

INTRASPECIFIC VARIATION OF PHYLLOTAXIS STABILITY IN *ANAGALLIS ARVENSIS*

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ABSTRACT

Qualitative transformations of phyllotaxis in *Anagallis arvensis* occur typically during the generative phase of development. In the vegetative phase phyllotaxis usually does not change. An exception to this rule is one peculiar population of *Anagallis arvensis*, belonging to a pink-flowered form *carnea*, in which phyllotaxis often transforms in both the generative and vegetative phase of development. The presence of a relatively large number of vegetative transitions, virtually absent in two control red-flowered populations of form *arvensis*, is accompanied also by the higher frequency of generative transitions. This might be a result of a genetically determined instability of shoot apex geometry. It has been proved experimentally that changing conditions of growth affect the transition frequencies and the time lapse from the beginning of either the vegetative or generative phase to the transition (measured in plastochrons). A comparison of two types of transitions shows that divergence changes are less abrupt in vegetative transitions. The differences between the two types also pertain to the vertical spacing of leaves and to the pattern formed by leaf bases and wings on the stem surface in the transition zone.

KEY WORDS: *Anagallis arvensis*, phyllotactic transitions, intraspecific variation.

INTRODUCTION

Although a phyllotactic pattern is traditionally regarded as characteristic for a plant species and used as a taxonomic trait, examples of plants with variable phyllotaxis are well documented. The variation results from transformations of phyllotaxis. The best known are transformations associated with a change in ontogenetic phase of development, as in members of *Scrophulariaceae* (Szafer, Kulczyński, Pawłowski 1967), *Silene* (Lyndon 1978), *Ranunculus* (Meichenheimer 1979), and *Epilobium* (Meichenheimer 1982). They may be either quantitative or qualitative. The first type is characteristic for *Ranunculus*; the latter – for the other species listed above. In some plants, however, also shoots in the same phase of development may undergo transformations. This is true for vegetative axes of *Ambrosia artemisiaefolia* (Soma, Kurijama 1970), *Bougainvillea glabra* (Puławska 1972), *Bryophyllum tubiflorum* (Gómez-Campo 1974), *Abies balsamea* (Zagórska-Marek 1985), *Juniperus chinensis* (Meichenheimer, Zagórska-Marek 1989; Zagórska-Marek unpublished data), for *Magnolia* flowers (Zagórska-Marek 1994), capitula of *Carlina acaulis* (Szymanowska-Pułka 1994), and finally also for microphyll arrangement on *Lycopodium* stems (Gola 1996). Frequencies of various phyllotactic patterns may vary among different members of the same genus or other closely related taxa. Various *Magnolia* species are the best example of this variation: in gynoecea of *M. soulangeana* the main Fibonacci pattern is dominating; whereas in *M. acuminata* many patterns are frequent (Zagórska-Marek 1985, 1994). Two genera of *Cactaceae*

are yet another case of this phenomenon. *Mammillaria* has different phyllotaxis spectrum than *Rebutia*; the latter resembling generative structures of *M. acuminata* (Gola 1997). The same is true for different taxa of *Crassulaceae* (Jensen 1968), especially species of *Sedum* (Astruc 1949, Deschatres 1954), and for various species of *Lycopodium* (Gola 1996). Interestingly, pattern frequencies may vary also among individuals or populations of the same species. Intraspecific variation of phyllotaxis, however, has not yet been studied extensively. The only example is that of *Magnolia acuminata* where frequencies of phyllotactic patterns in gynoecea are not the same in different trees (Zagórska-Marek 1994), and for each tree typical distribution of pattern frequencies is maintained in consecutive years (Zagórska-Marek personal communication). Plants of *Lycopodium annotinum* deriving from various locations differ in the arrangement of their microphylls (Gola unpublished data). The origin of these differences may lay either in environmental conditions or in the genetic factor. Differentiating between these two will be possible in experiments similar to those presented in this work, in which *Anagallis arvensis* has been used as a model plant.

In *Anagallis arvensis* a variation of phyllotaxis in generative shoots is known from literature (Kwiatkowska 1992, 1995b). It results from qualitative phyllotactic transitions hitherto known to occur only in a generative phase of shoot development. They are further referred to as generative transitions. A shift from the vegetative to generative ontogenetic phase in this species is accompanied by relatively subtle reorganization in the shoot apex activity. The only change is that

flower primordia arise in leaf axils instead of lateral shoots, whereas the formation of leaf primordia and vertical growth of a main shoot are continued. Initially both types of axillary primordia are very similar (Vaughan 1955, Green, Havelange, Bernier 1991). However, flower primordia appear about two plastochrons earlier than lateral shoot primordia. They are initiated already during the first plastochron of the development of the subtending leaf primordia. Regular ontogenetic sequence of phyllotactic patterns is characteristic for *Anagallis* (Kwiatkowska 1994, 1995b). Decussate phyllotaxis typically persists during a vegetative development. When a generative phase begins, decussate pattern often changes, sooner or later, into a spiral Fibonacci pattern. This one sometimes is transformed into whorled trimerous phyllotaxis, which in a small percentage of shoots appears directly after the decussate pattern. Trimerous phyllotaxis may be further transformed into the spiral Lucas pattern, this however, is the least common type of transition.

Although phyllotaxis of vegetative shoots of *Anagallis arvensis* is typically stable, i.e. vegetative transitions of the decussate pattern do not take place, a single population of this species has been found in which this rule is not observed. Moreover, in this peculiar population the frequency of generative transitions is also different than in others. These deviations pose two provoking questions. The first is about the reasons of variation in phyllotactic transition frequencies among populations of the same species. The second refers to possible differences between the course of vegetative and generative transitions in *Anagallis*. These two problems became the subject of the presented investigation.

MATERIALS AND METHODS

Anagallis arvensis is a model long-day plant which requires only one day 18hrs long to be induced to flowering (Ballard

1969, Brulfert, Fontaine, Imhoff 1985). The plant can be easily propagated by cuttings in the vegetative phase of development (Brulfert, Fontaine, Imhoff 1985). Thus the same genotype can be propagated indefinitely. When shoot phyllotaxis in generative phase is spiral, pseudowhorls are formed (Kwiatkowska 1994, 1995a, b). These are groups of spirally arranged leaves between which internodes are virtually not elongating. Internodes between adjacent pseudowhorls are as long as those of shoots with whorled phyllotaxis. Stems of *Anagallis* are winged due to the extension of leaf margins from leaf bases downward (De Sloover 1958). It typically results in connection of two wings with every leaf. A pattern formed by leaf bases and wings on the stem surface is characteristic for every type of phyllotaxis (Kwiatkowska 1994). In spiral phyllotaxis the pattern is also affected by uneven elongation of internodes.

