GENETICAL STUDIES IN PEARS

III. INCOMPATIBILITY AND STERILITY

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(With Plates 1 and 2 and Three Text-figures)

CONTENTS

1. INTRODUCTION

THE studies on incompatibility and sterility in plums, cherries and apples, which have been in progress at the John Innes Horticultural Institution since 1911, have recently been extended to pears. In the pear the basic chromosome number is 17, and cultivated varieties comprise diploids, triploids, and tetraploids. Our results are summarized in Tables 1–5 and Text-figs. 1–3. Tables 1 and 2 give the set of fruit following self-pollination, Text-figs. 1–3 the set of fruit from cross-pollination, and Tables 3–5 the number of viable seeds formed, and other details.

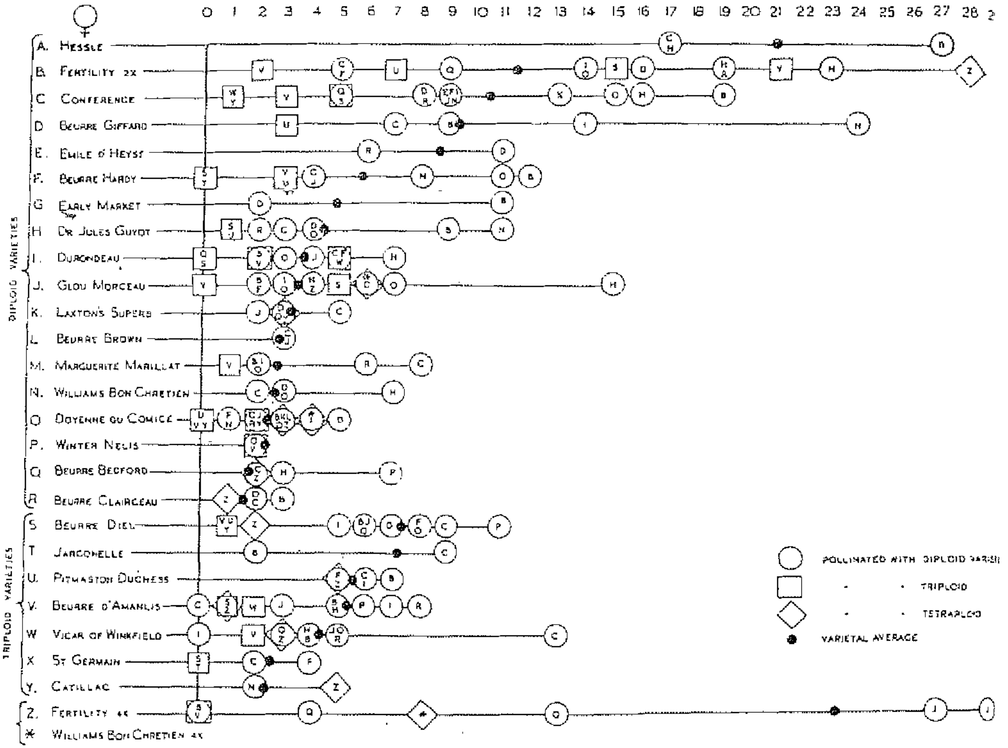
The results set out in Table 1 and Text-figs. 1–3 were all from trees flowered in pots under glass. Many of the pollinations were repeated in different years, results being summed in the table. The figures in Table 2 were from pollinations made in 1941 on trees growing in the open.

2. INCOMPATIBILITY IN DIPLOIDS

It is clear from Tables 1 and 2 that the failure or poor set of fruit following self-pollination is mainly due to incompatibility, since many of the varieties cropped heavily when suitably cross-pollinated.

In Table 1 the total results obtained from the self-pollinations made are detailed. In most varieties a number of separate pollinations have been made and the results have varied in different years. Thus twenty-six separate self-pollinations have been made on the variety Conference, and the percentage of fruit set and matured has ranged from 0 to 3·4 %; and in Fertility $2x$ twelve separate pollinations were carried out and the results ranged from 0 to 3·2 % matured.

