantana12.



- Figure 2. The 2015 replicated lantana field trials in Balm, FL on 21 Oct. 2015. Flowers were
- 347 collected from these plants for pollen viability tests. The plants were evaluated monthly for four
- months (Aug.-Nov.) for fruit production, plant and flower morphology, and plant performance.

349



350

Figure 3. Plants of 'Pink Caprice' lantana propagated by cutting, grown in a soilless mix for 95

days, and grown outdoors in the ground bed in Balm, FL for 131 days. Photo was taken at the

University of Florida Gulf Coast Research and Education Center in Balm, FL on 21 Oct. 2015.

354 'Pink Caprice' plants were very vigorous, grew very rapidly, and showed an erratic branching355 habit.



Figure 4. Flowers and infructescences of 'Pink Caprice' grown outdoors in ground beds in fullsun in Balm, FL. The plant was propagated by cuttings, grown in a soilless mix, and then grown

outdoors in the ground bed. Photo was taken at the University of Florida Gulf Coast Research
and Education Center in Balm, FL on 21 Oct. 2015. Flowers of 'Pink Caprice' were light pink

- 362 and each seed head bore numerous fruit.



- 370
- Figure 5. Plants of 'UF-1013-1' lantana propagated by cutting, container-grown in a soilless mix
- for 95 days, and grown in the ground beds in Balm, FL for 124 days. Photo was taken at the
- University of Florida Gulf Coast Research and Education Center in Balm, FL on 14 Oct. 2015.
- 374



- 375 376
- Figure 6. Flowers and inflorescences of 'UF-1013-1' grown outdoors in ground beds in full sun
- in Balm, FL. The plant was propagated by cuttings, container-grown in a soilless mix, and then
- 379 grown outdoors in the ground bed. Photo taken at the University of Florida Gulf Coast Research
- and Education Center in Balm, FL on 14 Oct. 2015.
- 381

			(P = 0.4733)				
	Simple ANOVA – Balm, 4 <sup>th</sup> evaluation	NA	Not significant (P = 0.4444)	Significant (P=0.0040)	NA	Table 4	Page 2, 2.1.4
	Simple ANOVA – Balm, 4 evaluations combined	NA	Not significant (P = 0.7599)	Significant (P <0.0001)	Significan t ( <i>P</i> = 0.0274)	Table 4	Page 3, 2.1.5
Fruit production – Ft. Pierce	Simple ANOVA – Ft. Pierce, 1 <sup>st</sup> evaluation	NA	Not significant (P = 0.4552)	Significant (P <0.0001)	NA	Table 4	Page 3, 2.2.1
	Simple ANOVA – Ft. Pierce, 2 <sup>nd</sup> evaluation	NA	Not significant (P = 0.4447)	Significant (P <0.0001)	NA	Table 4	Page 3, 2.2.2
	Simple ANOVA – Ft. Pierce, 3 <sup>rd</sup> evaluation	NA	Not significant (P = 0.4426)	Significant $(P = 0.0002)$	NA	Table 4	Page 3 & 4, 2.2.3
	Simple ANOVA – Ft. Pierce, 4 <sup>th</sup> evaluation	NA	Not significant ( <i>P</i> = 0.4547)	Significant $(P = 0.0002)$	NA	Table 4	Page 4, 2.2.4
	Simple ANOVA – Ft. Pierce, 4 evaluations combined	NA	Not significant (P = 0.4785)	Significant ( <i>P</i> <0.0001)	Significan t (P = 0.0149)		Page 4, 2.2.5
					at 1.7		
Fruit production	2 sites & 4 evaluations combined	Not significa nt ( <i>P</i> = 0.1080)	Not significant (P = 0.4040)	Significant ( <i>P</i> <0.0001)	$\frac{\text{Significan}}{(P = 0.0006)}$	Page 5	Page 5, 2.3

(Note: NA = Not applicable).

**Question 2:** Also, I don't have my files here with me, but I think we normally consider invasive type invasive *Lantana camara* (<u>https://assessment.ifas.ufl.edu/assessments/lantana-camara/</u>) as the "resident" species. The justification for that is we want to compare the cultivar to what is currently invading natural areas, not another cultivar. So I am curious why pink caprice was used and if this is what we have done in the past.

**Response:** Ten years ago (2009) when we began testing infertile lantana lines, we examined all of the available literature as well as naturalized lantana plants found in public parks and along road ditches in central and south Florida. The literature we studied included the article by Dr. Langeland et al., the article posted at the UF/IFAS Center for Aquatic and Invasive Plants (<u>https://plants.ifas.ufl.edu/plant-directory/lantana-camara/</u>), the article by R.L. Hammer (2004), Atlas of Florida Plants (<u>http://florida.plantatlas.usf.edu/Plant.aspx?id=1789</u>), etc.

We also conducted a replicated study comparing the seed germination of Pink Caprice and the resident Taxon (seeds were collected from a ditch along Rock Road in Fort Pierce, FL). Pre-germination viability

of seeds were 76% for 'Pink Caprice' and 44% for the resident taxon, presumably due to observed seed predation of plants. Nevertheless, both flowered similarly, fruited prolifically, and germinated readily between 14-60 days.

Based on our literature search and examination of naturalized lantana plants, we found out that Pink Caprice was highly similar to the naturalized lantana plants, including vigorous growth up to 6 feet or more in height, multicolored flowers that change color over time, being extremely prolific in seed production, and producing thousands of fruit per plant a year (<u>https://plants.ifas.ufl.edu/plantdirectory/lantana-camara/</u>). We also felt that the representative "resident" *Lantana camara* plant to be used should be readily available to other researchers who might want to use in their research or evaluation. If we had used the lantana plants we collected locally, other researchers might not be able to find naturalized lantana plants that can serve as common controls. Considering these factors, we felt that Pink Caprice could represent the "resident" lantana species better than locally found plants. When we presented our first ITP request forms to the IFAS Invasive Plants Working Group in 2011, our choice of Pink Caprice as the resident species was accepted, and our genetic sterilization work on lantana received positive comments from the Invasive Plants Working Group. In subsequent ITP requests in 2016, Pink Caprice was also accepted as a resident species.

Below are some photos of invasive Lantana camara and Pink Caprice:

a) Invasive *Lantana camara*: Photo in the IFAS Assessment database (<u>https://plants.ifas.ufl.edu/wp-content/uploads/images/lanspe/lantana2.jpg</u>)



b) Invasive *Lantana camara*: Photo at the Atlas of Florida Plants website (<u>https://florida.plantatlas.usf.edu/Plant.aspx?id=1789#</u>)



c) Invasive *Lantana camara*: Photo at the Atlas of Florida Plants website (<u>https://florida.plantatlas.usf.edu/Plant.aspx?id=1789#</u>)



d) Invasive Lantana camara: Photo taken in Lithia Springs Park, Hillsborough County, FL



e) Invasive Lantana camara: Photo taken along Rock Road, Ft. Pierce, FL



f) Lantana camara Pink Caprice in Florida