Hybridization and Cutting of *Buddleja* Genus

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Abstract: *Buddleia davidii*, also known as butterfly bush, is a unique species in China, with extensive cultivation and promotion values. The purpose of cross-breeding is to promote environmental adaptation and improve its economic benefits as a garden plant. As *Buddleja* is asexual, cutting can aid in mass production of the hybrid offspring, thus stabilising local ecosystem.

1 Introduction

Buddlejoideae, under the family of *Scrophulariaceae*, is a widely cultivated and extremely popular flowering shrub with attractive foliage, diverse colours and strong aroma. As a member of *Buddlejaceae*, the genus *Buddleja* has about 100–150 subspecies (e.g. *Buddleja davidii*, *B. lindleyana*, *B. japonica*, *B. × weyeriana*) that is distributed in north and south America, south and east Africa, as well as south-central and southeast Asia (Sandra B. Wilson *et al.* 2004). There are about 100 species in the world and about 23 species in Asia, of which 21 species are distributed in China (Chen *et al.*, 2007). Out of these species, *B. davidii* Franchet is the most common.

B. (Synonyms. Buddleia davidii; also known as butterfly bush) is a perennial, semi-deciduous, multi-stemmed shrub that readily establishes in disturbed areas of temperate, subtropical, and tropical climates. Native to central and southwestern China, it could reach up to 3,500 meters above sea level. (Tallent-Halsell & Watt, 2009).

B. davidii has been introduced to the Americas, Australia, Europe, and New Zealand as an ornamental for its fragrant and showy flowers. It is a widely cultivated as a popular garden plant for its economic values to the horticultural industry. Certain *B. davidii* cultivars are worth over \$200,000/year to plant growers in Georgia, USA, (Tallent-Halsell & Watt, 2009). In addition to the aesthetic and fragrant appearance of *B. davidii*, the flowering shrub has been closely related to butterflies, moths, and hummingbirds (Laere *et al.* 2009). There are also certain species of *Buddleja* which have been used as folk medicine for a variety of illnesses for centuries. In addition, the leaves of some species of *Buddleja* have been used for centuries for fishing in China as they have proven detrimental to the fish (Tallent-Halsell & Watt, 2009). W

However, many countries having introduced *B.* start to consider it as invasive and problematic. There are concerns that it has potential negative and irreversible

impacts on local agriculture and wild lands (Tallent-Halsell & Watt, 2009).

The use of native plants as ornamental would help avoid invasions from exotic species. The local ecosystem of China as a whole consists of biocenosis and the environment they live in, which could adjust and organize itself to maintain balance through adaptation and development. Failures of introduced species to adapt to the local ecosystem and potential unrestricted growth factors may eventually threaten other species, breaking down balance of the local ecosystem (Shi & Liu, 2013).

2 Aim of the research

The idea of using native plant is important. Therefore, *Buddleia davidii*, as a local species of China, is worth promoting. The paper also I focused on cross-breeding of this genus and asexual reproduction through hybridization and cutting.

The purpose of cross breeding aims at facilitating the adaption to the external environment and improve its economic benefits as a garden plant by perfecting its appearance. Cutting can help mass-produce hybridization as it is asexual, thus stabilising the local ecosystem.

3 The previous research and development of the breeding and cutting of *Buddleja* genus:

Breeding:

Starting as early as 1920, *Buddleja davidii* breeding programs have been focusing on the scale of breeding and diverse colours under harsh conditions for plantation (Tallent-Halsell & Watt, 2009). Meanwhile, the hybridization programs are concentrating on the traits of flower colours, inflorescence morphology, compact growth habits and grey pubescent foliage.

These breeding programs generally include intergeneric hybridization, physical breeding, bioengineering, and chemical breeding.

