STUDIES IN ANNONACEAE. VIII. A CLADISTIC ANALYSIS OF *TETRAMERANTHUS*

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Summary

The small genus *Tetrameranthus* (five species) stands isolated within the Annonaceae. A cladistic analysis was carried out using macromorphological characters in order to find possible apomorphies and to attempt a phylogenetic reconstruction. In the "best" cladograms there appear two subsets, one formed by *T. duckei, T. macrocarpus,* and *T. pachycarpus,* the other by *T. laomae* and *T. umbellatus.* Both are supported by a number of apomorphic character states. Any other conclusions remain speculative.

Introduction

Recently, a taxonomic revision of the genus *Tetrameranthus* was published by Westra and collaborators (Westra, 1985). That paper essentially deals with presentation of data (including two newly described species), it does not make any statements on possible relationships other than accepting the genus as a member of the Annonaceae. This is because *Tetrameranthus* has a unique combination of characters and stands rather isolated within the family. Principal among these characters are: 1) leaves arranged in a spiral, 2) the perianth in whorls of four, and 3) flowers subtended by a verticil of four bracts; for more data the reader is referred to Westra (1985).

Various positions for *Tetrameranthus* have been proposed. In Fries's (1959) comprehensive survey of the Annonaceae it is placed as the sole member of a tribe Tetramerantheae in subfamily Annonoideae. Walker (1971), in a classification based on his palynological studies, places the genus in the *Uvaria* tribe of the *Malmea* subfamily. It should be remarked in this context that there is a considerable discrepancy between palynological data by Hesse and Waha (1984) and those by Walker. Most recently, karyological investigations by Morawetz (1986b) show that *Tetrameranthus* could have a closer connection with the African genus *Uvariopsis* and a looser connection with the neotropical *Guatteria* tribe, rather than forming a monotypical tribe or being part of a heterogeneous *Uvaria* tribe sensu Walker.

All attempts to find allies for *Tetrameranthus,* so far, led to nothing or to contradictions at the most. Therefore a logical step now was to subject *Tetrameranthus* to a cladistic analysis in order to find out apomorphic character states, and to attempt to give a phylogenetic reconstruction. Only macromorphological data could be used, since a complete set of data of other kind is not available for each of the five species at this moment. Cladistic analysis is performed by means of a method that integrates the concepts of compatibility and parsimony (Zandee, 1985). A description and application can be found in Roos (1986) and Zandee and Geesink (1987).

The Data-Matrix

Within *Tetrameranthus* several aspects of variation may be noticed:

—arborescent vs. shrubby habit; four out of the five species now known are trees; *T. duckei* is a shrub to a small tree (note: in the revision (Westra, 1985) *T. macrocarpus* is erroneously described as "Shrub to tall tree");

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Table 1. List of code numbers used in the data matrix and cladograms indicating character states in *Tetrameranthus* and *Asimina tetramera*. Taxa: 1 = *Asimina tetramera*, 2 = *Tetrameranthus duckei*, 3 = *T. laomae*, 4 = *T. macrocarpus*, 5 = *T. pachycarpus*, 6 = *T. umbellatus*.

