STUDIES ON THE FRUCTIFICATION OF "SHINANO WALNUT", A STRAIN OF JUGLANS REGIA L.*

[]. Morphology and Ecology of the Pistillate Flower and the Cluster

By

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INTRODUCTION

So-called Shinano walnut, a strain of *Juglans regia* L, has been cultivated in Japan. It is said as the hybrid between the ancient variety (*J. regia* L. var. *orientis* Kita.) and some varieties of Persian walnut (*J. regia* L.). Therefore, the essential characters resemble closely those of Persian walnut, though the both strains more or less differ from each other in detailed points.

Since many years ago the author has investigated the morphology and the ecology of Shinano walnut, especially on the fluctification.

The flowering period and dichogamy, the habit of shoot, the morphalogy and ecology of the staminate catkin and the pollen were reported in the previous reports (Machida, 1952, 1955, 1974). In the present paper the author will introduce a part

^{*} This study was presented as a dissertation for the doctor's degree at Tokyo University of Agriculture. The present paper is a part of it.

of studies on the fluctification, in which the morphology and ecology of the pistillate flower and the cluster are contained.

Before going further the author wishes to acknowledge the indebtedness to Dr. S. MATSUBARA for his cordial guidance throughout the study. Thanks are also due to Dr. Y. ASAMI, Dr. N. SUGIYAMA and Dr. M. IWATA for their kind advices and supports for the study. Further the author desires to express his gratitude to Dr. N. KOYAMA, Mrs. Y. IKEDA and the other friends who assisted the experiment.

RESULTS

1. Florescence and Receptive period

Approaching to the flowering time in spring, the budding flower comes in sight at the tip of the newly grown shoot. The duration from germination to flowering is commonly $7\sim10$ days, differed according to individual tree and influenced by the weather.

The author observed the florescence of the pistils prior to an artificial pollination, and divided the progress of the florescence into 8 stages as shown in the following $(A \sim H, Plate 1)$.

- A stage : Young stigma enclosed by sepals at top of ovary.
- B stage : Young stigma grows a little, but not visible though sepals loosened.
- C stage : After 1~2 days from B stage. Sepals opened and a part of stigma comes out. The author judged this stage as the beginning of flores-cence.
- D stage : After1~2 days from C stage. Stigma begins to open, and young glanduler pleat appears in inner surface of stigma. The author decided this stage as the early stage of flowering.
- E stage : After 2~3 days from D stage. Stigma elongates and extends with developement of pleats, having a polish by secretion of liquid. The author decided the stage as the middle stage of flowering.
- F stage : After 2~3 days from E stage. Stigma appears completely, and its surface becomes highest in polish, bending itself back at tip. The author named this stage the best stage of flowering.
- G stage : After $2\sim3$ days from F stage. Stigma loses polish and dried. Here and there discoloration occurs in pleats. The author called the stage the last stage of flowering.
- H stage : After $1\sim 2$ days from G stage. Polish of stigma fades away entirely, discoloration extends, and dryness goes in periphery. Stigma seems to lose its receptibity. The author judged the stage as the end of

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florescence.

The progressive stages mentioned-above expand in the florescence of one pistil. It is important to ascertain the most receptive stage of the stigma for conduction of an artificial pollination.

Then, the following method was adopted.

Method : Many pistils at A or B stage were enclosed with paraffin-paper bags to exclude the pollen from the stigmas. When the pistils reach each appointed stage (B, $B_1 \sim B_3$), they were pollinated by the pollens which were shed in the previous day with a hair-pencil.

The test sections are as follows.

A, Control section : Pistils were pollinated freely.

B, Artificial pollination section :

B_i, Early pollination sect. : Pistils were pollinated at D stage.

 B_2 , Middle pollination sect. : Ditto at E or F stage.

B₃, Last pollination sect. : Ditto at G stage.

C, Non-pollination section : Pistils were enclosed with paper bags from A stage to H stage.

The results of the above test are shown in Table 1.