Three wild *Anagallis* populations of different origin were analysed. One of them was a weed in the Botanical Garden of Wrocław University (seeds and specimens collected in 08.1990); others came from Przeclawice in the vicinity of Trzebnica (09.1991) and from Rząśnik in the Kaczawskie Mts (08.1992). Plants of these populations belonged to two different forms of *Anagallis arvensis* L. (Kornaś 1962). These from Rząśnik represented a pink-flowered form *carnea* (Schrank) Schinz, whereas plants from the other two populations belonged to the red-flowered form *arvensis* L. (Fig. 1).

In 1993 seeds collected from natural habitats of these three populations were sown and about ten plants of every provenience were obtained. All these plants were grown in conditions not inducing flowering, in a phytotron at the Institute of Botany, Wrocław University. Later, they were propagated only vegetatively, i.e. by cuttings, always in a standard medium, in a way described by Brulfert, Fontaine, Imhoff (1985). A number of experiments has been designed in which the plants deriving from three different populations were treated with various growth conditions.



Fig. 1. Flower corollas of plants representing two different forms of *Anagallis arvensis* used in the present investigation: f. *carnea* (the Rząśnik population) on the right and f. *arvensis* (the Wrocław population) on the left. Bar = 1 cm.

Experiments SD1-4 were carried out in a phytotron under short days (SD; 10hrs day/14hrs night) with illumination of 100-120 $\mu\text{Mm}^{-2}\text{s}^{-1}$ of photosynthesis active radiation (PAR) measured with photometr FF-01; with temperature and humidity not controlled:

- SD1 – from 4.09.1995 to 23.11.1995;
- SD2 – from 23.11.1995 to 27.04.1996;
- SD3 – from 27.04.1996 to 28.08.1996;
- SD4 – from 28.08.1996 to 21.11.1996.

SD5 was carried out in the other room under the same day length and illumination; with temperature and humidity not controlled; from 21.11.1996 to 28.02.97.

LD1 was performed outdoors in conditions inducing flowering (LD); from 27.04.1996 to 23.06.1996.

LD2-3 – were conducted in a phytotron under long days (LD; 16hrs day/8hrs night), illumination of 25 $\mu\text{Mm}^{-2}\text{s}^{-1}$ of PAR, with temperature 26°C and humidity not controlled:

- LD2 – from 10.09.1996 to 25.11.1996;
- LD3 – from 21.11.1996 to 3.03.1997.

For every population a stability of phyllotaxis was determined on a basis of a frequency of phyllotactic transitions in each experiment. Only transitions from decussate phyllotaxis to the spiral Fibonacci or to trimerous pattern were considered. The percentage of shoots with transforming decussate phyllotaxis from the total of all the axes with at least two elongated internodes was calculated. The data on the number of elongating internodes in a sample of at least thirty shoots per population for every experiment carried out under short days, were also gathered. In experiments performed in inductive conditions, numbers of vegetative and flowering nodes in elongating parts of all the shoots were reckoned. In shoots with transitions of decussate phyllotaxis, decussate nodes (vegetative and flowering) were counted from the base of a shoot to the level where the transition occurred. The pattern formed by leaf bases and wings on the stem surface within the transition zone was analysed for every axis. In samples of shoots from all the experiments angles between leaves were measured in the serial transverse microtome sections made of the transition zone.

A number of shoots from every experiment (about 50 vegetative and 30 generative axes) was fixed in formalin-aceto-al-

cohol, sectioned in paraffin (serial transverse sections 10 μm thick) and stained with Erhlichs hematoxylin (Johansen 1940). Phyllotaxis of all these shoots was determined in serial transverse sections in a way described earlier (Kwiatkowska 1992, 1995b). The divergences were measured with the aid of computer program PHYL-2 for Indy (Silicon Graphics), written by Radosław Karwowski, M.S., from the Institute of Botany, Wrocław University on images of transverse sections obtained with the Scalar VMS 70 video lupe. Because the number of generative shoots with nondecussate phyllotactic patterns was insufficient for qualitative analysis of leaf spacing, the plant material collected from natural habitats of the three populations (about 60 generative axes) was also sectioned. It was used for the comparison of leaf spacing only, and not for the calculation of phyllotactic transition frequencies.

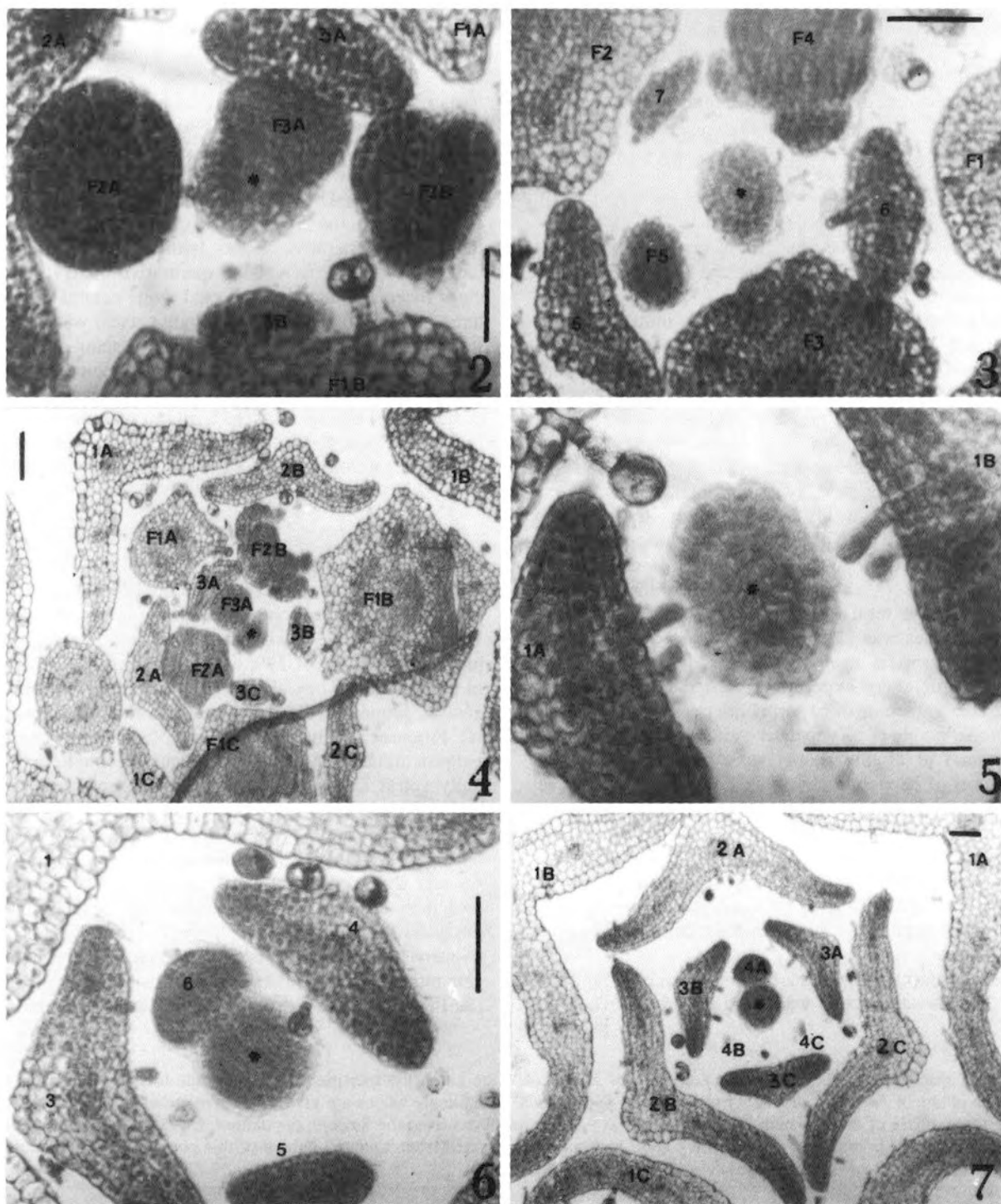
Numeric data concerning divergencies as well as numbers of nodes and internodes were analysed with the aid of CSS Statistica 3.1 (Statsoft Inc. 1993, Tulsa). Certain mean values were tested for a difference with t-test for independent samples.