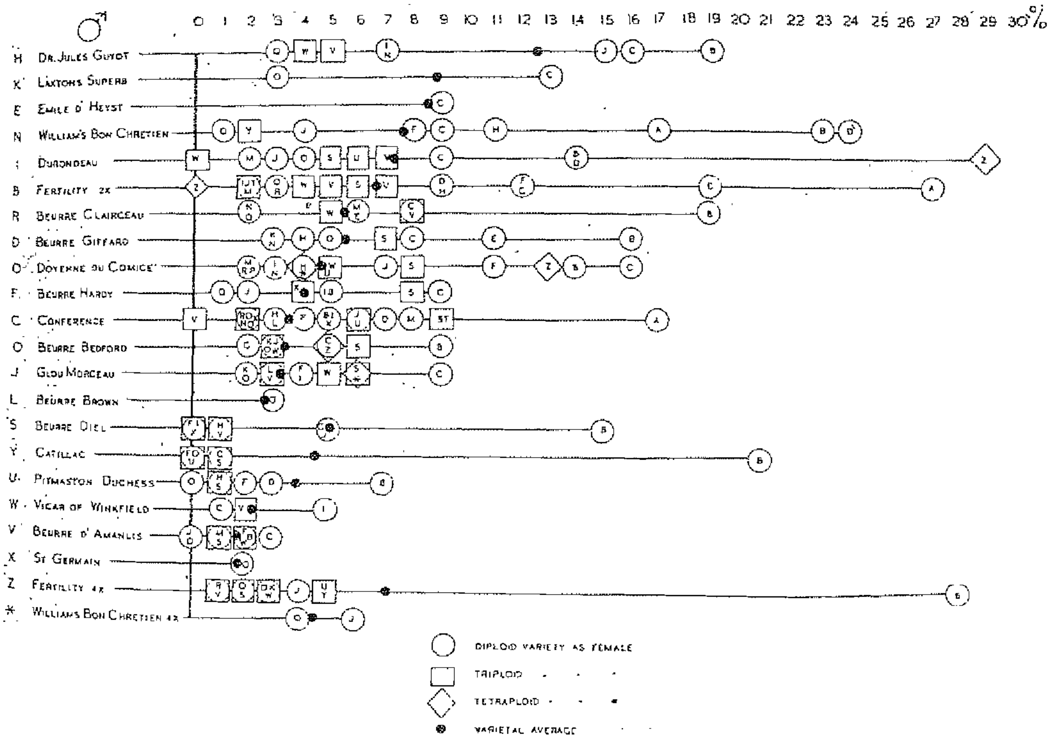
The results obtained from cross-pollinations are set out in Text-figs. 1–3. In Text-fig. 1 the female fruit fertility of the varieties is shown, and in Text-fig. 2 the fertility of the same varieties when used as males. In Text-fig. 3 the mean fruit fertility of the diploid and triploid varieties is set out, male fertility against female. These figures show that in some



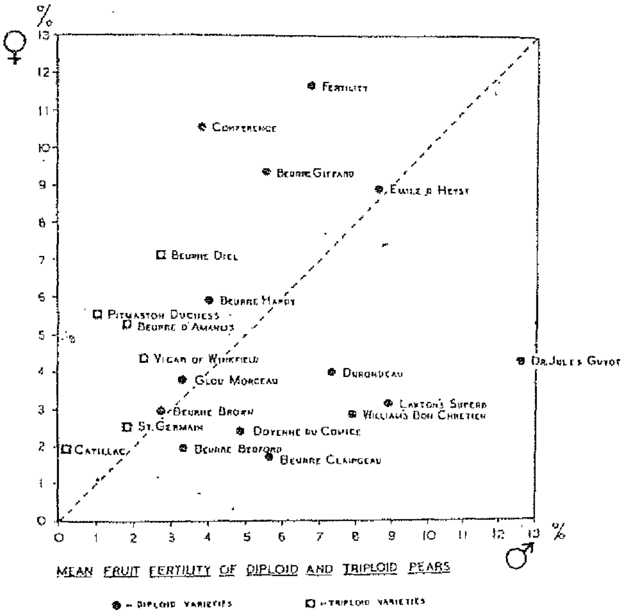
Text-fig. 1. Female fruit fertility; see text.

varieties the fertility on the two sides differs considerably. Thus the mean female fertility of Conference is over 10 %, its male fertility less than 4 %. In contrast, the average female fertility of Dr Jules Guyot is less than 5 %, whilst its average male fertility is over 12 %.