Tree		A	B_1	B_2	B_3	С
	No. of pistils	21	62	39	66	57
No. 9	19, June	71.4	43.5	61.5	21.2	8.8
	24, July	66.7	40.3	48.7	12.1	5.3
	No. of Pistils	56	48	39	51	56
No. 43	19, June	82.1	72.9	59.0	27.5	17.9
	24, July	67.9	62.5	38.5	13.7	0
	No. of pistils	58	44	54	62	69
No. 101	19, June	89.7	52.3	33.3	30.6	24.6
•	24, July	46.6	22.7	18.5	7.9	7.2
Total	No. of pistils	135	154	132	179	182
Mean	19, June	83.7	55.2	49.2	26.3	17.6
(3 trees)	24, July	58.5	42.2	33, 3	11.7	4.4

 Table 1.
 Set percentage in artificial pollination

According to Table 1, it is clear that the set-percentage is highest in A sect., and next in B sect., in which it is largest in B_1 sect., next in B_2 sect. and lowest in B_3 sect. In C sect. the nut should not be set, nevertheless some percentages are recorded here. The fact may be due to the parthenogenesis, regarding which WOOD

(1934) ZARUBIN (1949) and SCHANDERL (1964) reported in Persian walnut. As in the above, the percentages in B sect. are smaller than those in A sect., because the dropping of the pistils or young nuts was often occurred by overheating of the paper bags, and also, as KAVECKAJA and TOKAR (1963) described, by trouble of the stigma tissue brought by excessive pollens.

At all events it is indicated that the pollination at the early stage of flowering results the highest percentage of nut setting, whereas the stigma at the last stage of flowering keeps a little receptivity.

These results are agreed with the studies of WOOD (1934).

2. Morphological Variation of Stigma

The flowering time and the dichogamy in Shinano walnut are excluded in the present pater, because they have been discussed in the previous papers (MACHIDA 1952, 1955).

The external form of the pistillate flower in Shinano walnut is nearly the same as that of Persian walnut.

According to the study of MANNING (1940), the pistillate flower was composed of one bract, two bracteoles, four sepals and pistil.

The author has observed a large number of the pistillate flowers of Shinano walnut. As seen in Plate Π -1, the bract and the bracteoles as well as the sepals are fused with the ovary at their basal part. The tips of both bracteole and sepal are separated with each other at the top of ovary, although the former is so small as indistinguished and the latter is easily observed. The hair is grown densely on the surface of the involucre. Each of two styles exserted from the ovary forms the stigma respectively (Plate Π -1).

In the flowering time, the diameter of the ovary is $3\sim4$ mm and the length of the stigma is $4\sim5$ mm.

The shape of the stigma in Shinano walnut differs considerably from that of Japanese walnut (*J. sieboldiana* MAX.) and of American black walnut (*J. nigra* L.), but has a likeness to that of Persian walnut. The stigma is a plumous lobe with milky white color and bears many glandular pleats. However, some individuals have the stigmas taking a light pinkish color at the top portion.

The author considers that the character is originated by a genetic factor. Therefore, it must be used as an indicator of classification of Shinano walnut, because GLENN (1951) described such a fact on some varieties of Persian walnut. Also the stigma color varies according to species, viz. red in Japanese walnut, milky white with pale green in American black walnut and milky white in Persian walnut.

The forms of stigmas at the best stage of florescence were observed in detail for many years. As the results the author discovered that the shape of the stigma in plane view varied fairly according to individual tree, and that could be classified into three types or more.

These types are shown in the bellow (Plate II 2, 3, 4).

- 1. Semi-opened fan type
- 2. Spindle type

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- 3. Long oval type
- 4. Other types

Then, the author researched the stigma type in seedling trees in our orchard. The results obtained are shown Table 2.

Table 2. Frequency of individual tree based on stigma type

					-
Type	Semi-opened fan	Spindle	Long oval	Others	
No. of tree	26	22	2	15	

Most of "Others" in the above table take a rectangular shape, which resembles closely the semi-opened fan type. Therefore, it can be said that Shinano walnut belongs to the semi-opened fan type on morphology of stigma.

The shape of the stigma in Japanese walnut is more slightly elongate than that in the spindlle type of Shinano walnut, and that in American black walnut belongs to the spindle type though having a curled part at under side (Plate II-5, 6).

The type of the stigma seems to be an indicator of the varietal classification on Shinano walnut.

3. Character of Poly-flowered Cluster

It is well known that the pistillate flowers of Japanese walnut are grown generally at the long cluster which bears more or less 10 flowers (Plate Π -7), and that those of the Shinano walnut have normally two-flowered cluster, as seen in Persian walnut.

The author carried out the research on the habit of flowering for many years, and found out an interesting individual, which bears a lot of poly-flowered clusters, ten years ago. It is presumed to be originated by the inherent character of the individual.

Then the author studied the number of the poly-flowered clusters and further productivity of nuts in this tree.

This problematical individual was compared with a common tree in regard to the number of the poly-flowered clusters. The results are shown in Table 3.