RESULTS

Phyllotaxis of generative shoots of *Anagallis arvensis* plants belonging to populations growing in natural habitats in Wrocław and Przecławice, has been described in the previous paper (Kwiatkowska 1995b). Four patterns were encountered in those plants. They always occurred in a characteristic ontogenetic sequence: the decussate pattern was followed by spiral Fibonacci; trimerous (in very few shoots the trimerous pattern immediately followed decussate phyllotaxis); and finally spiral Lucas pattern. In plants propagated vegetatively and grown in inductive conditions (experiments LD1-3; Figs 2-4), belonging to all the examined populations, the first three patterns occurred in the same sequence. Only Lucas phyllotaxis was absent. This, however, is the least common pattern in *Anagallis arvensis*. In vegetative shoots (experiments SD1-5) the same three patterns were observed (Figs 5-7) as in generative ones. Differences between divergences in vegetative and generative phases are statistically insignificant (Tab. 1). Spiral Fibonacci and trimerous phyllotaxis occurred predomi-

TABLE 1. Leaf spacing in stable phyllotactic patterns and in the transition zone (the transition from decussate to spiral Fibonacci phyllotaxis exclusively) in plants of *Anagallis arvensis* from different populations. Mean angle values are given in degrees, standard error – in parentheses. V – the vegetative phase of development; G – generative; A – shoots of plants from the Rzęśnik population, experiments SD1-5 (V) and LD1-3 (G); B – the Przecławice population, experiments SD1-5 and material collected from a natural habitat of this population; C – material collected from a natural habitat of the Wrocław population.

Plants provenience	A		B	B + C
	V	G	V	G
Stable decussate phyllotaxis:				
– angles between leaves of the same node	181.6 (1.6)	177.0 (3.8)	179.3 (1.4)	180.0 (1.0)
– angles between leaves of adjacent nodes	89.7 (1.5)	91.1 (5.8)	92 (1.3)	90.0 (1.4)
Angles along the ontogenetic spiral in the spiral Fibonacci pattern	140.0 (1.8)	140.5 (1.5)	--	139 (1.0)
Angles in the transition zone:	154 (3.4)	168.7 (4.1)		175.0 (2.5)
Between leaves of the last decussate node				
Between successive leaves along the detectable ontogenetic helix (ascending):				
– between the leaf of the last decussate node and the successive leaf on the helix	131 (7.9)	121.1 (5.3)		121.8 (2.3)
– between the first two spirally arranged leaves	144 (2.7)	146.1 (3.5)		146.8 (2.3)
– between the second and the third of spirally arranged leaves	127 (4.1)	135.6 (6.8)		131.7 (2.0)
Percentage of shoots with wings not connected with any leaf from all the shoots exhibiting a phyllotactic transition	43%	4%	--	8%



Figs 2-7. Photomicrographs of transverse sections of generative (Figs 2-4) and vegetative (Figs 5-7) shoot apices of *Anagallis arvensis* exhibiting different phyllotactic patterns. The dome of every apex was indicated with an asterisk. Every leaf primordium was labelled with a number depending on its age or the age of a whorl to which it belongs. The younger the primordium or the whorl, the bigger the number. In whorled phyllotaxis a letter follows every number in order to differentiate between members of one whorl. Every floral primordium was labelled with "F" and a symbol of a leaf primordium in which axil it arises. Five shoots (Figs 3-7) are of plants from the Rzańnik population; one shoot (Fig. 2) – from the Przeclawice population.

Fig. 2. A generative apex exhibiting the decussate pattern with distinct anisoclady: F3A is apparent whereas F3B is not big enough to be sectioned in the same plane. An anisophylly is manifested by the difference between the size of 3A and 3B. Bar = 10 μ m.

Fig. 3. An apex with the spiral Fibonacci pattern. An ontogenetic spiral is ascending counterclockwise. Bar = 10 μ m.

Fig. 4. An apex with trimerous phyllotaxis. Both anisoclady (e.g. in the youngest whorl) and anisophylly (3A is bigger than 3B and 3C) are manifested. Although the arrangement of leaf primordia composing older whorls is distorted due to the rapid growth of floral primordia, orthostichies are apparent between 1A and 3A; 1B and 3B; etc. Bar = 100 μ m.

Fig. 5. A vegetative apex exhibiting decussate phyllotaxis. Only one pair of leaf primordia (1A and 1B) is apparent. No lateral shoot primordia are present. Bar = 10 μ m.

Fig. 6. An apex with spiral Fibonacci phyllotaxis and the ontogenetic spiral ascending clockwise. Bar = 10 μ m.

Fig. 7. Trimerous phyllotaxis. Due to anisophylly 4A is apparent whereas 4B and 4C are much smaller, and only their very tips can be seen in this section. Bar = 100 μ m.

TABLE 2. Frequencies of vegetative and generative phyllotactic transitions in three populations of *Anagallis arvensis*. Only axes with at least two elongated internodes were counted. Standard error (in B, C, E) or percentage of all the axes under consideration (in D) are given in parentheses. A – a number of all the axes in the experiment; B – a mean number of elongated internodes in a vegetative part of a shoot (in LD1-3 – internodes preceding a generative shoot portion); C – a mean number of flowering nodes; D – a number of axes with phyllotactic transitions: vegetative (SD1-5) or generative (LD1-3); E – mean number of decussate nodes (vegetative in SD1-5 or flowering in LD1-3) preceding the transition level. Experiment labelling corresponds to that in the text.