As shown in Text-fig. 3, the triploids are more fertile when used as females than when used as males. In this figure the results obtained from using Fertility $2x$ as a female with triploids have been omitted. As shown in Text-fig. 1, this variety gives exceptionally high results when pollinated



Text-fig. 2. Male fertility; see text.



Text-fig. 3.

with triploids. In an earlier paper (Crane & Thomas, 1939) it was suggested that these unusual results were due either to apomixis or to gametic selection. Since then Dr Thomas, from further cytological studies, has concluded that the gametic output of the triploids indicates gametic selection rather than apomictic development on the part of the variety Fertility as responsible for the high fertility of this diploid variety when pollinated with triploids. Furthermore, in families raised from Fertility $2x$ with triploids, an unusually high proportion of the seedlings are diploids, but the variation which occurs within the families is considerable and not in accordance with apomictic reproduction. Fertility $2x$ has the capacity for eliminating aneuploid pollen grains which are not eliminated by other diploid varieties. How this system works requires further investigation.

Table 1

Variety	Flowers self-pollinated	Fruits matured	% matured
Beurré Bedford	672	2	0·3
Beurré Clairgeau	1676	0	0·0
Beurré Giffard	666	0	0·0
Beurré Hardy	2164	5	0·2
Brown Beurré	123	0	0·0
Clapp's Favourite	212	0	0·0
Conference	5313	41	0·7
Dr Jules Guyot	2030	1	0·05
Doyenné du Comice	4364	16	0·4
Durondeau	1661	14	0·8
Early Market	378	3	0·8
Emile d'Heyst	520	0	0·0
Fertility $2x$	1994	16	0·8
Glou Morceau	2555	6	0·2
Hessle	175	0	0·0
Laxton's Superb	1064	0	0·0
Marguerite Marillat*	1362	0	0·0
Williams Bon Chrétien	1056	1	0·1
Winter Nelis	307	0	0·0
Beurré Alex Lucas	84	1	1·2
Beurré d'Amanlis	907	7	0·8
Beurré Diel	1749	5	0·3
Catillac	452	0	0·0
Jargonelle	171	1	0·6
Pitmaston Duchess	1420	0	0·0
St Germain	260	0	0·0
Vicar of Winkfield	1822	0	0·0
Fertility $4x$	144	21	14·5

In this table diploid varieties are shown in ordinary type, triploids in italics, and tetraploids in heavy type.

* Marguerite Marillat has no good pollen.

Fruits of diploid and triploid pears obtained from self-pollination usually have few good seeds, and often nothing but shrivelled and empty testas. For example, in the twenty fruits obtained from the variety Fertility in Table 2 there was no embryonic development. Conference and Durondeau are other varieties which commonly produce parthenocarpic (seedless) fruits.

Table 2

Pollinations	Flowers pollinated 25. iv. 40	Fruits set 17. v. 40	Fruits set 29. v. 40	Fruits matured	% matured
Conference selfed	188	73	36	0	0
Fertility $2x$ selfed	175	42	33	20	11·4
Beurré Diel selfed	357	0	0	0	0