According to Table 3, A-tree bears a small number of the two-flowered cluster, on the contrary, has a lot of the poly-flowered clusters as compared with the other trees. The poly-flowered clusters are poor in B, C and D trees, and scarcely found

No. of flower per cluster Year Indi-No. of vidual cluster 2 1 3 4 5 6 7 8 9 10 2,5 13.0 41.3 28.1 8.1 3.11,3 0.6 160А 38.7 53.2 3.6 0.9 В 111 3.6 С 526,0 62.0 22.0 6.0 2.0 2.0 7742.9 1964 15.6 38.9 2.6 D 2.3 57.5 87 36.8 3.4 Ε F 65.0 1238,1 26.9 G 10414.4 61.5 23.1 1.0 A 100 16.0 35.0 34.0 5.0 4.0 2.0 2.0 1.0 1.0 82 21.9 54.9 23.2 В 1965 С 31 29.0 61.3 6.5 8.2 50 32.0 60.0 8.0 D 59 13.6 66.120.3 Е 718.5 54.9 35.2 1.2 F

Table 3. Percentage of poly-flowered cluster

Remarks : A.....problematical individual.

B,C,D,.....Individuals which bear some resemblance to A-tree E,F,G,.....Common individuals.

in E, F and G trees.

Conclusively it is clear that A-tree has the special character which bears many poly-flowered cluster.

On the other side, the nuts of A-tree are provided with nice characteristics for practical use.

The author named it "Birei" which means "Beautiful" in Japanese.

Recently *Birei* grafted on a common tree was investigated to know the number of the poly-flowered cluster (Plate $\Pi - 8, 9$). The results are shown in Table 4.

Year	No. of cluster	and in a few many second of the second s			cluste	r grade	*		
i cai	cluster	1	2	3	4	5	6	7	8 9
1970	101	27.7	30.7	23.8	9.9	5.0	1.0	1.0	1.0
1971	116	16.4	40.5	33.6	8.6		0.9		1997 - 19
1972	152	11.2	26.3	32.2	15.1	11.9	3.3		· -

Table 4. Percentage of poly-flowered cluster in Birei

* Cluster grade was estimated by numbers of flower per cluster

The percentages of the poly-flowered cluster in *Birei*, varying according to year, are smaller than those of the mother tree (A-tree in Table 3), though larger

than those of common trees.

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The author conformed here again that such a character as bearing much polyflowered cluster in *Birei* is genetically decided.

Next, the author examinated the growth of the shoot bearing the poly-flowered clusters. The results are shown in Table 5.

year	Indiv-	No. of			Average length					
idua idua	idual	shoots 1		2	3	4	5	6	7-10	of total shoot
	A	160	4.8	6.6	7.4	9.6	10.7	10.1	6.9	7.9
1964	В	111	3.0	7.5	10.9	10.6	10.0			8.4
1001	С	50	9.7	6.5	7.6	11.2		14.0	25.4	12.4
	D	77	4.5	8.7	15.5	10.8				9.9
	А	100		5.9	7.1	8.1	10.8	8.5	5.8	7.0
1965	В	82		10.0	14.4	13.2				12.5
2000	С	31		8.0	9.1	11.5	9.0			9.4
	D	50		8.4	10.5	12,5				10.5

Table 5. Relation between shoot length and cluster grade

As seen in Table 5, the lengths of shoot having the poly-flowered cluster are longer in general than those bearing the 2-flowered cluster, although the shoot lengths are different in each individual, respectively. Therefore, it is assumed that the shoots with the poly-flowered cluster are generally superior in growth.

During the developmental process of the flower into fruit the physiological nut drop tends to occur. The author ressearched the number of dropping nuts in each poly-flowered cluster of the variety, *Birei*. The results are indicated in Table 6.

Year Tree age	Tree		****	Average					
	1	2	3	4	5	6	7~10	Average	
1964	*33	0	52.1	49.5	13.9	52.3	73,3	63.4	50.9
1965	*34		53.1	54.3	47.1	40.0	58.3	74.4	61.1
1970	10	11.5	17.4	19.4	12.5	8.0	16.6		15.8
1971	11	0	3,8	5.4	8.7	7.8	10.0		10.0

 Table 6.
 Percentages of physiological dropping nuts in poly-flowered cluster of *Birei*

* Same tree (mother tree of Birei)

As seen in Table 6, the percentages of the physiological nut drop in the mother tree are so much higher than those in the young tree, which was grafted. It seems

that the fact depends on the declining of vitarity of the mother tree.

Even so, the percentages in more than 6-flowered cluster are evidently higher than those in 2-flowered cluster, except in the case of young tree.

However, such a difference $(10\sim20\%)$ in the percentage seems not to result the reduction of productivity, because the number of bearing nuts in the 6~10-flowered cluster must be greater than that in the 2-flowered one if the percentage is the same in the both clusters. The fact is understood in Table 7.

