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Helen Aufderheide
Butler University

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CHROMOSOME NUMBERS IN FAGUS GRANDIFOLIA AND QUERCUS VIRGINIANA

By HELLEN AUFDERHEIDE

INTRODUCTION

The chromosomes have been counted in a number of species of Fagaceæ, both in Europe and America. The opportunity now arises to add to the rapidly growing list one more species of Quercus and one species of Fagus whose numbers have not until now been determined.

Within the order Fagales, to which the genera Quercus and Fagus belong, Woodworth (13), 1929, determined 14 to be the reduced number in 17 species, varieties and hybrids of Corylus. The genus Betula has been found to contain polyploid forms with 14 as the basic haploid number (Wetzel (12), Jaretsky (7), Woodworth (13), (16), Helms and Jorgensen (5)), while Alnus has 14 as the basic number of a 2x and a 4x series (Woodworth (14), Jaretsky (7), Wetzel (12)). The Fagaceæ have been regarded as very regular indeed. In 1929 Wetzel (12) determined the chromosome numbers in *Fagus silvatica*, *Castanea sativa* and the following species of Quercus: *Q. cerris*, *Q. coccinea*, *Q. Dalechampii*, *Q. glandulifera*, *Q. Libani*, *Q. macranthera*, *Q. nigra*, *Q. Pontica*, *Q. Robur* and *Q. sessiliflora*. In each species he found the reduced number to be 11. Later, Jaretsky (7) took the same slides from which Wetzel (12) had made his counts. For every species studied Jaretsky (7) found 12 instead of 11 as the reduced number. Ghimpu (2) in 1929 reported 12 for each of the following species: *Q. cerris*, *Q. coccifera*, *Q. Ilex*, *Q. palustris* and *Q. suber*. Hoeegg (6) reported 12 chromosomes for *Q. sessiliflora*, *Q. robur* and *Q. suber* Sno-Eg.

In January, 1930, Friesner (1) published the results obtained from a study of the following ten species of Quercus: *Q. alba*, *Q. borealis*, *Q. coccinea*, *Q. macrocarpa*, *Q. marylandica*, *Q. Michauxii*, *Q. Muhlenbergii*, *Q. prinoides*, *Q. Prinus* and *Q. velutina*. His work was done on the root tips, the number determined being 12. In 1931 Tischler (11), in his "Pflanzliche Chromosomen-Zahlen," gave the reduced number for all the Fagaceæ, so far reported, as 12, stating in a footnote that Friesner (1) had made his counts correctly but that he had misinterpreted what he had seen, having mistaken the metaphase with its 24 chromosomes for the anaphase.

In the genus *Carpinus* the reduced number has been shown to be 8 in seven species and 32 in one variety (Wetzel (12), Jaretzky (7), Woodworth (16)). Three species and one variety of *Ostrya* (Wetzel (12), Jaretzky (7), Woodworth (16)) and one species of *Ostryopsis* (Woodworth (16)) each show 8 as the reduced number.

MATERIAL AND METHODS

Nuts of *Fagus grandifolia* were gathered October 18, 1930, at Trevelac, Indiana, and were planted in moist sawdust in the greenhouse October 20, 1930. On March 7, 1931, two nuts had germinated with roots long enough to be cut, while three nuts had roots too short to be of use. These were replanted. By March 14 four nuts had germinated with roots sufficiently long to be cut.

Acorns of *Quercus virginiana* were obtained December 30, 1930, at Allandale, Florida. They were planted January 3, 1931, in moist sawdust and set in the greenhouse to germinate. January 22, 1931, the seedlings had roots long enough to be cut. When the radicles from any particular seedling were long enough, they were removed and washed free of sawdust particles. The root tips were killed in Navashin's Killing Fluid, which consists of:

Part A—1 per cent chromic acid, 10 parts; glacial acetic acid, 1 part.

Part B—40 per cent formalin: 40 cc commercial formaldehyde; 100 cc distilled water.

Parts A and B were mixed in the proportion of 11:4.

Root tips were washed free from the killing solution, dehydrated and brought into paraffin. They were sectioned 10 microns thick and stained in iron hæmatoxylin. All chromosome counts were made with a Spencer research microscope equipped with apochromatic objectives and aplanatic condenser and at a magnification of 1900 x. Counts were made from both side and polar views of the metaphase and early anaphase stages of somatic mitosis.