Table 3

	No. of crosses	Flowers pollinated	Fruits matured	% fruits matured	Seeds	Seeds per fruit	% seeds germinated	% survived
$2x$ selfed	13	29,846	99	0·3	157	1·5	65·6	59·3
$3x$ selfed	8	6,471	13	0·2	2	0·1	42·8	42·8
$4x$ selfed	1	144	21	14·5	86	4·1	58·8	58·8
*$2x \times 2x$	164	18,963	1065	5·6	7000	6·6	86·7	86·3
$2x \times 3x$	37	5,030	182	3·6	589	3·2	75·4	73·1
†$3x \times 2x$	40	6,076	343	5·6	474	1·3	52·4	46·6
$3x \times 3x$	8	2,167	18	0·8	10	0·5	40·0	40·0
$2x \times 4x$	7	1,019	80	7·8	476	5·9	88·1	87·7
‡$4x \times 2x$	3	63	12	19·0	70	5·8	77·1	75·7

* Excluding Marguerite Marillat × Beurré Bedford.
† Excluding Beurré d'Amaulis × Conference.
‡ Excluding **Fertility** $4x$ × Fertility $2x$.

Table 4

Parents	Fowers pollinated	Fruit matured	% fruit matured	Seeds	Seeds per fruit
Beurré Bedford used as male					
Conference × Beurré Bedford	540	25	4·6	8	0·3
Doyenné du Comice × Beurré Bedford	487	16	3·3	10	0·6
Laxton's Superb × Beurré Bedford	106	3	2·8	4	1·3
Fertility $2x$ × Beurré Bedford	28	1	3·6	0	0·0
Beurré Bedford used as female					
Beurré Bedford selfed	439	1	0·2	0	0
Beurré Bedford × Dr Jules Guyot	148	4	2·7	16	4·0
Beurré Bedford × Conference	90	2	2·2	6	3·0
Beurré Bedford × Winter Nelis	30	2	6·7	11	5·5
Beurré Bedford × **Fertility** $4x$	83	2	2·4	5	2·5

The expression of incompatibility is often obscured by the modifying influence of sterility and parthenocarpy. It is evident that the results shown in Tables 1 and 2 represent productivity rather than fertility; as shown later, the best measure of fertility and compatibility in pears is the number of viable seeds per fruit.

So far, only two cases of complete cross-incompatibility have been established. In each case one partner is a polyploid and the incompatibility is in one direction only, namely, when the polyploid is used as female, as follows:

	Flowers	Fruits
Fertility $4x$ × Fertility $2x$	45	0
Beurré d'Amanlis $3x$ × Conference $2x$	523	0
Fertility $2x$ × Fertility $4x$	190	54
Conference $2x$ × Beurré d'Amanlis $3x$	845	25

In the reciprocal pollinations Fertility $2x$ gives a heavy crop, but Conference gives a comparatively light crop when pollinated with Beurré d'Amanlis.

As shown in Text-figs. 1–3, the proportion of flowers which develop to give fruits after compatible *pollination* has varied considerably. The age of the tree, the crop carried in the previous year and varietal differences in fruit size are often involved in these variations. If two trees bear the same number of fruits, but one is more free-flowering than the other, the shyer-flowering variety will of course show a higher percentage set. Reinecke (1930), for example, states that flower-bearing capacity is a varietal characteristic and that a 3% set with a free-flowering variety will produce a big crop, while a variety that flowers sparingly will require a set of 15–20% for a satisfactory crop. In our experiments a 4 or 5% set, as illustrated in Pl. 1, fig. 1, is on the average a good crop.

Some of the cross-pollinations in Text-figs. 1 and 2, where a small group of pollinations has given a poor set, should not be too seriously regarded as a repetition may give a better result.

3. INCOMPATIBILITY IN POLYPLOIDS

As already shown (Crane & Lawrence, 1929), in different fruits the expression of incompatibility is of many kinds. Thus in the diploid cherry, *Prunus avium*, the inheritance and phenotypical behaviour of incompatibility is sharply discontinuous and is a comparatively simple phenomenon. All varieties are completely self-incompatible, and they fall into intra-sterile inter-fertile groups on the oppositional factor scheme of East & Mangelsdorf (1925). In the hexaploid plum, *P. domestica*, incompatibility is more complex; some varieties are self-incompatible, others completely or partly self-compatible. Cross-incompatibility and reciprocal differences involving both partial and complete self-incompatibility also occur, but cross-incompatibility is the exception rather than the rule as in cherries. In the apple, which is a secondary polyploid (Darlington & Moffett, 1930), incompatibility is even more complex than in the plum. Complete self- or cross-incompatibility is rare, and among the numerous varieties of apples tested, incompatibility grades almost imperceptibly. It is therefore evident that there is a connexion between incompatibility and chromosome make-up.

