# Pollination of a cultivated fig, *Ficus pumila* var. *awkeotsang*, in South China

Yong Chen<sup>1\*</sup>, Hong-Qing Li<sup>2</sup>, Shao-Jiang Ruan<sup>1</sup>, and Wei-Liang Ma<sup>2</sup>

<sup>1</sup>Department of Biology, Ningde Teachers College, Ningde, Fujian 352100, China, Email. cytx2007@126.com;

<sup>2</sup>School of Life Science, East China Normal University, Shanghai 200062, China

(Received February 20, 2007; Accepted May 17, 2007)

#### Abstract

Fruits of Ficus pumila var. awkeotsang can be made into jelly, which has been a special delicacy in South China for centuries. We studied the potential for cultivation of F. pumila var. awkeotsang. Young cuttings of F. pumila var. awkeotsang were planted in Ficus pumila var. awkeotsang Cultivation Garden, Dayangke Mountain (long. E 119°03′, lat. N 25°42′, altitude 420 m). They established easily and plants reached reproductive size after just 3 years. However, although wild populations of F. pumila var. awkeotsang existed 35 km from our plot, and populations of another variety, F. pumila var. pumila, thought to be pollinated by the same wasp, occurred at the site, pollination success was initially zero. Pollination experiments indicated that F. pumila var. awkeotsang is reproductively isolated from F. pumila var. pumila, and that the pollinator of the latter cannot breed in the syconia of the former variety. Deliberate introduction of wild pollinators led to the establishment of a pollinator population in the cultivated plot containing 60 male plants. However, syconia abortion rates were still high (>50%) after four years. This was partly due to the seasonal phenology of F. pumila var. awkeotsang and the low overlap between wasp reproduction and the receptive phases in the fig. We expect pollination rates to increase as the plants get larger, or if more male fig plants are established.

Keywords: Ficus pumila var. awkeotsang, Wiebesia pumilae, pollination biology, cultivation

## 1. Introduction

Ficus pumila L. belongs to section Synoecia, but recent molecular data indicate it should be placed in section Ficus (Rønsted et al., 2008, this volume). In South China, Ficus pumila var. awkeotsang is a climbing fig that grows on hillsides above 400 m altitude, whereas F. pumila var. pumila is usually found below 400 m. In some areas their distribution overlaps. Ficus pumila var. awkeotsang is endemic to northern Fujian, southern Zhejiang, and Taiwan.

The pollinator of *F. pumila* var. *awkeotsang* was described as *Wiebesia pumilae* by Hill (1967), and has been regarded as the same wasp species that pollinates *F. pumila* var. *pumila* (Ma, 1989; Kjellberg et al., 2001). Morphological traits and molecular data now place *Wiebesia pumilae* in the genus *Blastophaga*. Wasps in this genus pollinate other figs in section *Ficus* which fits in with the new placement of *Ficus pumila* in this section (J.-Y. Rasplus, personal communication).

No other wasps have been collected from the syconia of *F. pumila* var. *awkeotsang* (Ma, 1989). The fruit of *F. pumila* L. var. *awkeotsang* has been used to make an edible jelly (Huang et al., 1979), or a popular natural drink, traditional Chinese delicacies, for over 180 years. Cultivation of *F. pumila* var. *awkeotsang* began in Taiwan at the end of last century, following a sharp decline in wild populations of this fig. Recently, trial cultivation and related research was initiated in Fujian, China where *Ficus pumila* var. *awkeotsang* is expected to become an economically important species.

In this paper, we describe how a cultivated plot of *F. pumila* var. *awkeotsang* was set up in South China in 1996, and how successful fruit production was established.

## 2. Material and Methods

The *Ficus pumila* var. *awkeotsang* Cultivation Garden, Dayangke Mountain (long. E 119°03', lat. N 25°42', altitude 420 m) has a subtropical oceanic climate with four clear seasons. Cuttings of *F. pumila* var. *awkeotsang* 

<sup>\*</sup>The author to whom correspondence should be sent.