OBSERVATIONS

Fagus grandifolia Ehrh. ($2x=12$). Figures 1 and 2 show polar views of the anaphase. Chromosome behavior is normal with 24 daughter chromosomes. Figures 3 and 4 show side view of the early anaphase.

Figure 4 shows an early anaphase with one lagging chromosome still in the equatorial plane. Figures 5 and 6 show polar views of the metaphase with chromosomes larger and fatter than those in the anaphase in Figures 1-4.

Q. virginiana Mill. ($2x=12$). Figures 7 and 8 show polar views of the anaphase with 24 chromosomes. Figures 9 and 10 show the early anaphase, side view, with 12 daughter chromosomes moving toward each pole. Figure 11 shows a polar view of the metaphase with 12 large, fat chromosomes in the equatorial plane. These are twice the size of those found in Figure 9. Figure 12 shows a side view of the metaphase with 12 large, fat chromosomes.

DISCUSSION

The low haploid number of 6 determined for 10 species of *Quercus* studied by Friesner (1) and one by the present writer, seems to be very significant, in view of the fact that Jaretzky (7), Ghimpu (2), Hoeegg (6) and Wetzel (12) found double that number in a number of species including two (*Q. alba* and *Q. coccinea*) studied by Friesner. If it were true, as Tischer (11) says, that Friesner (1) misinterpreted what he saw, mistaking the metaphase with its 24 chromosomes for the anaphase, cells should be found showing polar views of the anaphase with 48 daughter chromosomes. With this possibility in mind, the present writer has searched diligently for any such cells in her material. No cells were found showing more than 24 chromosomes in polar views. On the other hand, many cells were found showing polar views of the metaphase in which 12 larger and fatter chromosomes were scattered in the equatorial plane. There can be no doubt about these numbers, since repeated drawings of the same cells yielded the same numbers. Furthermore, these cells were entire cells, since the chromosomes could be focused entirely out of view without losing sight of the cytoplasm of the particular cell under study. If 12 were the reduced number, the metaphase should show 24 in such a view taken from root cells. Since 12 chromosomes were seen so clearly and distinctly in a large number of cells, the conclusion must be reached that the diploid number for the root cells of the species under study is 12 and not 24. If 12 were the reduced number, that number should not occur in any root cells at all. As further evidence that 12 is the diploid and not the haploid number for *Quercus virginiana*, side views of cells in both metaphase (Figure 12) and anaphase (Figures 9 and 10) stages are presented. Figures 9

and 10 show very clearly 12 chromosomes (and not 24) going to each pole of the dividing cell.

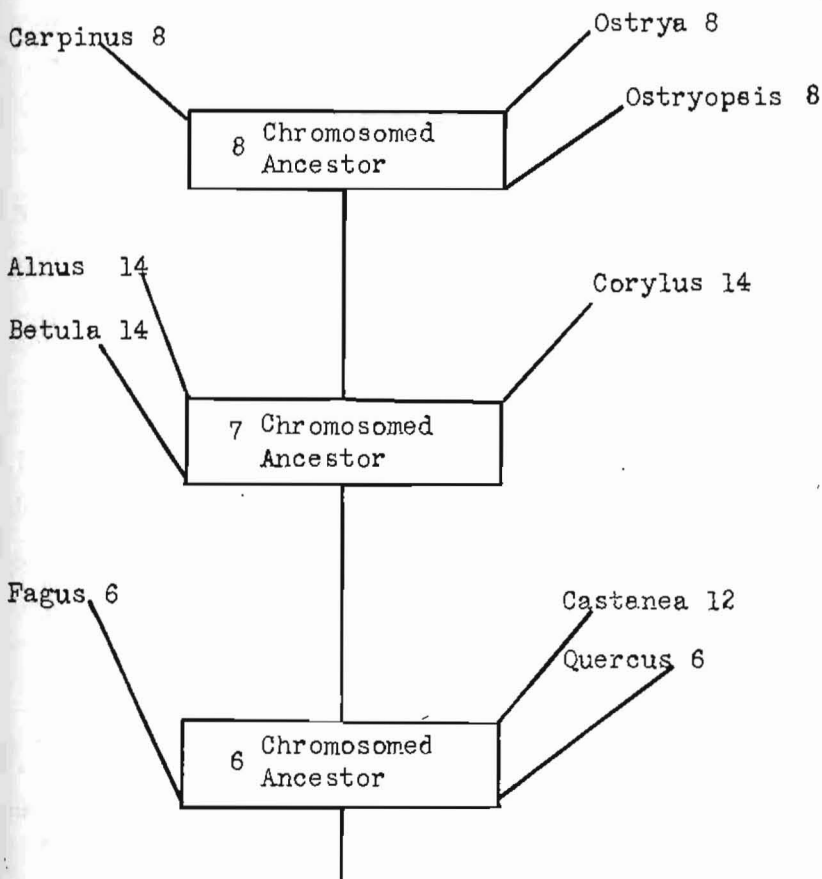
It is possible that polyploidy is present in *Quercus* as well as in *Fagus*, *Alnus* and *Betula*, and that the American species of *Quercus* have just half as many chromosomes as the European species. These are from different geographical sections of the world. Those made by Friesner (1) are from American material and those made by Kostoff and Kendall (8) and by Jaretsky (7) are from European material. It is entirely possible that American and European races of the same species have different chromosome numbers and that Tischler (11) is incorrect in his contention that Friesner's (1) interpretations are wrong, since Friesner's figures show polar views of metaphase plates with 12 chromosomes moving to each pole of the dividing cell. A careful re-examination of the preparations from which Friesner's (1) counts were made is now under way.