	No. of		Avarage							
	Cluster	1	2	3	4	5	6	7	8	Avalage
*A	160	1.0	1.0	1.5	1.8	2.2	1.6	2,5	3.0	1.6
Birei	152	1.0	1.9	2.8	3.6	4.6	5.4	-		2.8

Table 7. Nunber of mature nuts per cluster

*A : mother tree of Birei

Further, the author examined some practical characteristics of the dry nuts. The results are shown in Table 8.

Year	Cluster	No. of	:	Nut	Kernel			
grade		nuts	Weight	Volume	Weight	Percentage		
	1	20	9.5	22,5	5.1	54.1		
	2	20	9.5	22,6	5.1	53.8		
	3	20	9.2	22,5	5.0	54.2		
1970	4	20	9.2	22,5	5.0	54.2		
	5	20	8.8	17.2	4.5	50.7		
	6	20	8.7	18.9	4.8	55.5		
7	7	7	10.0	20,6	5.1	50.9		
	1	20	8.3	22,4	5.1	61.8		
	2	20	8.4	22,8	5.0	59.8		
	3	20	8.6	22.0	5.1	59.2		
1971	4	20	8.5	21,6	5.0	58,5		
	5	20	8.6	22,0	5.2	60.0		
	6	20	7.9	20,9	4.6	58.4		

 Table 8. Characteristics of dry nuts classified by cluster grade in *Birei*

According to Table. 8, each value of the nut-weight, the nut-volume and the kernel-weight in 1970 and 1971 slightly decreases with the elevation of the cluster

grade, and becomes evidently small at the cluster grade $5\sim 6$, although such a decrement is not seen in the kernel-percentage.

In fruit-tree such as apple-tree which fruits in poly-flowered cluster, size and quality of the fruit cannot generally be in normal state, provided the cluster keeps a natural condition.

In Shinano walnut, however, the nuts matured in the clusters bearing more than 5 flowers are not so small as it is reputed.

Conclusively the nuts matured in the poly-flowered cluster contribute to the yield in the number of the nuts rather than in the size, so that the characteristic of bearing poly-flowered cluster is a factor enhancing the productivity.

4. Abnormal Inflorescence

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Shinano walnut tree is monoecious. It bears an anemophilous unisexual flower as seen in all other species on the genus *Juglans*. The pistillate flowers are grown on the tip of young shoot whereas the staminate flowers on the biennial twigs.

We can rarely find an abnormal phenomenon in a flower-organ or in an inflorescence in general fruit-trees.

The author observed such abnormal phenomenons in Shinano walnut as described in the following during the past twenty years.

- (1) June, 1956. Secondary growth of catkin bud.
- (2) May, 1959. Simultaneous occurrence of germination and florescence of pistillate flower.
- (3) May, 1962. Both pistillate and staminate flowers grown in the same inflorescence.
- (4) June, 1964. Secondary growth of catkin bud.
- (5) May, 1971. Both pistillate and staminate flowers grown in the same inflorescence.

The abnormal phenomena above-mentioned are xpressed in detail as in the following.

(1) Secondeary growth of catkin bud

The new catkin bud begins to grow after about one month from natural flowering time, and comes into the staminate catkin. However, several abnormal points are detected in this secondarily growing catkin compared with the primary catkin; the axis is thicker and grows upwards, the staminate flower is slightly smaller and easily separated from the axis and a projection like primitive stigma is found in some flowers which lie at the middle part of the axis (Plate III-1, 2).

(2) Simultaneous occurrence of germination and florescence of pistillate flower

In normal, pistillate flower opens on the tip of the young shoot which elongates after germination. In this case, however, Pistillate flower opens at the same time

with germination, while no growth of the shoot and the infant leaf is occurred. The reason remains unsolved (Plate III-3).

(3) Both pistillate and staminate flowers grown in the same inflorescence

It occurred in *Birei* above-mentioned (Chap. 3) during the natural flowering time. The inflorescence is compose of the pistillate cluster and the staminate catkin which grows on the former. There are 10 and 11 flowers in the cluster and the catkin, respectively. The dichogamy is of a plotogynous type. The following abnormal points are described in each flower (Plate $\Pi -4, 5$).

Pistillate flower : The tip of the ovary constricted, the bracteole indistinct, the sepal short and wide (Plate III-6).

Staminate flower : The bracecole not developed, the form small and triangular in plane view, the anther a few and not shed pollen (Plate III-7).