Population origin	A	B	C	D	E
Rzańnik:					
SD1	333	7.0 (0.27)	--	39 (12%)	10.2 (0.56)
SD2	431	8.3 (0.51)	--	77 (18%)	10.5 (0.37)
SD3	246	11.7 (0.52)	--	4 (2%)	15.8 (--)
SD4	501	8.3 (0.27)	--	48 (10%)	10.2 (0.53)
SD5	556	5.3 (0.16)	--	236 (42%)	8.2 (0.02)
LD1	75	1.3 (0.20)	2.6 (0.14)	40 (53%)	3.6 (0.12)
LD2	69	3.1 (0.22)	4.6 (0.16)	10 (15%)	5.6 (0.40)
LD3	98	3.3 (0.21)	3.5 (0.24)	30 (31%)	4.8 (0.04)
Przeclawice:					
SD1	595	7.8 (0.48)	--	0	--
SD2	430	11.4 (0.72)	--	0	--
SD3	181	12.3 (0.43)	--	1 (0.6%)	0 (--)
SD4	537	8.1 (0.21)	--	3 (0.6%)	2.3 (--)
SD5	397	7.3 (0.03)	--	3 (0.01%)	6.3 (--)
LD1	306	1.1 (0.09)	2.6 (0.07)	29 (9%)	4.0 (0.33)
LD2	150	3.3 (0.14)	4.9 (0.14)	0	--
LD3	166	3.8 (0.18)	4.6 (0.14)	0	--
Wrocław:					
SD1	209	6.8 (0.38)	--	0	--
SD2	291	11.2 (0.68)	--	1 (0.3%)	11 (--)
SD3	226	12.5 (0.74)	--	0	--
SD4	366	8.2 (0.20)	--	0	--
SD5	314	6.8 (0.04)	--	0	--
LD1	209	1.0 (0.09)	2.7 (0.07)	29 (14%)	4.2 (0.13)
LD2	131	3.0 (0.12)	5.8 (0.13)	0	--
LD3	88	3.6 (0.23)	4.4 (0.21)	0	--

nantly in shoots of plants from the Rzańnik population characterised by a high frequency of decussate transitions (Tab. 2), and were practically absent in the others. The ontogenetic sequence of patterns in vegetative axes was identical with that in generative ones. An immediate transition from decussate to trimerous pattern was observed in as few as 4% of vegetative axes showing any type of the decussate transition.

In generative apices representing any of the three phyllotactic patterns, floral primordia grew to the size of the apex dome while they were still in its close vicinity (F3A in Figs 2, 4, F5 in Fig. 3). They were often bigger than leaf primordia of a similar age, especially in whorled phyllotaxis (compare 3B with F3A in Fig. 2; F3A with 3B in Fig. 4). This was never a case in vegetative apices.

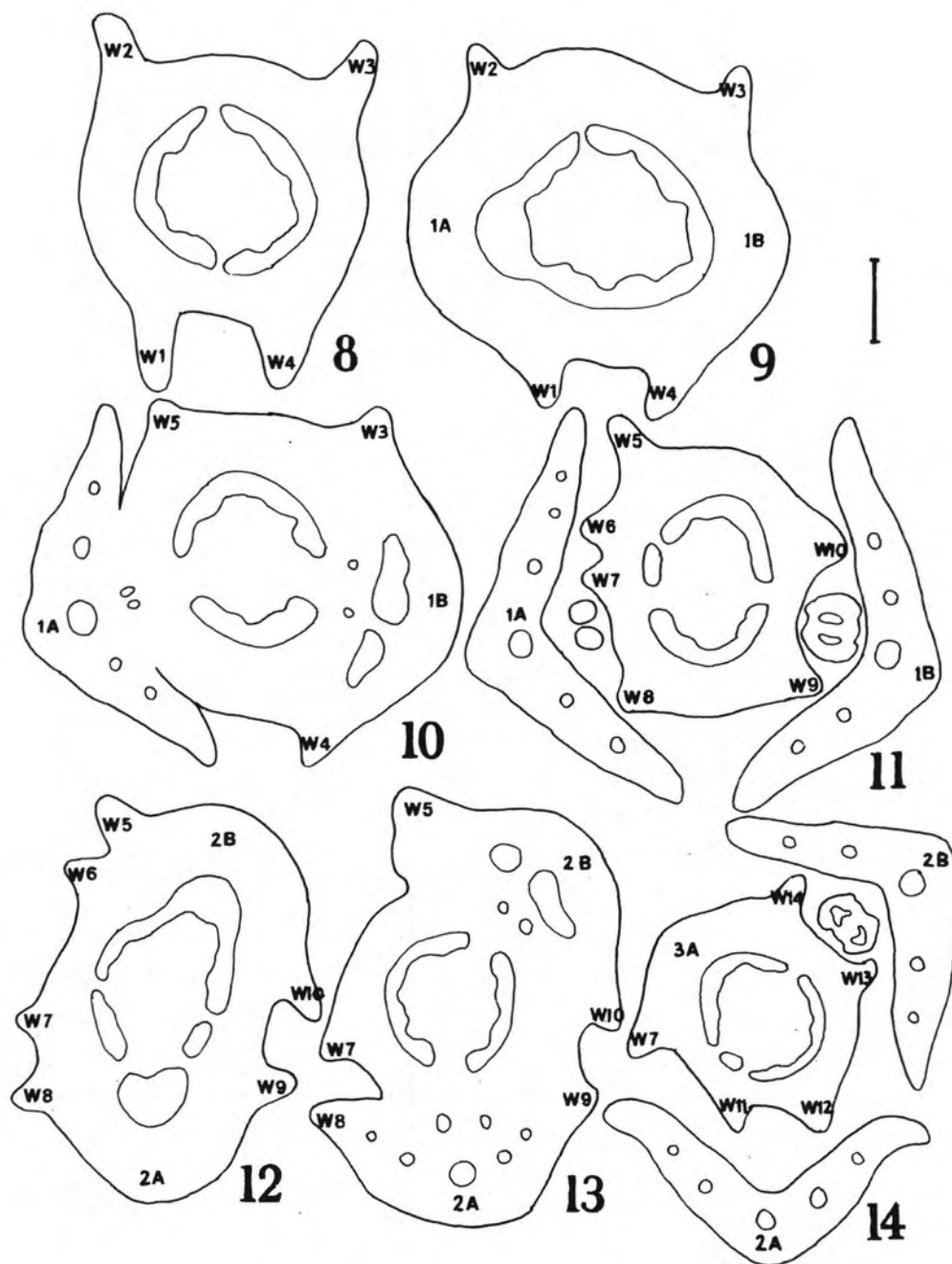
In elongating parts of vegetative shoots of *Anagallis* with whorled leaf arrangement helical anisoclady (Cutter 1972, Rutishauser 1981, Kwiatkowska 1994, 1995b) was manifested by different lengths of lateral shoots attached at the same node. It was apparent also in generative whorled apices (Figs 2, 4) where floral primordia of the same node differed in size. In vegetative apices anisoclady was not manifested, since lateral shoot primordia arise two plastochrons later than floral primordia (Fig. 7). Anisophylly was

characteristic for both vegetative (Fig. 7) and generative (Figs 2, 4) apices with whorled phyllotaxis.