When we come to pears we find that incompatibility is more sharply defined than in the apples, but less so than in the sweet cherries. From Table 1 it will be seen that about half the varieties tested are completely or almost completely self-incompatible, and the remainder only self-compatible to a low degree. This difference between apples and pears is another expression of a difference in chromosome behaviour. The pear shows less secondary association of chromosomes at meiosis than the apple (Moffett, 1933).

Morphological characters in apples and pears show an analogous difference. In apples such leaf characters as colour, hairiness and serration intergrade instead of segregating into clearly defined categories (Crane & Lawrence, 1931), but the same characters in pears show discontinuous and sharply defined differences (Crane & Lewis, 1940).

There is also a difference between apples and pears in the fertility of the triploids. When triploids are crossed, the low yield is mainly due to sterility, from unbalanced gametes and embryos: not to incompatibility. Now triploid varieties of apples, when intercrossed or crossed by diploids, are more fertile than triploid pears whether judged by fruit set or seeds per fruit (see Table 5). Numerically unbalanced gametes are more viable in a secondary polyploid, owing to the replication of the chromosomes. Thus all the evidence, from incompatibility, sterility, cytological behaviour and genetic variation, shows that while both are secondary polyploids in origin, the pear is functionally more like a diploid than the apple. If the apple and pear had a common ancestor, as their taxonomy and cytology suggest, it is remarkable that such divergences should have occurred in the evolution of these two species. Evidently differentiation, by some means we do not yet understand, has gone further in the pear than in the apple.

There is, however, another possibility. It is probable that in more recent times, through hybridization, a greater number of species have taken part in the origin of the cultivated apple than of the cultivated pear. Heterogeneity would therefore be greater in apples than in pears and would account for the differences detailed above.

4. PARTHENOCARPY

Fruits which reach maturity yet have no seeds are commonly termed parthenocarpic, and in what follows we shall use the term in this sense. Little is known about parthenocarpic development; it may embrace three types of abnormal fruit formation: (1) fertilization followed by the death of the embryo at a very early stage, (2) pollination, where the growth of the pollen tubes, though not leading to fertilization, stimulates fruit development, (3) fruit development without pollination.

None of the varieties of pears investigated can be described as unconditionally parthenocarpic, as none are able, under all conditions, to develop a crop of entirely seedless fruits. But, as we have shown, the tendency to form parthenocarpic fruit is strongly developed in certain varieties. Many varieties, however, never form fruits without a proportion of seeds (see Table 5). The varieties which crop most freely are those in which parthenocarpy is most pronounced. Thus Conference and Fertility have in some cases given over 20% of matured fruits. At the other extreme, Doyenne du Comice, with one unique exception, has not given entirely seedless fruits, and in cross-pollinations this variety has only yielded about 5% of fruit. Lying in between are such varieties as Beurré Hardy and Glou Morceau, which are intermediate both in productivity and in tendency towards parthenocarpic development.

Table 5

	Triploid selfed				Triploid × diploid			
	Flowers pollinated	Fruit matured	% fruit matured	Seeds per fruit	Flowers pollinated	Fruit matured	% fruit matured	Seeds per fruit
Pears	6471	13	0·2	0·1	6076	343	5·6	1·3
Apples	5806	145	2·5	1·9	3095	112	3·6	3·0
	Triploid × triploid				Triploid × tetraploid			
Pears	2167	18	0·8	0·5	758	22	2·9	2·6
Apples	930	23	2·5	2·1	—	—	—	—
	Diploid × triploid							
Pears	5030	183	3·6	3·2				
Pears*	3877	66	1·7	1·8				
Apples	4373	270	6·1	2·4				

* Excluding Fertility $2x$ as ♀.

