



Research Article

Gall wasp *Leptocybe invasa* (Hymenoptera: Eulophidae) management in Eucalypts

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ABSTRACT: The invasive gall wasp *Leptocybe invasa* (Hymenoptera: Eulophidae) induces galls on the shoots of *Eucalyptus camaldulensis* Dehnh. and *E. tereticornis* Sm. and damage nursery seedlings young plantations and coppice shoots. The magnitude of the damage in nurseries and young plantations necessitates increased efforts to reduce the incidence and impacts of the pest. Identification of feasible and appropriate management methods will help in reducing the high cost of containing the pest in outbreak situations and avoid loss of planting material by state Forest Departments, Corporations and farmers while raising commercial *Eucalyptus* plantations. Deployment of gall tolerant clones and biological control agents has been attempted in the present study. Results show that eucalyptus gall wasp in nurseries can be managed by deploying identified gall tolerant, productive clones. Release of natural enemies like *Quadrastichus mendeli* and *Megastigmus* sp and regular monitoring of its population level in nurseries can further bring down the gall infestation.

KEY WORDS: Invasive species, eucalyptus, gall, biological control, parasite, resistance.

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INTRODUCTION

Leptocybe invasa Fisher & La Salle (Hymenoptera: Eulophidae) has emerged as the most serious pest of *Eucalyptus* outside Australia. Its outbreak has been observed since 2002 in the Mediterranean region (Mendel *et al.*, 2004), African countries (CABI 2007) and Asia (Jacob *et al.*, 2007; Thu 2004)). This gall wasp has been inflicting considerable damage in the eucalypts nurseries and plantation of *Eucalyptus*. The tiny adult wasp induces galls on shoot terminals, petioles and midribs in saplings and trees of eucalypts. This insect reproduces parthenogenically (Mendel *et al.*, 2004). Adult female insect lays eggs in young growing shoot tips besides midribs of juvenile leaves. The presence of cork tissues appearing at the egg insertion areas can be used to identify gall wasp infestation. Typical bump shaped galls start developing at these areas which become more visible after 40-45 days of egg laying. The larva develops inside the plant tissue within the larval chamber, deriving its nutrition from the surrounding plant tissue. Characteristically cylindrical galls develop on either young stems or petioles or midribs. Mature galls are

usually deep pink or red. Adult wasps emerge by cutting a hole through the gall wall after 3-4 months of growth inside the gall.

The minute size of the adult fly makes it impossible its detection in the field when the nucleus population is in the initial stage of breeding and establishing. The natural enemies keep them at a low profile in Australia and it is not a pest in Australia. Extensive plantations established using clones of *E. camaldulensis* in India facilitated successful establishment and spread of the wasp species in states of Tamil Nadu and Andhra Pradesh.

Several methods were adopted for control of the pest. Conventional control measure like sweep net collection, sticky traps, colour and light trap and pesticides were not successful due to its concealed nature (Jacob and Sivakumar 2012). Meanwhile variations in the incidence and intensity of gall in clones and seed origin plants have been observed (Jacob and Ramesh Kumar 2009; Thu *et al.*, 2009). Classical biological control was also reported successful in Israel (Kim *et al.*, 2008). The present study describes an

attempt to understand the variation in gall incidence on selected clones of eucalypts and efficiency of natural enemies in bringing down the gall incidence.