Pseudowhorls were formed in all the axes, generative and vegetative, with spiral Fibonacci phyllotaxis. They consisted of two or three leaves attached at almost equal levels. The number of wings extending within the elongating internode, basipetally adjacent to the pseudowhorl, varied from five to six regardless the number of leaves in the pseudowhorl.

Both in vegetative and in generative shoots decussate phyllotaxis transformed to the spiral Fibonacci, or less often to the trimerous pattern. A shoot segment referred to as a transition zone included: the last two-leaved node of decussate phyllotaxis with four wings extending basipetally; an elongating internode adjacent acropetally; and the subsequent pseudowhorl composed of two or three spirally arranged leaves with more than four wings, or a three-leaved trimerous whorl. It has already been shown (Kwiatkowska 1992, 1995a, b) that in *Anagallis* shoots with unchanging decussate phyllotaxis leaves composing one whorl are usually shifted vertically with respect one to another. Therefore the borders of the transition zone were estimated on the basis of the circumferential spacing of leaves and a number of wings connected with them.

Generative transitions from decussate to spiral Fibonacci phyllotaxis were identical in shoots of all the three populations examined. They differed from those in vegetative shoots (Tab. 1). During the vegetative transitions the last decussate node preceding Fibonacci phyllotaxis (i.e. the older node of the transition zone) was less symmetrical than in generative (Tab. 1; Figs 9-10, 21). In former the two leaves were c. 150° apart, whereas in latter – c. 170°. This difference is statistically significant. Also the vertical spacing of leaves in the transition zone was not the same in transitions of the two developmental phases. In generative shoots all the leaves in the transition zone could be easily connected by a detectable ontogenetic helix. The vertical displacement of leaves composing the last decussate node was concurrent with leaf sequence on the helix (Figs 33-36). Angles between successive leaves along the helix slowly approached the Fibonacci divergence (Tab. 1). In the transition zone of vegetative shoots angles also fluctuated around the Fibonacci divergence (Tab. 1). However, successive leaves along the detectable helix, especially those of the last pair, were most often attached at levels not concurrent with leaf sequence along the helix. For example leaf 1B in Figs 9-10 arises at the level higher than 1A although as suggested from the circumferential leaf spacing, it should precede 1A along the detectable helix ascending clockwise. The same applies to the leaves 1A and 1B in Fig. 21 which could be connected with a counterclockwise helix. Also the pattern formed by leaves and wings on the stem surface in the transition zone was often different in vegetative and generative shoots. Wings not connected with any leaf margin (Figs 11-13 and 15-17; 23-26 and 18-20) were much more common in vegetative than in generative transition zones (Tab. 1). The basal end of these atypical wings appeared in an axil of a leaf belonging to the last decussate node (W6 in axil of 1A in Figs 11, 15, 22), like in case of typical wings. The apical end was located at the level of the subsequent pseudowhorl, and was not connected with any leaf margin (Figs 13, 17, 26). Atypical wings passed an internode always



Figs 8-14. Contours of transverse sections through the transition zone of a vegetative shoot from the Rzaśnik population. Outlines of stems and leaf bases were made with heavy lines; those of vascular bundles – with thin lines. Every leaf was labelled with a number and a letter. The number depends on the age of a whorl or a pseudowhorl to which the leaf belongs. The younger the whorl or the pseudowhorl, the bigger the number. A letter follows every number in order to point out a particular leaf. Wings are labelled with "W" followed by a number to differentiate between various wings. The decussate pattern is transformed into a spiral Fibonacci. A detectable ontogenetic helix, ascending clockwise, passes from leaf 1B to 1A, then further to 2B, 2A, and to 3A. Levels at which 1A and 1B arise are in disagreement with their order along the helix. The same concerns leaves 2A and 2B. Bar = 10 μ m.

Fig. 8. A section through an internode basipetal to the last decussate node preceding Fibonacci phyllotaxis. Four wings (W1-4), two per leaf, are present.

Fig. 9. A section at the base of the last decussate node. 1A is attached slightly lower than 1B, the leaf spacing is distinctly asymmetric, i.e., the upper angle between 1A and 1B is bigger than the lower one.