The influence of cultural conditions should not, however, be overlooked. As shown in Table 2, under outdoor conditions the variety Fertility $2x$ developed a full crop of seedless fruits, and although the selfed fruits of Conference (also without embryonic development) failed to reach maturity, they grew to a considerable size before falling from the tree. It appears therefore that conditions out of doors may be more favourable to parthenocarpic development than under glass.

The condition of the fruits, whether fertile or seedless, is of concern to the grower, as fruits with a full complement of seeds are of a normal symmetrical shape, while in many varieties fruits with few or no seeds tend to be misshapen or distorted.

The above conclusions apply particularly to diploid varieties and pollinations between them. In triploids, sterility adds a further complication, and crosses between diploids and triploids are discussed in the next section.

5. STERILITY IN POLYPLOIDS

Sterility resulting from chromosome unbalance, as distinct from incompatibility due to inhibition of pollen-tube growth, is particularly evident in crosses involving triploids. Table 3 gives the results of self-pollinating diploid, triploid and tetraploid varieties. The tetraploid is highly self-compatible, producing 14 % of fruit with 4·1 seeds per fruit. The diploid varieties selfed mature 0·3 % of fruits, with an average seed content of 1·5; the triploids about the same amount of fruit, but with only 0·1 seed per fruit. Assuming then that the diploids, which will set a full crop when suitably crossed, are only affected by incompatibility, the further reduction of seed fertility in the triploids must be due to chromosomal sterility. But this comparison does not show us the full effect of sterility; for incompatibility, on the general theory, will be less strong in the triploids: incompatibility depending on the chance of similar genes meeting, which is lower in triploids than in diploids.

In the tetraploid neither cause of unfruitfulness operates; the chromosome behaviour is orderly and the self-sterility mechanism has broken down. The cause of this breakdown in the self-sterility mechanism which follows doubling of the chromosomes is described in detail in the accompanying paper on pollen-tube growth in polyploids (Lewis & Modlibowska, 1941).

The results from reciprocal diploid-triploid crosses (Table 3) show a significant contrast between the two measures of fertility, fruit set and seed set, as follows:

	Fruit set	Seed per fruit	Germination %
$2x \times 3x$	3·6	3·2	75·4
$3x \times 2x$	5·6	1·3	52·4

In the diploid × triploid pollinations, despite the lower set of fruit, the average seed yield was more than twice that of the reciprocal and the germination of the seed was also better. A similar result is reported in apples by Brittain & Eidt (1933). The low seed content of the $3x \times 2x$ fruits is accounted for by the lack of opportunity for selection between female gametes; on the male side greater selection is possible. The relatively high fruit set with poor seed formation is due to the higher proportion of good pollen and successful pollen tubes in diploids. The diploid pollen succeeds in fertilizing the triploid egg; but chromosome unbalance in the egg often makes the resulting zygote inviable: thus many pears are formed but few seeds.

It is often presumed that seed formation is essential for fruit development. With pears, however, our results show that the penetration of the style and embryo-sac by the pollen tubes can, at least in some cases, supply the necessary stimulus for fruit development. As shown above, varieties differ in their capacity to set fruit without seed, and it may be that these differences account for the triploids producing no smaller crops than diploids despite their lower seed content. Perhaps in selecting for fruit yield in triploids breeders have been compelled to select for parthenocarpy.

Triploid pollen, however, is less effective than diploid in stimulating fruit setting of either diploids or triploids. It seems therefore that parthenocarpy depends on pollen germination as well as on the parthenocarpic abilities of the female parent. This is not surprising, since parthenocarpy in our sense includes, in addition to true parthenocarpy, fruit formation with arrested seed development.