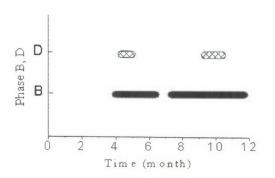


Figure 1. Phenological observation of cultivated Ficus *pumila* var. *awkeotsang*.

were imported from Taiwan and planted in the garden in spring of 1996. From these, 259 female plants and 60 male plants were established. A 4 m tall column of stones was used as the structure for each plant to climb over. The growth and performance of the plants was monitored.

In 2000, 15 male and 15 female plants were selected for pollination studies. We planned to introduce pollinators from a wild population of *F. pumila* var. *pumila*, located at Nanshan Mountain of Ningde City, Fujian Province (long. E 119°37', lat. N 25°42', altitude 450 m) by transplanting *F. pumila* var. *pumila* to the *Ficus* Cultivation Garden, Dayangke Mountain. Thirty wild plants of *F. pumila* var. *awkeotsang* and *F. pumila* var. *pumila* were marked. From 2001 until 2004, we followed the phenology of these plants and tagged all the receptive syconia.

In order to test whether the wasps dispersing from the syconia of F. pumila var. pumila can survive in the syconia of F. pumila var. awkeotsang, we conducted bagging and introduction experiments. We bagged 80 syconia of F. pumila var. awkeotsang (30 male, 50 female) before the receptive phase. The bags were left on ten male and ten female syconia as no-pollination controls. For natural pollination controls, we removed bags and replaced then again, after one naturally arriving wasp had entered the syconium (male n=10, female n=10). Using a medical syringe, we artificially pollinated female syconia with pollen from F. pumila var. awkeotsang and F. pumila var. pumila (n=10 for each variety). We also artificially introduced wasps, emerging from a F. pumila var. pumila syconium, into ten male syconia of F. pumila var. awkeotsang using one wasp per fig. To do this we took a glass tube of 5 mm in diameter, heated it and drew it out to make a fine pointed tube. The tube was marked with a ring at the spot where the diameter equaled 2 mm near the sharp end, which was left in place. The sharp end of the glass tube was inserted into receptive-phase syconia until the girdling ring was just within the syconia cavity. The tube was then broken into two parts and the sharp end pulled out of the fig. A single wasp was then introduced into the  $\emptyset$  5 mm end of the glass tube and blown into the syconia cavity. The wounds in the syconia automatically sealed and repaired themselves. We found in our experiments that introducing one wasp was sufficient to prevent syconia from aborting.

In a separate trial, wild male *F. pumila* var. *awkeotsang* syconia at the wasp emerging phase were hung in the cultivation plot. The trial was repeated twice in 2001. Fifty syconia were introduced to the plot each time. We recorded the abortion rate of syconia on ten sample trees, over the subsequent 4 year period. In 2004, we sampled 10 syconia of each sex in the cultivated plot to assess the seed- and gall-set.

## 3. Results

The cultivated plants grew quickly, the height of year-old plants being 80–100 cm, at two years >180 cm, and in the third year they reached 250 cm. They then started to produce syconia. And the vegetative growth slowed down. Vegetative branches had adventitious roots and small (2–3 cm long) heart-shaped leaves that lay flat against the stones. Branches with syconia did not have adventitious roots and had large wide lanceolate-shaped leaves (7–12 cm long), forming a bushy crown with syconia developing in the leaf axils.

Observations on the 30 cultivated and 30 wild *F. pumila* var. *awkeotsang* and the 30 wild *F. pumila* var. *pumila* sample trees in 2001 revealed that both *F. pumila* var. *awkeotsang* and *F. pumila* var. *pumila* had two peaks of inflorescence production in a year, in spring and in autumn (Fig. 1). In spring, both intra- and inter-plant synchrony was obvious, especially in the wild populations. In the autumn inflorescences, there was far greater asynchrony. Female plants of *F. pumila* var. *pumila* bore syconia only in spring, but the male plants bore syconia in both spring and autumn. The autumn crop supports pollinating wasps over the winter and this has also been reported for *F. carica* (Kjellberg et al., 1987).