	1	2	3	4	5	6
1. Trees, height ≥ 10 m	0	0	1	1	1	1
2. Shrubs, or small trees, height <10 m	1	1	0	0	0	0
3. Branchlets stout, diam. shortly below apex >2 mm	0	1	0	1	1	1
4. Branchlets slender, diam. shortly below apex <2 mm	1	0	1	0	0	0
5. Base of petiole slightly thickened	0	1	0	1	1	0
6. Petioles of uniform diameter throughout	1	0	1	0	0	1
7. Primary vein prominent to flat adaxially	1	0	1	0	0	1
8. Primary vein impressed adaxially	0	1	0	1	1	0
9. Stellate hairs (length of rays) <0.2 mm long	0	0	1	0	0	1
10. Stellate hairs (length of rays) >0.2 mm long	0	1	0	1	1	0
11. Stellate hairs absent	1	0	0	0	0	0
12. Inflorescence (up to) 5-flowered	0	0	0	0	0	1
13. Inflorescence 1(–2)-flowered	1	1	1	1	1	0
14. Peduncle >5 mm long	0	1	0	1	1	1
15. Peduncle <5 mm long	1	0	1	0	0	0
16. Callus of outer petals over ⅙ of the total petal length	1	1	0	1	0	0
17. Callus of outer petals under ⅙ of the total petal length	0	0	1	0	1	1
18. Apex of connective conical	0	1	0	1	1	0
19. Apex of connective flat	1	0	1	0	0	1
20. Diameter of (dried) fruit becoming >3.5 cm	1	0	0	1	1	0
21. Diameter of (dried) fruit remaining <2.5 cm	0	1	1	0	0	1
22. Two-seeded fruit with oblique lateral constriction	1	1	1	1	0	1
23. Two-seeded fruit not or inconspicuously constricted	0	0	0	0	1	0
24. Leaves in spiral arrangement	0	1	1	1	1	1
25. Leaves distichous	1	0	0	0	0	0
26. Perianth in whorls of 4	1	1	1	1	1	1
27. Perianth in whorls of 3	0	0	0	0	0	0
28. Bracts verticillate	0	1	1	1	1	1
29. Bracts otherwise	1	0	0	0	0	0

—indument of comparatively large stellate hairs (trichomes = rays mostly ≥0.5 mm long) in *T. duckei, T. macrocarpus, T. pachycarpus* vs. minute stellate hairs (trichomes mostly ≤0.1 mm long) in *T. laomae* and *T. umbellatus*;

—rather large fruits with a diameter (in sicco) exceeding 3.5 cm in *T. macrocarpus* and *T. pachycarpus* vs. distinctly smaller fruits not exceeding 2.5 cm in diameter (in sicco);

—a prominent to flat primary vein on the adaxial side in *T. laomae* and *T. umbellatus* vs. an impressed primary vein on the adaxial side in *T. duckei, T. macrocarpus, T. pachycarpus*;

—flowers single or flowers (up to) five in umbels in leaf axils. The latter condition is only found in *T. umbellatus*, although *T. duckei* may exceptionally develop two flowers together;

—peduncle well-developed, the bracts well beyond the base of the inflorescence vs. peduncle reduced, the bracts at the base of the inflorescence, tiny and partly missing. The latter condition is found in *T. laomae* only;

—anthers with a protruding, conical apical prolongation of the connective in *T. duckei, T. macrocarpus, T. pachycarpus* vs. with a flat, cushion-shaped top in *T. laomae* and *T. umbellatus*.

Together with some less conspicuous characters which need not to be mentioned at all in detail here, these characters form the base of a binary data-matrix (Table 1).

Choice of the Outgroup

For outgroup selection, the following features were especially taken into consideration:
—leaves in a spiral. Leaves in Annonaceae are placed in two alternating rows (1/2) in essentially all representatives but *Tetrameranthus*. Among the rare exceptions (fide Fries, 1959) are a few species of *Artabotrys* and possibly *Annona crotonifolia,* where leaves on some shoots have been found in three rows (1/3). A further exception is *Disepalum anomalum* where, similarly, a phyllotaxis of 2/5 has been observed;
—tetramerous flowers. Such flowers may exceptionally be found (usually among the regular flowers) on individuals in probably all species of Annonaceae that normally have trimerous flowers. Deviations of this kind are nothing unusual, and occur in other families of flowering plants as well. Truly tetramerous flowers are met with, apart from *Tetrameranthus,* in *Asimina tetramera* and in *Reedrollinsia* (only in *R. cauliflora*);
—stellate trichomes. Within the family, predominantly simple (to occasionally furcate) hairs are found. Stellate (and/or scaly) hairs are encountered, however, in a variety of not necessarily related genera and species in Annonaceae.

Out of those, the number of perianth members per whorl seems to offer the best handhold. There are no species (known) in Annonaceae outside *Tetrameranthus* that have leaves in an arrangement other than 1/2 over the whole plant (in the cases mentioned only some of the shoots on a plant are involved). The indument, on the other hand, seems to leave too wide a variety of choice, and so does not lead us anywhere.