MATERIALS AND METHODS

Assessment of impact of gall damage on young nursery seedlings

Eucalyptus clones showing varying degrees of susceptibility to gall wasp infestation identified during multi location field level screening in plantation areas in earlier studies were selected and raised in Silviculture Research Nursery of Institute of Forest Genetics and Tree (IFGTB), Coimbatore, India nursery (Jacob 2009, Balu *et al.*, 2013). Highly susceptible clone IFGTB* -17, moderately susceptible clone IFGTB 69, and highly tolerant clone IFGTB -19 were selected for the study. Clones were compared in nursery in terms of percentage gall incidence and assessed every 10 days for 40 days. Incidence of galls was observed comparatively higher in seedlings since germination in nurseries till the end of one year in out planted saplings in plantation areas. Intensity of gall incidence comes down in plantation areas two years after planting. Percentage incidence of galls in nursery beds with 1000 seedlings in each bed was assessed based on the percentage of gall incidence per plant, following Jacob and Ramesh Kumar (2009): up to 30% — low, between 30% and 70% — medium, and >70% — high. Growth performance of young seedlings with different levels of gall damage was compared in terms of height. Girth of 1- 2 months old seedlings was not recorded as it is very minimal. Since gall incidence was more prevalent in the field during first year of growth after planting in the field, assessment of growth due to gall wasp infestation was carried out in clones IFGTB -17, IFGTB 69 and IFGTB -19 for height (cm) and girth variation (cm) for the first year of growth in the field.

Classical biological control

One thousand numbers of gall wasp infested, 20 days old eucalypts seedlings were shifted to net house and a nucleus culture of natural enemies imported from Israel were introduced into the net house. 15 adults each of *Quadrastichus mendeli* and *Megastigmus* sp. were released. Population assessment of natural enemies was done every 10 days by sweep net. Captured adults of natural enemies were counted and released back in the nursery. The counted galls were spotted red, using commercial nail varnish, to avoid erroneous re-counting. Percentage reduction in gall incidence was assessed every 10 day for 40 days. Beds where biocontrol agents were not released served as control.

Statistical Analysis

Data were subjected to one way ANOVA followed by Duncan's multiple-range test to assess the significance level.

RESULTS

Impact of gall damage on young nursery seedlings

Clear increase in percentage gall incidence was observed in IFGTB 17 and IFGTB 69 from 10th day to 40th day after germination in nursery. There was also significant variation in the gall incidence among the 3 clones. Gall tolerant clone IFGTB 19 recorded a minimal gall incidence of 4% on an average over a period of 40 days where as moderately susceptible clone (IFGTB 69) and highly susceptible clone (IFGTB 17) recorded 44% and 86% respectively. Growth performance in terms of height was significantly different among the three clones. Highest overall height gain during the 40 days period was in the moderately susceptible clone IFGTB 69 followed by highly susceptible clone IFGTB 17 and tolerant clone IFGTB19 (Table 1).

Classical biological control

The average number of parasite *Q. mendeli* decreased initially and increased 30 days after release. *Megastigmus* sp. showed an increase in population 40 days after release. There was decrease in the percentage gall incidence on seedlings 40 days after release of parasites. Percentage gall incidence in young meristems reduced from a level of 48% in the initial stages to 10.33% 40 days after release of parasites (Table 2).

Assessment of growth variation in seedlings due to gall wasp infestation

Comparison of gall infestation on the three clones at the end 40 days of growth showed that mean number of seedlings infested were more in highly susceptible (IFGTB 69) and moderately susceptible (IFGTB 17) clones. The proportion of the three infestation levels was also high in these clones. A similar trend was observed at the end of one year of growth (Table 3). Comparison of growth parameters showed variation between gall infested and uninfested seedlings in all the three clones. Highly susceptible clones recorded decrease in height and girth at the end of one year of growth. (Table 3).

DISCUSSION

Eucalyptus species in the introduced locations is colonized by many native and invasive species of insects causing significant levels of injury and damage. Insecticides

Table 1. Effect of clonal variation on gall incidence on nursery seedlings

DAT	Clone –IFGTB -17		Clone – IFGTB 69		Clone – IFGTB- 19		Sig.**
	Gall %	Height	Gall %	Height	Gall %	Height	
10	49.33	14.80	54.67	15.40	3.00	13.53	a
20	63.67	25.20	45.67	24.27	5.33	22.53	a
30	83.00	33.67	50.00	38.47	5.67	37.60	b
40	86.00	53.33	44.33	53.60	1.80	48.73	b
Overall	70.50	31.75	48.67	32.93	3.95	30.60	
Sig.**	c	2	b	1	a	3	

DAT- Days After Treatment *p<0.01 Letters within a column do not vary significantly