We found that 1) bagging effectively stopped wasps from entering receptive phase syconia; 2) In *F. pumila* var. *awkeotsang* a single naturally-arriving foundress, can result in the production of 1328±199 fruitlets and 610±74 galls in female and male syconia respectively; 3) artificial pollination of female syconia with pollen from *F. pumila* var. *awkeotsang* led to successful and substantial seed set. However, pollen from *F. pumila* var. *pumila* resulted in zero seed set; and 4) induction of wasps from the syconia of *F. pumila* var. *pumila* to male syconia of *F. pumila* var. *awkeotsang* failed to produce any galls (Table 1). These results indicate that there is reproductive isolation between *F. pumila* var. *pumila* and *F. pumila* var. *awkeotsang*, and that the pollinating wasps of *F. pumila* var. *pumila* cannot reproduce in the syconia of *F. pumila* var. *awkeotsang*.

Table 1. Results of different pollination treatments for the syconia of *F. pumila* var. *awkeotsang*. The number of seeds and galls produced in female and male syconia, respectively (n=10 for all treatments).

Treatment	No. of seeds (Mean±SD)	No. of galls (Mean±SD)
Bagging, no pollinators permitted to enter	0±0	0±0
One naturally arriving pollinator permitted to enter	1328±199.4	610±74.5
Artificial pollination with pollen from F. pumila var. awkeotsang	11273±710.7	_
Artificial introduction of pollinator emerging from F. pumila var. pumila	-	0±0
Artificial pollination with pollen from F. pumila var. pumila	0±0	=

Following the introduction of pollinators from wild *F. pumila* var. *awkeotsang* populations to the cultivated plot in 2001, the abortion rate of syconia decreased each year. The retained figs were 11.14% in 2001, 39.21% in 2002, 43.71% in 2003 and 50.75% in 2004. The data are the average for the spring and autumn production seasons in each year. After 4 years still about 49% of the figs, which contrasts with our observations on wild populations of *F. pumila*. var. *awkeotsang*, where less than 1% of syconia aborted.

In the Spring of 2004, we assessed the seed- and gall-set in pollinated syconia in the cultivated population of F. pumila var. awkeotsang. From ten female syconia, we found on average that  $11394\pm651$  flowers were formed that produced  $7320\pm1525$  seeds (64%). In male syconia there was on average of  $13153\pm386$  gall flowers that produced  $8463\pm3049$  galls (64%), and there were  $1360\pm88$  male flowers (n=10). The average foundress number is  $4.2\pm0.5$  either in male or in female syconia (n=10, respectively). In 2004, we also monitored the overlap of reproductive phases in the cultivated population. Receptive syconia are available for about 70 days but the wasp emergence phase only lasted 20 days in spring; hence there was only a 30% overlap. The overlap was even less in autumn being only 23%.

## 4. Discussion

Successful cultivation of *F. pumila* var. *awkeotsang* depends on establishing the correct conditions for vegetative growth and pollination of the syconia. We found that *F. pumila* var. *awkeotsang* grew easily from cuttings and reached reproductive size after just three years. However, initially 100% of syconia aborted through pollination failure. Investigation of the pollination system established that *F. pumila* var. *awkeotsang* is reproductively isolated from *F. pumila* L. var. *pumila* and the pollinators of *F. pumila* L. var. *pumila* and the pollinators of *F. pumila* L. var. *awkeotsang*. Thus, wild populations of *F. pumila* L. var. *pumila* cannot supply pollinators for the cultivated *F. pumila* var. *awkeotsang*.