For practical reasons, *Reedrollinsia* had to be dropped, since it is too incompletely known at the moment. This leaves *Asimina tetramera* as the only candidate out of all applicants for the job, even though populations of this species certainly do not have exclusively tetramerous flowers (Kral, 1960).

Asimina tetramera is a shrub to about 3 m tall with axillary, single flowers with a perianth consisting of one whorl of four or three sepals and two whorls of four or three petals, and with (rather) large, fleshy monocarps with several seeds of 1–2 cm long. It occurs in eastern peninsular Florida (Kral, 1960). Note that axillary inflorescence position and comparatively large monocarps with large seeds (1–2) are (also) characteristic of *Tetrameranthus*!

Recently it was found that karyologically the African genus *Uvariopsis* shares a great deal with *Tetrameranthus* (Morawetz, 1986a, 1986b; see also introduction to this paper). Morphologically there seems to be little similarity between the two, however, at least no more than between *Tetrameranthus* and the bulk of annonaceous genera. The tetramerous corolla in a single whorl (!) in *Uvariopsis* might be regarded as made up of two dimerous whorls rather than a single truly tetramerous whorl; the number of sepals, after all, is two!

Character Analysis and Discussion

The outgroup choice just discussed cannot be but a provisional one, as complete data with regard to other candidates are still lacking. Therefore it was decided to have separate analyses: first, with the five species assigned to *Tetrameranthus* only, and, secondly, with the addition of *Asimina tetramera* as the outgroup.

An analysis of *Tetrameranthus* using *partially monothetic sets,* as cladogenetic units (Zandee and Geesink, 1987) produced 12 fully resolved cladograms (Figs. 1.1–1.12).

Ten of these cladograms fit the data equally well, the other two (Figs. 1.1 and 1.2) only fall behind because of one more ambiguity remaining in the character state pairs 16–17 and/or 20–21, due to lack of an outgroup.