The same argument presented to support the view that 12 is the diploid and not the haploid number in *Quercus* is likewise applied to *Fagus grandifolia*, since the count given by Jaretsky (7) for *Fagus sylvatica* is twice the number found in the present investigation. The many clearly defined cells leave little doubt as to the number to be found or as to the interpretation.

If it is true, as it has been maintained by Osawa (10), Longley (9), Heilborn (7, 8) and others, that the lower the number of chromosomes possessed by members within any closely related group the more primitive they are in respect to their phylogeny, then the genera *Quercus* and *Fagus* are more primitive than any other of the Fagales, since the basic chromosome numbers within the order are 6 for *Quercus* and *Fagus*, 8 for *Carpinus* and *Ostrya*, 12 for *Castanea* and 14 for *Corylus*, *Alnus* and *Betula*. Both *Alnus* and *Betula* contain polyploid forms, the former with tetraploid, pentaploid and hexaploid forms, and the latter with triploid, tetraploid, pentaploid, hexaploid and aneuploid forms. The basic number 14 for *Alnus*, *Betula* and *Corylus* may possibly represent a tetraploid form of a more primitive ancestor with 7 as the haploid number. *Castanea* with its haploid number 12 may likewise represent a tetraploid form of a more primitive ancestor whose haploid number was 6. This then would make a possible group of ancestral forms with 6, 7 and 8 as haploid numbers possible progenitors of the present members of the Fagales. The American members of the order are:

Fagales	Family Betulaceæ	Tribe I	Ostryopsis	8
			Corylus	14
			Ostrya	8
			Carpinus	8
		Tribe II	Betula	14
			Alnus	14
	Family Fagaceæ	Tribe I	Fagus	6
		Tribe II	Castanea	12
			Quercus	6

The following diagram will help to show a possible phylogeny of the probable progenitors of the modern Fagales.



SUMMARY

1. Chromosome studies reveal 12 as the diploid number of chromosomes in root cells of *Q. virginiana* and *F. grandifolia*.
2. Mitotic behavior is regular and normal in both species.
3. A diagram of the possible progenitors of modern Fagales is presented.

Grateful acknowledgment is made for helpful criticism given by Dr. R. C. Friesner during the process of this study.

EXPLANATION OF FIGURES

- Figures 1, 2. *Fagus grandifolia*: polar view showing 24 chromosomes in early anaphase.
- Figure 3. *Fagus grandifolia*: side view of early anaphase showing 12 daughter chromosomes moving toward each pole.
- Figure 4. Same, with one chromosome lagging in the equatorial plane.
- Figures 5, 6. *Fagus grandifolia*: polar view of metaphase showing 12 undivided chromosomes.
- Figures 7, 8. *Quercus virginiana*: polar view of early anaphase showing 24 daughter chromosomes.
- Figures 9, 10. *Quercus virginiana*: side view of early anaphase showing 12 daughter chromosomes moving toward each pole.
- Figure 11. *Quercus virginiana*: polar view of metaphase showing 12 undivided chromosomes.
- Figure 12. *Quercus virginiana*: side view of metaphase showing 12 undivided chromosomes in the equatorial plane.



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