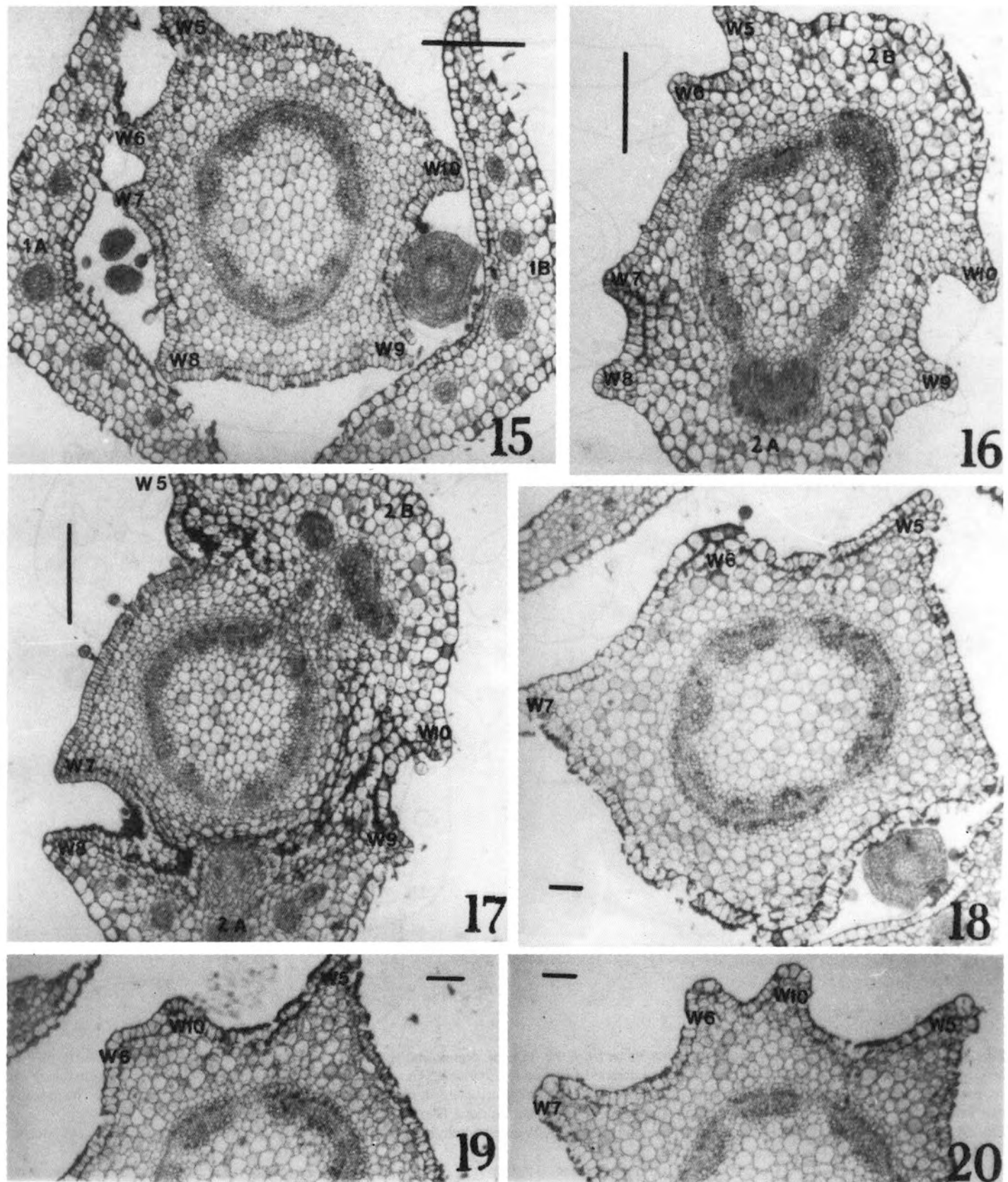
Fig. 10. An apical part of the same node. A new wing W5 is visible in the axil of 1A.

Fig. 11. A basal part of an internode following acropetally. Six (W5-10) instead of four wings are present. This is the only one elongating internode included in the transition zone.

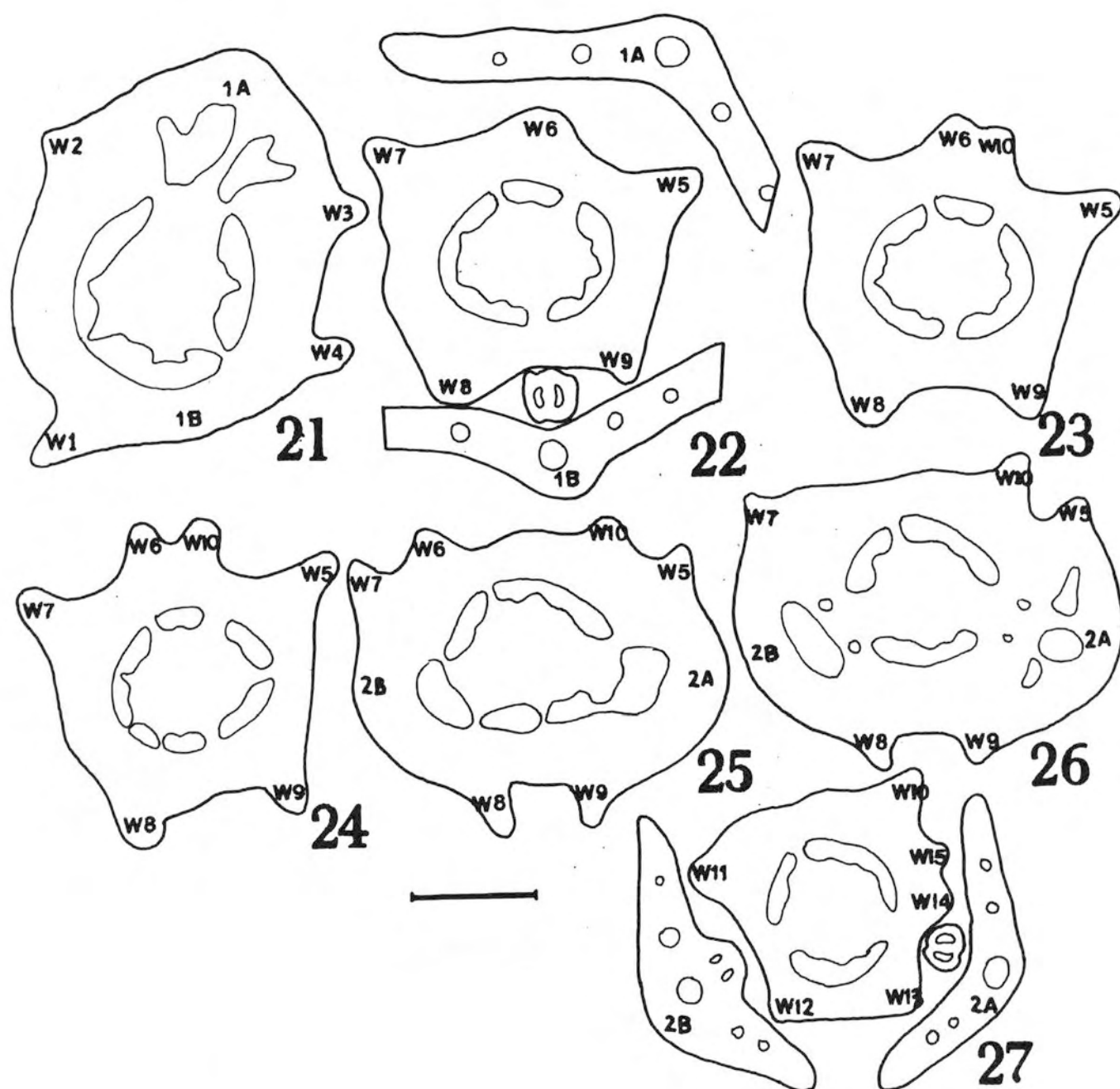
Fig. 12. A base of the subsequent pseudowhorl consisting of two asymmetrically spaced leaves (2A and 2B) arising at slightly different levels. W6 and W7 are not connected with any leaf of this pseudowhorl.

Fig. 13. An apical part of the same pseudowhorl. W6 is no longer continued. Its apical end has not been connected with any leaf base. W7 has passed through the pseudowhorl level.