A special example of the importance of pollen tube growth for fruit setting is seen in the variety Beurré Bedford. This variety is a diploid and has the characteristic appearance of a diploid, but its behaviour is very exceptional. After the second division of meiosis the cell wall fails to form, and the four nuclei are enclosed within one cell. In subsequent development these nuclei may fuse to a varying extent, giving $2x$, $3x$ and $4x$ nuclei, of which only the last, resulting from complete fusion, are comparable with normal pollen grains. The others form branched pollen tubes, presumably owing to their multinucleate constitution (Thomas, 1941).

As shown in Table 4, the results from crossing Beurré Bedford with other diploids are unique. With Bedford as male, the set of fruit is comparable with that from an ordinary diploid cross-pollination, but the fruits formed have very few or no seeds, the average being 0·5 seed per fruit and the occasional seeds are usually inviable. Beurré Bedford pollen therefore has the quality of stimulating fruit formation without seed formation. For fruit development in most diploid crosses complete pollen tube growth, followed by fertilization and seed formation, is essential for fruit production. Doyenné du Comice, for example, as female produced no seedless fruit in any cross but that with Bedford. These results recall the work of Gustafsson (1939) and Dollfus (1936) who applied pollen extracts to the styles of many plants and obtained seedless fruits. Kostoff (1930) has also reported that in certain intergeneric crosses in the Solanaceae, seedless fruits can be stimulated to form by the penetration of the pollen tubes into the micropyle.

When Beurré Bedford is used as the female parent, the fruit set is slightly lower than in normal diploids, and the seed content is lower than normal, though much higher than when Bedford is used as a male parent.

6. SUMMARY

1. Tests of self-compatibility have been made in nineteen diploid, eight triploid and two tetraploid varieties of pears. Nine of the diploid and four of the triploid varieties completely failed to set fruit with their own pollen. With two exceptions, one diploid and one tetraploid, the others are only self-compatible to a low degree.

2. The diploid form of the variety Fertility when selfed can set a full crop, but the fruits are seedless. The tetraploid form of Fertility sets a full crop of fruit and seed with its own pollen.

3. In 193 intervarietal cross-pollinations made, only two cases of complete cross-incompatibility have been established. In each case one partner is a polyploid, and the incompatibility only acts when this polyploid is the female parent.

4. Parthenocarpy, following self-pollination, is frequent in pears. Parthenocarpic fruits are often inferior in size and shape to fruits with seeds.

5. The expression of incompatibility is more discontinuous in pears than in apples.

6. Some varieties are more fertile on the male than on the female side, others on the contrary more fertile on the female than on the male side.

7. Sterility in triploid varieties is expressed by a low proportion both of good pollen and good seeds. Poor crops are obtained, both on diploids and triploids, when triploids are used as pollinators.

8. Beurré Bedford, a diploid, gives aberrant results in pollinations

with other varieties. This is associated with abnormal pollen development after a regular meiosis.

We are indebted to Mr Gavin Brown for assistance in these experiments.

REFERENCES

Brittain, W. H. & Eidt, C. C. (1933). Seed content, seedling production and fruitfulness in apples. *Canad. J. Res.* **9**, 307–33.

Crane, M. B. & Lawrence, W. J. C. (1929). Genetical and cytological aspects of incompatibility and sterility in cultivated fruits. *J. Pomol.* **7**, 276–301.

—— (1931). Sterility and incompatibility in diploid and polyploid fruits. *J. Genet.* **24**, 97–107.

Crane, M. B. & Lewis, D. (1940). Genetical studies in pears. II. A classification of cultivated varieties. *J. Pomol.* **18**, 52–60.

Crane, M. B. & Thomas, P. T. (1939). Genetical studies in pears. I. The origin and behaviour of a new giant form. *J. Genet.* **37**, 287–99.

Darlington, C. D. & Moffett, A. A. (1930). Primary and secondary chromosome balance in *Pyrus*. *J. Genet.* **28**, 327–8.

Dollfus, H. (1936). Wuchsstoffstudien. *Planta,* **25**, 1–21.