Table 2. Effect of biocontrol agents on gall incidence on nursery seedlings

DAT	<i>Q. mendeli</i>	<i>Megastigmus</i> sp.	% Gall incidence
10	1.87b	1.60b	48.00b
20	0.53b	0.53c	65.00b
30	6.33a	3.47b	50.33b
40	9.67a	10.47a	10.33a
Overall	3.35	4.02	43.42
Sig.	**	**	**

*p<0.01 Letters within a column do not vary significantly

Table 3. Gall incidence and growth parameters of Eucalyptus clones in Nursery

CLONE	After 40 days in nursery							
	Infected seedlings	Infestation Level			With Gall infestation		Without Gall infestation	
	Mean	LOW	MEDIUM	HIGH	Ht (M)	Girth (cm)	Ht (M)	Girth (cm)
IFGTB 69	13.5±0.6	45.94±2.3	38.44±1.3	15.6±1.2	0.50±0.04	0.90±0.02	0.52±0.01	0.42±0.05
IFGTB 17	12±0.5	50.4±3	66.1±2.5	74.8±1.3	0.36±0.01	0.81±0.04	0.44±0.01	0.26±0.01
IFGTB 19	2±0.3	8.8±0.8	4±0.7	0±0	0.68±0.02	0.84±0.01	0.69±0.02	0.43±0.05
CD	0.074	0.068	0.078	0.032	0.132	0.125	0.149	0.186
After one year in plantation								
IFGTB 69	18.20±0.6	42.52±2.0	28.52±	20.52±2.2	2.31±0.05	8.00±0.05	2.40±0.02	13.69±0.1
IFGTB 17	23.20±0.7	64.36±2.8	70.90±	84.14±2.8	1.74±0.01	8.03±0.07	1.82±0.01	11.97±0.09
IFGTB 19	6.00±0.4	10.12±1.2	8.34±	2.11±0.4	1.94±0.01	5.99±0.02	2.40±0.02	13.60±0.1
CD	0.098	0.058	0.081	0.038	0.152	0.184	0.232	0.195

Values represent mean ±SD

cannot be used in plantations in an outbreak situation due to environmental and financial implications. An alternative means of combating pest in such situation is through the use of genotypes resistant /tolerant to pests.

L. invasa (approximately originating from Australia) spread throughout India during 2006 to 2007 causing damage to Eucalyptus nurseries and plantations. In order to enhance productivity per unit area, superior quality eucalypts with high biomass productivity leading to maximum pulp yield has been raised by various government and private agencies besides farmers. Extensive plantations using such clones of *E. camaldulensis* facilitated fast spread of *L. in-*

vasa in the states of Andhra Pradesh and Tamil Nadu (Jacob 2009). Meanwhile variation in the incidence of gall and intensity of injury and damage existed among these clones then in use. Variation in the incidence among different seed source raised seedlings was also evident (Jacob and Ramesh Kumar 2009). Host plant resistance a component of a suite of other identified integrated control measures is an obvious candidate for development.

In the present study high level of gall incidence and damage was observed in susceptible clone like IFGTB 69, IFGTB 17 whereas minimum gall incidence was observed in resistant clone IFGTB 19. Presence of thick cuticle and

compactly arranged vascular bundle sheath was evident in gall tolerant seed origin plants of eucalyptus (Jacob and Ramesh Kumar 2009). Similar situation in clones tends to resist the wasp in laying more number of eggs in tolerant candidates. In the present study galls in some clones got aborted in between and no emergence of adult wasp was observed. Manifestation of aborted galls in certain varieties of *Mangifera indica* by cecidomyiid *Procontarinia matteiana* has been attributed to secondary resistance by way of blocking the sub cellular events leading to cell enlargement and division in those varieties which may be causing mortality of early stage larva (Githure *et al.*, 1998). Variations in the level of defoliation caused by larvae of autumn gum moth, *Mnesampela privata* (Johns *et al.*, 2002) and of leaf necrosis caused by larvae of leaf blister sawfly, *Phylacteophaga froggatti* was observed in different provenance of *E. globulus* in Australia (Farrow *et al.*, 1994). Similar variations in the levels of susceptibility of *Mangifera indica* varieties to the cecidomyiid *Procontarinia matteiana* (Sathianantham *et al.*, 1973; Rao *et al.*, 1991) infestations have been recorded in southern India and in South Africa (Schoeman *et al.*, 1996).