Fig. 14. A basal part of the following pseudowhorl. New wings (W11-14) appeared in axils of 2A and 2B. One of them (W14) is connected with 3A as well as the long wing W7.



Figs 15-20. Photomicrographs of the transverse sections which contours are presented in Figs 11-13 (photomicrographs 15-17) and Figs 22-24 (photomicrographs 18-20). They show an atypical wing not connected with any leaf margin and the splitting of wings, respectively. In Figs 19-20 only the upper part of a section, where the wing splitting takes place, was shown. Labelling as in Figs 8-14. Bars = 10 μ m.



Figs 21-27. Another transition zone of a vegetative shoot from the Rzańnik population shown in contours of transverse microtome sections. Labelling as in Figs 8-14. The decussate pattern is transformed into spiral Fibonacci. The detectable ontogenetic helix, ascending counterclockwise, passes from leaf 1B to 1A, 2B, and 2A. Levels at which 1A and 1B arise are in disagreement with their order along the helix. The same is true for leaves 2A and 2B. One of wings (W6) appearing in the transition zone is splitting. Bar = 500 μ m.

Fig. 21. A section through the base of the last decussate node. 1A is attached slightly lower than 1B, the leaf spacing is distinctly asymmetric. All the four wings (W1-4) are connected with leaf margins.

Fig. 22. A basal part of the long internode of the transition zone. It follows the node from Fig. 21 acropetally. Five wings (W5-9) are present.

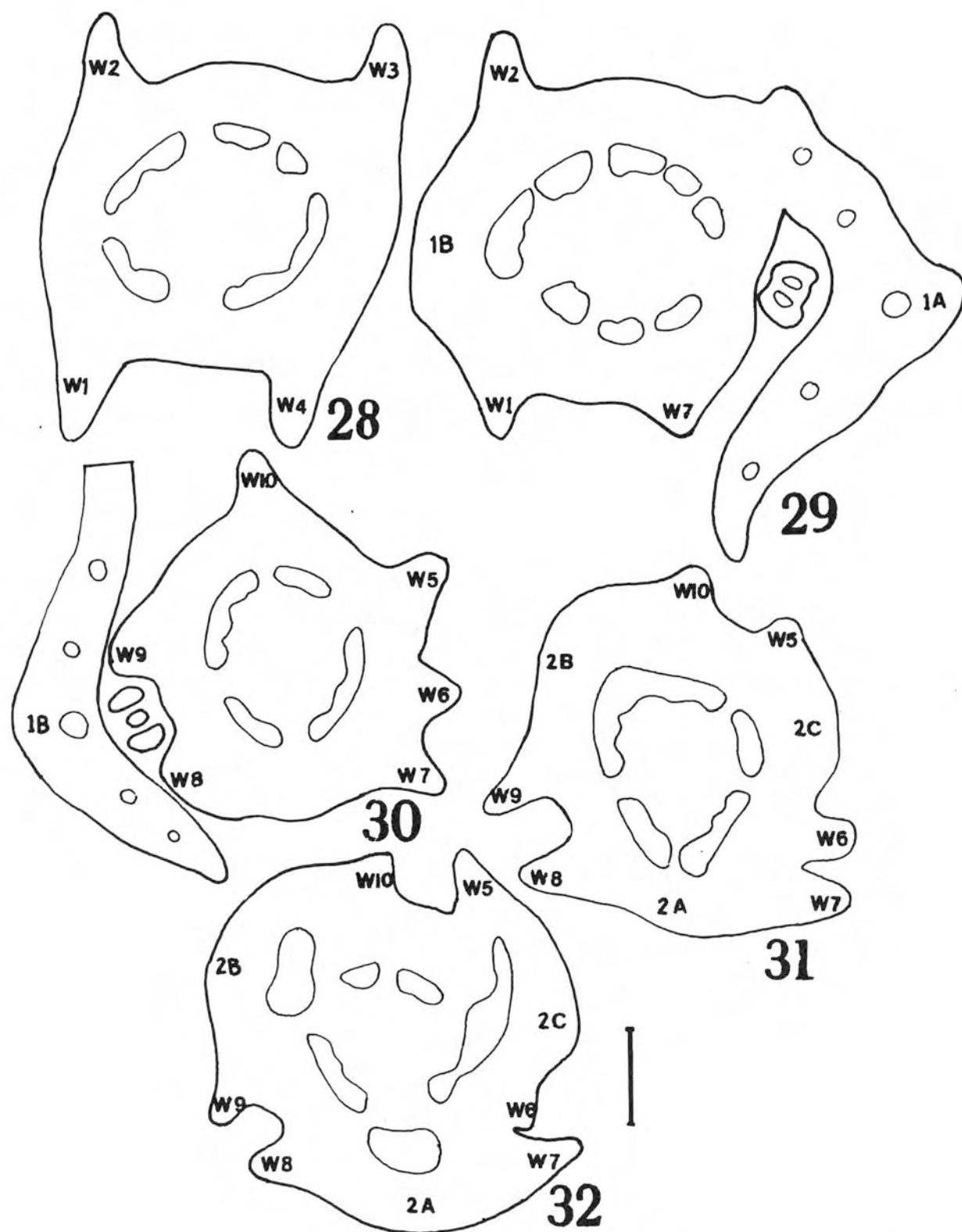
Fig. 23. A section at the level 50 μ m higher than that in Fig. 22. An additional wing (W10) becomes apparent due to the splitting of W6.

Fig. 24. A middle part of the same internode. Six distinct wings are present.

Fig. 25. A base of the following pseudowhorl consisting of two asymmetrically spaced leaves, 2A and 2B, arising at slightly different levels. W6 and W10 are not connected with any leaf of this pseudowhorl.

Fig. 26. A middle part of the same pseudowhorl. W6 is no longer continued. It is an atypical wing, not connected with any leaf margin.

Fig. 27. A base of the internode following acropetally. New wings (W11-15) appeared in axils of 2A and 2B. W10 is visible in this section since it has passed a level of the pseudowhorl shown in Figs 25-26.



Figs 28-32. An immediate transformation of decussate phyllotaxis into trimerous pattern in vegetative shoot portion. Labelling as in Figs 8-14. Bar = 10 μ m.