Also a projection, like a primitive stigma, was found at the center of the anther group in some staminate flowers, which lie at the proximal part of the catkin. These flowers are recognized as hermaphrodite according to the opinion of SCEPOTJEV (1955), and NEDEV and BAEV (1972) (Plate III-8).

The author could not find the fruiting of pistillate flower on the inflorescence above-mentioned. However. PETROSYAN and ANTONENKO (1972) found the pistillate, the staminate and hermaphrodite flowers in the same inflorescence which grown secondarily, and certained the fruiting of their flower. TUZ (1958) observed also the fruiting in the secondary flower of persian walnut.

The causal factor, however, remains to be solved.

SUMMARY

The author studied on the fructification of so-called Shinano walnut, which had been cultivated mainly in Japan. It is said as the hybrid between the ancient variety (*J. regia* L. var. *orientis* KITA.) and some variety of Persian walnut (*J. regia* L.).

In the present paper, molphology and ecology of the pistillate flower and the cluster of Shinano walnut were dealt with.

The outline of the results is described as follow.

1. The process of the flowering of the pistillate flower was devided into five stages. It was recognized that the receptivity of the stigma was the best condition at the eraly stage in the flowering process, and decreased remarkably at the last stage by the artificial pollination.

2. The author discovered that the shape of the stigma varied at the best stage of the flowering process, It was classified into three types. The type seems to be used as an indicator of classification of the variety. 3. The author found that some individual trees kept the ability bearing the poly-flowered cluster.

The ability was recognized as a factor concerning the productivity.

4. Some abnormality was observed in the inflorescence, the opening of the pistillate flower and in growth of the catkin and the staminate flower.

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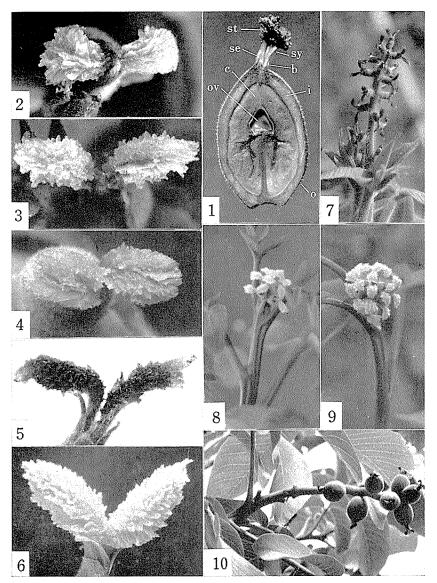
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E F В С G Η D

- A. Budding flower stage
- C. Biginning of florescence
- E. Middle stage of flowering
- G. Last stage of flowering
- B. Budding flower stage
- D. Early stage of flowering
- F. Best stage of flowering
- H. End of florescence



Plate II



- Longitudinal section of young nut after four weeks from end of florescence 1. b : Bracteole, c : Carpel, i : Involucle, o : Ovary, ov : Ovule, se : Sepal, st : Stigma, sy : Style,
 - Semi-opened fan type of stigma in Shinano walnut
- 2. 3. Spindle type of stigma in Shinano walnut
- 4. Long oval type of stigma in Shinano walnut
- 5. Stigma in Japanese walnut 6. Stigma in American black walnut
- 7. Poly-flowered cluster in Japanese walnut
- 8. Poly-flowered cluster in Shinano walnut (5 flowers)
- 9. Ditto (10 flowers) 10. Nut-setting on poly-flowred cluster

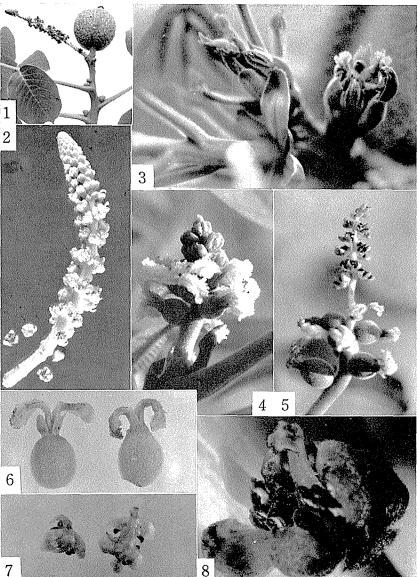


Plate III

1. Secondary growth of new catkin bud

- 2. Secondarily grown catkin
- 3. Simultaneous occurrence of germination and florescence of pistillate flower
- 4. Abnormal inflorescence (protogynous) on 11, May
- 5. Ditto, on 18, May
- 6. Pistillate flower of abnormal inflorescence (left) and of normal one (right)
- 7. Staminate flower of abnormal inflorescence (left) and of normal one (right)
- 8. Hermaphroditic flower