The overall height gained by the entire tolerant clones was on par with the susceptible clones 40 days after infestation. Though the height variation between infested and uninfested seedlings was not significant, a clear trend towards retardation in growth was observed in infested seedlings of clone at the end of one year growth. Infested growing shoot terminals and midribs enlarge in thickness and leaves present a contorted appearance. New leaves appearing from the gall infested shoot terminals are stunted. Disfiguration, rosette appearance and slow growth of growing shoot region and drooping of young saplings were observable in nurseries and plantations. After the emergence of the adult, the surrounding tissues, shoot terminals and leaves become dry leading to leaf shedding, retarded growth, poor stem form and loss of vigour in susceptible *Eucalyptus* clones and seedlings. Repeated attack and gall formation tends to cause a net decrease in the balance of nutrients leading plants to a stress situation and poor growth. Eucalyptus provenances which were more resistant to gum moth performed the best in terms of survival, height and growth (Farrow *et al.*, 1994).

There was significant decrease in the percentage gall incidence on seedlings 40 days after release of parasites. *Q. mendeli* tends to be an efficient parasitoid of *L. invasa* than *Megastigmus* sp. This is evident from the more number of parasites *Q. mendeli* collected 30 days after release compared to *Megastigmus* sp. which showed an increase in population from 40 days after release. The average number of days for development of *Q. mendeli* was 29 and 40 days for

Megastigmus sp. This is in agreement with results of Kim *et al.*, (2008) where it is reported that *Q. mendeli* develops faster than *Megastigmus* sp. Complete control has been achieved in the classical biological control of *L. invasa* in Israel with *Q. mendeli* (Kim *et al.*, 2008) and *Megastigmus* sp (Protosov *et al.*, 2008). In the present study also the combined effect of both the parasites had brought down the percentage gall incidence significantly after 40 days. This may account for the efficient parasitisation of *Q. mendeli* and *Megastigmus* sp on *L. invasa* induced galls in Eucalypts.

CONCLUSION

High level of gall infestation can interfere with the growth performance of seedlings in nursery. From the present study it is clear that eucalyptus gall wasp in nurseries can be managed by deploying identified gall tolerant, productive clones. Deployment of natural enemies like *Quadrastichus mendeli* and *Megastigmus* sp and regular monitoring of its population level in nurseries can further bring down the gall infestation. Moderately susceptible clones can also be included to sustain the parasite population in nurseries. Integrating deployment of gall tolerant clones and release of biocontrol agents can be used by farmers, tree growers and forest based industries in their nurseries to significantly reduce the high cost of containing the gall wasp outbreak situations and to avoid loss of planting material. Deployment of mixed clones will help to avoid epidemic outbreaks of pests as well as to gain / stabilize the economic loss resulting due to pest damage when limited clones are deployed.

DEDICATION

Fondly dedicated to my teacher T N Ananthakrishnan, who turns 90 on 15 December 2015.

REFERENCES

- Balu A, Jayaraj RSC, Vimal P, Rajarishi R, Mahalakshmi R, Thangapandian K, Deeparaj B, Eswaran S, Senthilkumar P, Kalaiselvi R. 2013. Screening of productive clones for tolerance to the gall pest, *Leptocybe invasa* Fisher & La Salle (Hymenoptera: Eulophidae) In: Balu, A., Jayaraj, R. S. C., Regupathy, A., Mohan, V., Rekha Warriar, Raghunath, T. P. and Krishnakumar, N. (Eds) Forest Health Management. IFGTB, Coimbatore, Tamilnadu, India. 395–410.
- CABI 2007. *Leptocybe invasa* Fisher & La Salle. Distribution Maps of Pests. 698,2pp.
- Farrow RA, Floyd RB, Neumann FG. 1994. Inter provenance variation in resistance of *Eucalyptus globulus* juvenile foliage to insect feeding. *Aus For.* **57**: 65–68.