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Firstly, in terms of intergeneric hybridization, W. van de Weyer developed interspecific hybrids in 1920 from crosses between *B. globosa* and *B. magnifica*, which was classified as *B. ×weyeriana* and generally characterized by followers with colours between yellow and orange in interrupted patterns along the inflorescence length. (Tallent-Halsell & Watt, 2009). Another hybrid was discovered by Raymond Moore in 1949 between diploid *B. madagascarensis* Lam. and the diploid *B. crispa*. Hybrids from this intersectional cross resulted in polypanicles of dense orange flowers that retained the indehiscent fruit characteristic. In addition, a controlled cross between *B. davidii* var. *nanhoensis* (Chitt.) Rehd. ‘Nanho Purple’ and *B. lindleyana* Fort. ex Lindl. in 2004 produced a hybrid exhibiting characteristics of both parents as well as a number of intermediate traits. Flowers tend to be larger than either parent with an extended blooming period. The hybrid grows better at lower temperatures with fewer leaves dropped in winter and more flowers in blossom (Dunn & Lindstrom, 2007)

Secondly, gamma radiation has been introduced in University of Georgia as a physical breeding technique to induce sterility in *B. davidii* cultivars (Tallent-Halsell & Watt, 2009).

Thirdly, bioengineering has played a significant role in breeding development. Researchers used genetic engineering techniques to create hybrids with dwarf or non-functional reproductive organs or heavier seeds that are not carried by the wind. In addition, Lindstrom have sought to reduce potential invasions by producing hybrid varieties that either alter plant morphology or have an odd ploidy number (Tallent-Halsell & Watt, 2009). Introgression of flower colour in *B. davidii* is made possible by polyploidisation and interspecific hybridisation. There was an interspecific breeding programme with *B. globosa* that introduced yellow in the commercial *B. davidii* assortment through introgression.

Interspecific crosses between chromosome doubled *B. globosa* genotypes and *B. davidii* produced a progeny with yellow flower colour resembling *B. davidii*, which offers new opportunities for *Buddleja* breeding programmes (Laere *et al.*, 2011).

Finally, for chemical breeding, a study in 2007 determined that oryzalin could be used to restore fertility and alter morphological traits such as flower size or growth habit of other *Buddleja* species as longer and more intense exposure could lead to greater chromosome doubling potential (Dunn & Lindstrom, 2007)

Cutting:

There are a number of cutting programs of *Buddleja* in China.

Liu *et al.* (2008) conducted orthogonal experiments on cutting propagation of four *B. davidii* breeding lines. The result showed that all the four lines belonged to the lenticel rooting family. In the two media, the soil mixture composed of clay and sand in equal proportion + perlite had a positive effect on rooting rate while peat soil+perlite had a positive effect on rooting number. The study shows that the root number is positively correlated with rooting length.

Xu (2015) investigated three substrates and two rooting agents, and carried out cutting experiments on *B. davidii* by random combination. The results showed Peat soil: sand (1:1) matrix and Guoguang rooting agent works more effectively for cuttings breeding of *B. davidii*.

Zhang *et al.* (2017) studied the suitable cutting matrix of *Buddleja* by experimenting with different ratios of vermiculite, perlite, sand and soil. The results showed that the strong permeability of sand is beneficial to the growth of root elongation due to strong permeability of perlite and strong water conservation in soil. The combination of the two can increase the number of rooting and maintain moisture to increase the survival rate.

4 Research Hypothesis

- Hybridization of *Buddleja*

7. HC 5	×	JY 5	=	<p>Leaf: dark green white-hairy narrow ovate dentate weak</p> <p>Flower: lilac conical sparse horizontal free</p>

8. DY 54	×	ZH 3	=	<p>Leaf: white-hairy narrow ovate dentate strong</p> <p>Flower: white cylindrical dense horizontal free</p>

9. DY 52	×	HC5	=	<p>Leaf: white-hairy medium ovate entire strong</p> <p>Flower: lilac globular dense horizontal touching</p>

10. BA 2	×	ZH3	=	<p>Leaf: dark green white-hairy lanceolate dentate strong</p> <p>Flower: lilac globular sparse semi erect free</p>