When *Asimina tetramera* is introduced as an outgroup, only six completely resolved cladograms result from an analysis using partially monothetic sets as building blocks for cladograms. One of these stands out as the best regarding the balance between fitting (15) and contradictory (7) character states. It corresponds with Fig. 1.3, but it has *A. tetramera* now in the position of global outgroup (Fig. 2.1). In fact, it is the only cladogram out of these six that features *A. tetramera* in this position. All other grouping opportunities for
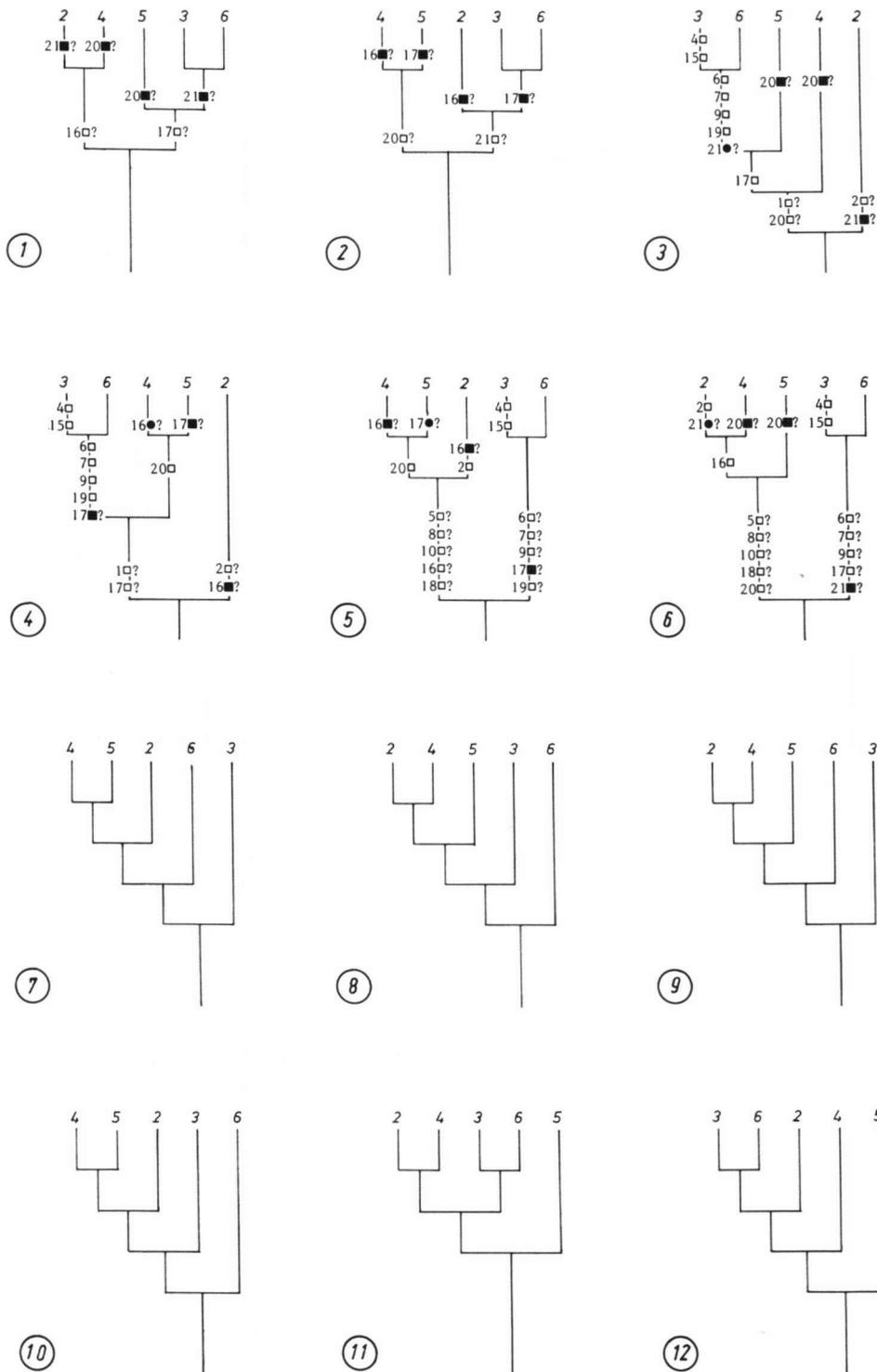
Fig. 1.   Cladograms indicating possible phylogenetic relations within *Tetrameranthus* without *Asimina tetramera* as sistergroup. Only those apomorphic character states mentioned in the text are given. For further explanation, see text. (See Fig. 2 for key to symbols used.)

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(1)

3 6 5 4 2 1

4●
15●

6●
7●
9□
19●
21■   21■

17□

1□

3□?   4■?
5□?   6■?
8□?   7■?
10□?   11□?
14□?   15■?
18□?   19■?

(2)

3 6 4 5 2 1

4●
15●   16△

6●
7●
9□
19●   20●

1□
17□

3□?   4■?
5□?   6■?
8□?   7■?
10□?   11□?
14□?   15■?
18□?   19■?
21□?   20■?

T. macrocarpus *T. pachycarpus* *T. duckei* *T. lacmae* *T. umbellatus* *A. tetramera*

(3)

4 5 2 3 6 1

17■   4●   12□
23□   2● 15●

20●?   21■?

5□
8□   17■
10□?   21■?
18□   9□?

1□?   2■?
3□?   4■?
14□?   11□?
21□?   15■?
24□?   20■?
28□?   23□?
       27□?

(26)

T. duckei *T. macrocarpus* *T. pachycarpus* *T. lacmae* *T. umbellatus* *A. tetramera*

(4)

2 4 5 3 6 1

2●
21■   4●   12□
          15●

16●?   17■?
       23□

5□
8□   17■?
10□?   21■
18□   9□?

1□?   2■?
3□?   4■?
14□?   11□?
17□?   15■?
24□?   16■?
28□?   23□?
       27□?