- Githure CW, Schoeman AS, Mc Geoch MA. 1998. Differential susceptibility of mango cultivars in South Africa to galling by the mango gall fly *Procontarinia matteiana* Kieffer and Cecconi (Diptera: Cecidomyiidae). *Af Entomol.* **6**: 33–40.
- Jacob Prasanth J, Devaraj R, Natarajan R. 2007. Outbreak of the invasive gall-inducing wasp *Leptocybe invasa* on *Eucalyptus* in India. *Newsletter for the Asia-Pacific Forest Invasive Species Network.* **8**: 4–5.
- Jacob PJ, Ramesh KA. 2009. Incidence of galls induced by *Leptocybe invasa* (Hymenoptera: Eulophidae) on seedlings of *Eucalyptus camaldulensis* and *E. tereticornis* (Myrtaceae) from different seed sources in Southern India. *Int J Ecol and Env Sci.* **35**: 187–198.
- Jacob PJ. 2009. Invasive insect pest *Leptocybe invasa* in Eucalyptus plantations in India. *ENVIS For Bull.* **9**: 65–70.
- Jacob PJ, Siva K. 2012. *Development of appropriate integrated management methods for the Eucalyptus Gall wasp problem in nurseries.* Project Completion Report Submitted to Indian Council of Forestry Research and Education, Dehradun. 26 pp.
- Jones TH, Potts BM, Vaillancourt RE, Davies NW. 2002. Genetic resistance of *Eucalyptus globulus* to autumn gum moth defoliation and the role of cuticular waxes. *Can J For Res.* **32**: 1961–1969.
- Kim Il-Kwon, Zvi M, Alexey P, Daniel B, John LS. 2008. Taxonomy, biology, and efficacy of two Australian parasitoids of the Eucalyptus gall wasp, *Leptocybe invasa* Fisher & L Salle (Hymenoptera: Eulophidae: Tetrastichinae). *Zootaxa.* **1910**: 1–20.
- Mendel Z, Protasov A, Fisher N, La Salle J. 2004. The taxonomy and natural history of *Leptocybe invasa* (Hymenoptera: Eulophidae) gen & sp. nov., an invasive gall inducer on *Eucalyptus*. *Aus J Entomol.* **43**: 101–113.
- Protasov A, Doganlar M, Salle JL, Mendel Z. 2008. Occurrence of two local *Megastigmus* species parasitic on the Eucalyptus gall wasp *Leptocybe invasa* in Israel and Turkey. *Phytoparasitica.* **36**: 449–459.
- Rao PK, Kasi V, Parvathamma K, Rao DVR. 1991. Relative susceptibility of different mango varieties to leaf gall fly *Procontarinia matteiana* Keifer and Cecconi (Cecidomyiidae: Diptera). *Ind J Entomol.* **53**: 449–452.
- Sathianantham VKR, Abraham EV, Bettai Gowder R, Thirumurthi S, Rajappan PV, Subramaniam TR. 1973. Relative resistance and susceptibility of different mango varieties to cecidomyiid galls. *South Ind Hortic.* **21**: 51–55.
- Schoeman AS, Mc Geoch MA, Githure CW. 1996. *Differential susceptibility of eleven mango cultivars to galling by the mango gall fly (Procontarinia matteiana Kieffer and Cecconi (Diptera: Cecidomyiidae)).* South African Mango Growers' Association Year Book **16**: 23–26.
- Thu PQ, Dell B, Burgess TS. 2009. Susceptibility of 18 eucalypt species to the gall wasp *Leptocybe invasa* in the nursery and young plantations in Vietnam. *Science Asia.* **35**(2009): 113–117.
- Thu PQ. 2004. The first record of gall forming wasp associated with eucalypt plantations in Vietnam. *Sci and Tech J of Agri and Rural Dev.* **11**: 1598–1599.