11. BA 2

×

MX9

=

Leave: white-hairy
lanceolate
dentate

Flower: white-hairy
yellow
globular
dense
free

12. BA 2

×

YY 1

=

Leave: dark green
white-hairy
tiny ovate
sinuate
weak

Flower: modena
globular
sparse
eret
free

13. YY 1

×

MX 8

=

Leave: white-hairy
lanceolate
sinuate

Flower: yellow
conical
sparse
eret
free

- Cutting of 5 different F1 hybrids of *Buddleja*

5 Research plan and schedule

WORK LIST	WEEK 1	WEEK 2	WEEK 3	WEEK 4
THESIS PROPOSAL	✓	✓		
TEST THE RHS COLOUR VALUE	✓			
MEASURING	✓			
PUTTING ON BAGS	✓			
POLLINATION		✓	✓	
CUTTING			✓	✓

THESIS PROPOSAL

✓

6 Result Predictions

- 65% of these pollinated ovaries will expand
- Collect seeds from this batch of hybrids
- Obtain 50% seedlings from the cuttings

7 Results

- By August 22, 2018, 62% of these pollinated ovaries expanded.

DATE	FEMALE PARENT	MALE PARENT	NO.	MARK	COMMENTS	SECCE D OR NOT	FERTILISED OVARY NO.	PERCENT AGE %
24/07/2018	BD 3	YY 1	60			yes	12	20
24/07/2018	DY 54	BA 2	48			no	0	0
24/07/2018	ZH 3	BA 2	13			no	0	0
24/07/2018	ZH 3	BA 2	9			no	0	0
24/07/2018	YY 1	BA 2	1			no	0	0
24/07/2018	YY 1	BA 2	1			no	0	0
24/07/2018	MX 9	BA 2	3			yes	2	7
24/07/2018	YY 1	MX 8	1			no	0	0
25/07/2018	ZH 3	DY54	26		raining	yes	20	77
26/07/2018	BD14	DY59	185		DY 59 little pollen	no		
26/07/2018	MX 8	BD14	4			yes	3	75
26/07/2018	MX 8	BD14	5			yes	2	40
26/07/2018	C 60	HC 22	25	green	HC 22 the flowering time has passed	yes	23	93
26/07/2018	HC 22	C60	5	green	HC 22 the flowers are small	yes	1	20
26/07/2018	JY 5	DY 52	23	green				
26/07/2018	DY 52	JY 5	15	green		yes	14	93
26/07/2018	HC 5	DY 52	15	green		yes	13	87
26/07/2018	DY 52	HC 5	27	green		yes	23	85
26/07/2018	HC 5	JY 5	21	red		yes	15	71
26/07/2018	JY 5	HC 5	26	red				
27/07/2018	MX 8	BD 3	3	green	MX 8 little inflorescence and the flowers are small	yes	1	30
27/07/2018	DY 54	BD 3	22		DY 54 many insects	yes	20	90
27/07/2018	ZH 3	BD 3	6			yes	4	66
27/07/2018	C 60	BD 3	35	green		yes	35	100
27/07/2018	MX 8	ZH 3	8	green	ZH 3 little pollen	yes	6	75
27/07/2018	YY 1	ZH 3	1	red	ZH 3 little pollen	no	0	0
27/07/2018	YY 1	ZH 3	2	red	ZH 3 little pollen	no	0	0
27/07/2018	C 60	ZH 3	26	green	ZH 3 little pollen	yes	26	100
27/07/2018	ZH 3	DY 54	14	red		yes	13	93
27/07/2018	MX 8	DY 54	13	red		yes	11	85
27/07/2018	C 60	MX 8	24	red		yes	23	96
			433				267	62

8 Conclusion

The table of results shows that as of August 22, 2018, 62% of the pollinated ovaries had expanded, as expanded. However, weather conditions and unavoidable states of the flowers could hinder pollination. Therefore, *Buddleja* is likely to expand further in more parts of China. This process of crossbreeding could facilitate *Buddleja's* adaption to the environment and boost its economic use as a garden plant, thus stabilising the local ecosystem.

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