(26)

□ : Apomorphy
■ : Apomorphy showing homoplasy, but undisputed because of the outgroup rule
△ : Reversal
● : Apomorphy showing homoplasy or reversal
? : Indicates uncertainty about the polarity of the complementary state of the
    character, either because of the lack of an outgroup, or because the alter-
    native transformation series are equally parsimonious

Fig. 2. Cladograms indicating possible phylogenetic relations within *Tetrameranthus* with *Asimina tetramera* as sistergroup. In Figs. 2.3 and 2.4 all possible apomorphic character states are recorded; in the other figures only those mentioned in the text are given. For further explanation, see text.

A. tetramera given the character states in the data-matrix renders its outgroup position impossible.

When *strictly monothetic sets* (Zandee and Geesink, 1987) are accepted in a cladogram, the analysis based on *Tetrameranthus* alone results in 70 cladograms. Ten out of these 70 represent the best possible fit to the data regarding the balance between supporting (15) and contradictory (1) character states. These ten cladograms are the same as found on the basis of partially monothetic sets (Figs. 1.3–1.12).

In six out of these ten cladograms there appears a subset formed by *T. duckei* (2), *T. macrocarpus* (4), and *T. pachycarpus* (5), though in different three-cladon permutations (Figs. 1.5–1.10).

When *Asimina tetramera* is accepted as outgroup, an analysis using strictly monothetic sets as building blocks resulted in no less than 190 possible cladograms, in 70 of which *Asimina* really occupies the outgroup position. Out of the 190, the three "best" cladograms also represent *Asimina* as an outgroup to *Tetrameranthus* (Figs. 2.2–2.4). As can be seen from Figs. 1 and 2, the internal structure of these three cladograms is similar to some of those found for *Tetrameranthus* alone (Figs. 2.2 ≡ 1.4; Figs. 2.3 ≡ 1.5; Figs. 2.4 ≡ 1.6). Two of these (Figs. 2.3 and 2.4) show the same amount of fitting character states (15), but have one homoplasy less (6) and are as a consequence one step shorter (19), than the cladogram in Fig. 2.1. The third one (Fig. 2.2) shows the same balance (9) between fitting (16) and contradictory (7) character states, but at the cost of two extra steps (21).

Cladograms 2.3 and 2.4 support the apomorphic nature of character states 5, 8, and 18. The cladograms do not give an answer about the apomorphic state for the hair types (9 vs. 10). On the base of cladograms 2.3 and 2.4, there are several possibilities: 11 → 10 → 9; 11 → 9 → 10; 9 ← 11 → 10. In contradiction to the best cladogram in Fig. 1, character states 3 and 14 are now indicated as possibly apomorphic. Within Annonaceae the inflorescence is considered to have evolved from positions exhibiting one terminal flower to such structures as (a.o.) leaf-opposed or axillary groupings of flowers through processes of reduction of leaves and foreshortening of internodes (Fries, 1919, 1959). Within this framework, the extremely short peduncle (peduncular part) in *T. laomae* (3) would seem a derived condition. In cladograms shown in Fig. 2, however, it can be interpreted as a reversal of the state shown by the global outgroup *A. tetramera*. Following the same reasoning, character state 14 as indicated by global outgroup comparison (peduncle >5 mm long in *T. duckei, T. macrocarpus, T. pachycarpus,* and *T. umbellatus*) can also be interpreted as the apomorphic condition, with character state 15 as the reversal to a state also shown much earlier in the phylogeny of the Annonaceae.

Placing all putative evidence together, the cladograms in Figs. 2.3 and 2.4 are chosen as the most likely representations of cladogenesis in *Tetrameranthus.* As indicated by this choice, we postpone a final decision with regard to the relationships among *T. duckei, T. macrocarpus,* and *T. pachycarpus.* The following character states are suggested as apomorphies:

1. trees (vs. shrubs) in *T. laomae, T. macrocarpus, T. pachycarpus,* and *T. umbellatus* (with reversal in *T. duckei*);

3. diameter of branchlets stout (vs. slender) in *T. duckei, T. macrocarpus, T. pachycarpus,* and *T. umbellatus* (with reversal in *T. laomae*);

5. base of petiole slightly thickened (vs. of uniform diameter throughout) in *T. duckei, T. macrocarpus,* and *T. pachycarpus*;

8. primary vein impressed adaxially (vs. prominent to flat adaxially) in *T. duckei, T. macrocarpus,* and *T. pachycarpus*;