East, E. M. & Mangelsdorf, A. J. (1925). A new interpretation of the hereditary behaviour in self-sterile plants. *Proc. nat. Acad. Sci., Wash.*, **11**, 166–71.

Gustafsson, F. G. (1939). Auxin distribution and its significance in fruit development. *Amer. J. Bot.* **26**, 189–94.

Kostoff, D. (1930). Ontogeny, genetics and cytology of *Nicotiana* hybrids. *Genetica,* **12**, 33–139.

Lewis, D. & Modlibowska, I. (1941). In the Press.

Moffett, A. A. (1933). Cytological studies in cultivated pears. *Genetica,* **15**, 511–18.

Reinecke, O. S. H. (1930). Pollination requirements of varieties of deciduous fruit trees.

Thomas, P. T. (1941). In the Press.

EXPLANATION OF PLATES 1 AND 2

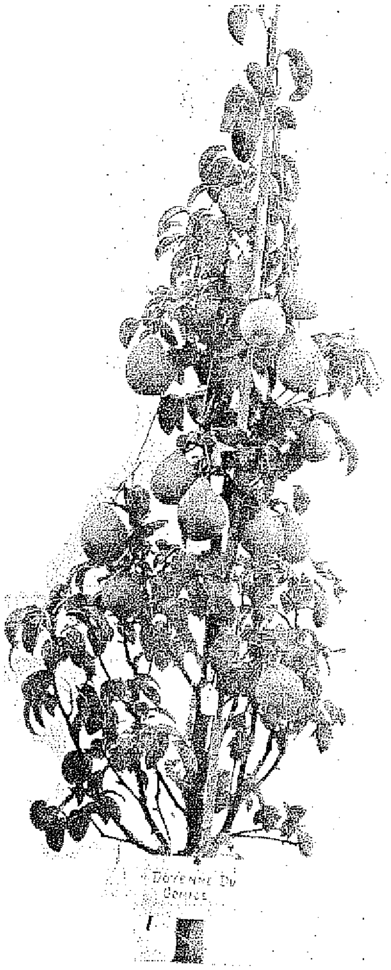
PLATE 1

Fig. 1. Doyenné du Comice

	Flowers	Fruits
Top: × Glou Morceau	188	6
Centre: × Beurré Giffard	76	6
Bottom: Selfed	74	0

Fig. 2. Beurré Hardy

	Flowers	Fruits
Top: Selfed	542	5
Bottom: × Fertility 2x	117	18
,, × Glou Morceau	170	18
,, × Doyenné du Comice	166	22



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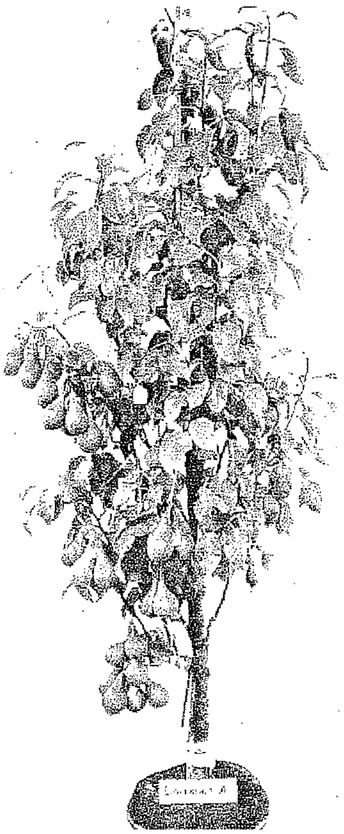

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PLATE 2

Fig. 1. Beurré d'Amanlis

	Flowers	Fruits
Top: × Beurré Clairgeau	282	23
,, × Dr Jules Guyot	227	11
,, × Fertility 2*x*	149	10
Left: × Conference	236	0
Bottom right: Selfed	504	1

Fig. 2. Conference

	Flowers	Fruits
Top: × Beurré d'Amanlis	61	1
Left: × Dr Jules Guyot	101	32
Right: Selfed	57	0