Wild populations of F. pumila var. awkeotsang can be found 35 km from our site, but its wasps cannot disper e such a long distance. This is in accordance with Harrison's hypothesis that dispersal range in many dioecious fig pollinators may be more restricted than that of monoecious fig pollinators (Harrison, 2003; Harrison and Rasplus, 2006). Thus, successful pollination of the cultivated population at the Dayangke Mountain Ficus Cultivation Garden depended on deliberate introduction of pollinators from a wild population. This was done twice in Spring 2001 and the pollinator population persisted until the end of observations in 2004 within our cultivated plot of 60 male trees. However, although syconia pollination rates increased each year from 2001 to 2004, abortion rates were still high (about 49%) after four years. This suggests that a larger cultivated population of male plants is required to maintain high rates of pollination (Kameyama et al., 1999).

F. pumila var. awkeotsang was found to have a highly seasonal phenology similar to that reported for F. carica (Kjellberg et al., 1987). This involves spring pollination of female syconia and a special over-wintering crop produced on male figs in the autumn. We found that there was little overlap in the cultivated plot between the wasp emerging phase and receptive phases in both spring and autumn. This contributed to high syconia abortion rates. The initiation of syconia is staggered in F. pumila var. awkeotsang and, hence, as the plants grow larger we expect the period of syconia production will be extended, as has been reported for other dioecious figs (Harrison and Yamamura, 2003), which should lead to great overlap in the wasp emerging and receptive phases and thus increased pollination rates.

Reproduction of natural *F. pumila* var. *awkeotsang* population depends on seed germination and this keeps the sex ratio at about two. Furthermore, in the wild populations, there is a large overlap between female and male-phase syconia and this results in high fruiting rates. In cultivated populations, owing to the lower overlaping of female and male-phase syconia, there is little to boost the tree's reproductive ability even if more males are planted. The best option may involve selecting cultivars with an improved synchronism.

## REFERENCES

- Harrison, R.D. 2003. Fig wasp dispersal and the stability of a keystone plant resource in Borneo. *Proceedings of the Royal Society London B* **270**: S76–S79.
- Harrison, R.D. and Rasplus, J.-Y. 2006. Dispersal of fig pollinators in Asian tropical forests. *Journal of Tropical Ecology* 22: 631-639.
- Harrison, R.D. and Yamamura, N. 2003. A few more hypotheses for the evolution of dioecy in figs. *Oikos* 100: 628–635.
- Hill, D.S. 1967. Fig-wasps (Chalcidoidea) of Hong Kong I Agaonidae. Zoologische Verhandlungen 89: 355.
- Huang, Y.-C. and Chen, W.-B. 1979. On the material plant of awkeo-jelly: Ficus awkeotsang Makino its historical review and future prospects. Chinese Horticulture 25: 103-111.
- Kameyama, T., Harrison, R.D., and Yamamura, N. 1999. Persistence of a fig wasp population and evolution of dioecy in figs: A simulation study. Researches on Population Ecology 41: 243–252.

- Kjellberg, F., Gouyon, P.H., Ibrahim, M., Raymond, M., and Valdeyron, G. 1987. The stability of the symbiosis between dioecious figs and their pollinators: A study of *Ficus carica* L. and *Blastophaga psenes* L. *Evolution* 41: 693–704.
- Kjellberg, F., Jousselin, E., Bronstein, J.L., Patel, A., Yokoyama, J., and Rasplus, J.-Y. 2001. Pollination mode in fig wasps: the predictive power of correlated traits. *Proceedings of the Royal Society of London B* 268: 1113–1121.
- Ma, W.-L. and Wu, X. 1989. A preliminary study of symbioses between *Blastophaga pumilae* Hill (Hymenoptera) and *Ficus pumila* L.(Moraceae). *Acta Ecologica Sincia* 9: 914 (in Chinese).
- Rønsted, N., Weiblen, G.D., Clement, W.L., Zerega, N.J.C., and Savolainen, V. 2008. Reconstructing the phylogeny of figs (*Ficus*, Moraceae) to reveal the history of the fig pollination mutualism. *Symbiosis* **45**: 45–55.