9 or 10. stellate hairs [length of rays] <0.2 mm long (vs. stellate hairs [length of rays] >0.2 mm long) in *T. laomae* and *T. umbellatus,* or stellate hairs [length of rays] >0.2 mm long (vs. stellate hairs [length of rays] <0.2 mm long) in *T. duckei, T. macrocarpus,* and *T. pachycarpus*;

12. inflorescence (up to) 5-flowered (vs. inflorescence 1(–2)-flowered) in *T. umbellatus*;

14. peduncle >5 mm long (vs. peduncle <5 mm long) in *T. duckei*, *T. macrocarpus*, *T. pachycarpus*, and *T. umbellatus* (with reversal in *T. laomae*);

18. apex of connective conical (vs. apex of connective flat) in *T. duckei*, *T. macrocarpus*, and *T. pachycarpus*;

23. two-seeded fruit not or inconspicuously constricted (vs. two-seeded fruit with oblique lateral constriction) in *T. pachycarpus*.

As regards the character state pair 16–17, global outgroup comparison indicates 17 to represent the apomorphic state for *Terameranthus*. As a consequence, and as registered by cladogram optimization, character state 16 is interpreted as a reversal for *T. macrocarpus* and *T. duckei* in cladogram 2.4 (one transformation series 16 → 17 → 16), while in cladogram 2.3 two independent acquisitions of character state 17 occur (two transformation series 16 → 17).

As to the character state pair 20–21, the reverse is true. The occurrence of character state 20 in *A. tetramera* indicates this to represent the plesiomorphic state. Consequently, and following cladogram optimization, a reversal to state 20 is shown in cladogram 2.3 (one transformation series 20 → 21 → 20), while cladogram 2.4 shows two independent origins of state 21; once in *T. duckei*, and once in *T. laomae* and *T. umbellatus* together (two transformation series 20 → 21).

It should be recalled that in both cases quantitative characters are involved, i.e., relative length of callus and diameter of (dried) fruit. For both, a reversal can easily be assumed. The choice of *A. tetramera* does not lead to one best cladogram.

The subset formed by *T. duckei*, *T. macrocarpus*, and *T. pachycarpus* is supported by a number of apomorphies. Within this subset, however, it is difficult to settle on a choice between the two alternatives, although the mutual resemblance between *T. macrocarpus* and *T. pachycarpus* is such that they were taken for one single species in the early course of the taxonomic work. This overall-similarity is represented by cladogram 2.3.

Turning to *T. duckei*, a derived nature of that species appears likely from recent field work by Morawetz (1986b). Morawetz has found that *T. duckei* is a tetraploid, and *T. umbellatus* a diploid ($2n = 28$ and $2n = 14$, respectively). *Tetrameranthus duckei* is a shrub to small tree found in savanna shrub vegetation, low forest, secondary vegetation, etc. Other species, as far as known, are (much) larger forest trees. This fits in quite well with Morawetz's conclusion from investigations in various families (including Annonaceae) that the step diploid-polyploid may have correlation with invasion from humid to more extreme, often xeric habitats (Morawetz, 1986a, 1986b). As long as chromosome numbers of the other species of *Tetrameranthus* and of *Asimina* are unknown, and more accurate data on habitat are lacking, it is not possible to include these characters in cladistic analysis. However, for the time being we think that data about them as far as known now, point towards the choice of cladogram 2.3 as the "best" one.

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Note Added in Proof

After this paper had gone to press, a new species of *Tetrameranthus* was discovered (to be published in a forthcoming issue of *Proc. Kon. Ned. Akad. Wetensch. Natuurk. Ser. 2*). The new species is unique in having globular fruits. All other features are also found in one or more other species of *Tetrameranthus.* A new cladistic analysis supports the conclusions of this paper. It was no longer possible to form a cladogram on the base of partially monothetic sets. Taking Figs. 2.3 and 2.4 as a starting point, nr. 7 and 2+4+5 are sister groups with 3+6 as outgroup. For the character pairs 16–17 and 20–21, now in both cladograms, only parallel developments are indicated.