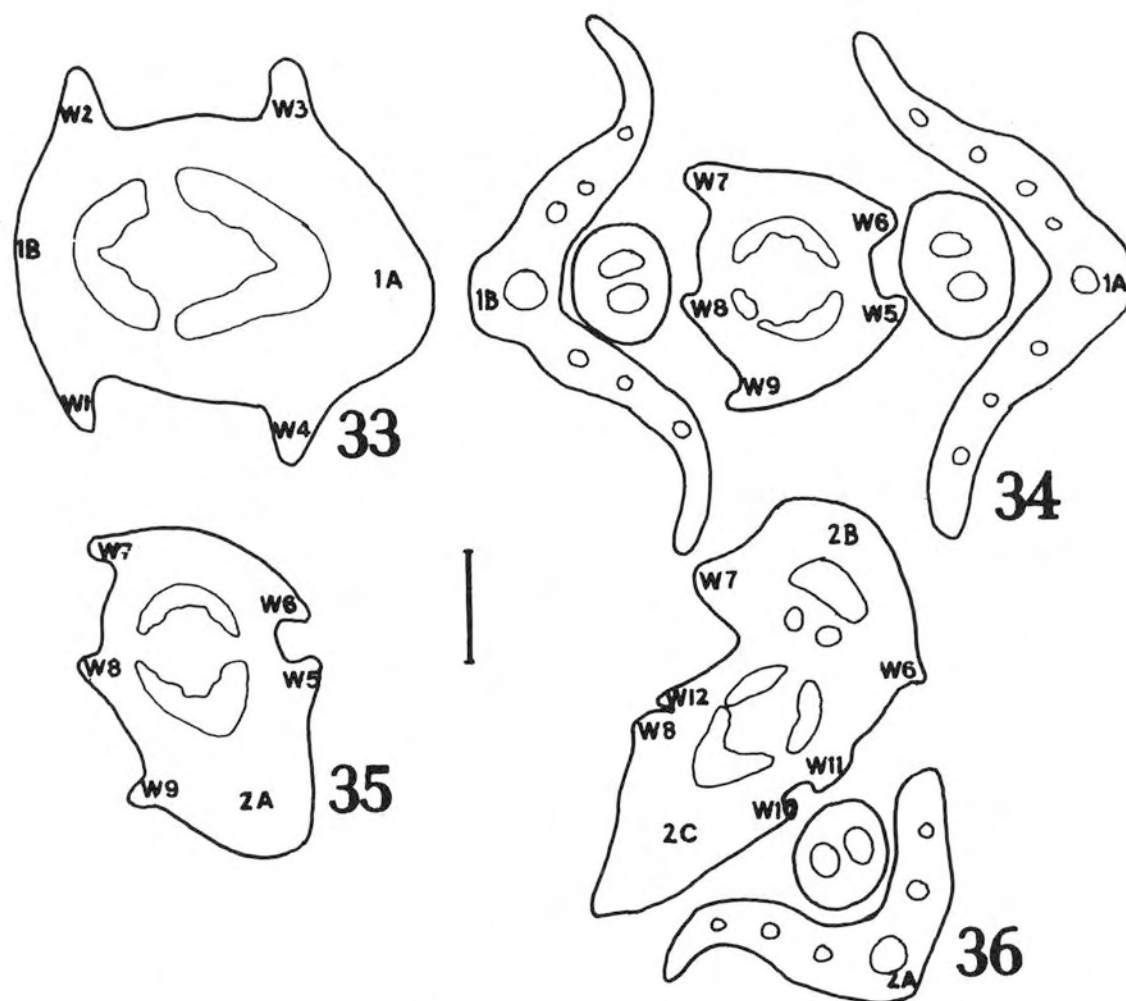
Fig. 28. A section through the internode preceding the last decussate node before the spiral Fibonacci pattern. Four wings (W1-4) are visible.

Fig. 29. A section through the last decussate node. Although 1A is attached slightly lower than 1B, leaf spacing in this node is almost symmetric. All the wings from the preceding internode (W1-4) are connected with margins of 1A and 1B. A new wing (W7) is already apparent.

Fig. 30. A basal part of the internode following acropetally. Six wings (W5-10) are present.

Fig. 31. A base of the following whorl consisting of three asymmetrically spaced leaves (2A-C) arising at almost equal levels. All the wings are connected with leaf margins.

Fig. 32. The middle part of the same whorl.



Figs 33-36. A transition zone of a generative shoot from the Rzaśnik population. The decussate pattern is transformed into spiral Fibonacci. A detectable ontogenetic helix ascending counterclockwise passes from leaf 1A to 1B, 2A, 2B, and 2C. Levels at which all the leaves in the transition zone are attached are in agreement with their order along the helix. Bar = 10 μ m.

Fig. 33. A section through the base of the last decussate node. 1A is attached lower than 1B, leaf spacing in almost symmetric. All the four wings (W1-4) are connected with margins of 1A and 1B.

Fig. 34. A basal part of a long internode of the transition zone. Five (W5-9) wings are present.

Fig. 35. The base of the following pseudowhorl. W5 and W9 are connected with 2A.

Fig. 36. The middle part of the same three-leaved pseudowhorl (2A-C). New wings W10 and W11 have appeared in the axil of 2A, and the other one W12 – on the opposite side of the stem. W6 and W7 are connected with 2B; W8 and W10 – with 2C. Since this is a young, only slightly elongated part of the stem, the recognition of pseudowhorls may be inaccurate.

in a certain sector of the stem circumference, i.e. in the wider of two sectors positioned between two typical wings connected with different leaves of the pseudowhorl (the left part of the stem in Figs 11-13 and 15-17; the upper stem sector in Figs 23-26). No atypical wings occurred in these few vegetative shoots where decussate phyllotaxis transformed immediately to the trimerous pattern (Figs 28-32). In generative shoots the lack of atypical wings was characteristic for transitions of the decussate pattern both to the spiral Fibonacci (Figs 33-36) and to trimerous phyllotaxis.

In a few vegetative shoots some wings splitted within a long internode of the transition zone (Figs 18-20 and 22-24). The split took place again in the wider stem sector of this internode.

Plants were grown for a different time period (2-5 months) depending on the experiment. Their shoots achieved different lengths measured in a number of elongated internodes. These values were similar for all the populations in the same experi-

ment both for vegetative and generative shoot portions (Tab. 2). Significant differences between the three populations were in frequencies of vegetative and generative transitions of the decussate phyllotaxis (to the spiral Fibonacci or to trimerous pattern). In experiments SD1, 2, 4, 5 numerous vegetative transitions took place in plants from the Rzaśnik population (in 10-40% of axes), and were almost absent in others (less than 1% of shoots). Only in a single experiment, SD3, the number of transformed shoots of plants from all the populations was very low. The percentage of generative shoots exhibiting phyllotactic transitions was also the highest in plants from the Rzaśnik population (Tab. 2). Especially in LD2 and LD3, the decussate pattern was transformed in a large number of shoots from this population, whereas no transitions occurred in others.

Mean numbers of decussate nodes counted from the base of an axis to the level of the vegetative transition showed no statistically significant differences for plants from experiments

SD1, 2, 4 (Tab. 2). In SD5 the number of nodes preceding the transition was significantly smaller than in other cases. This experiment, however, was performed in another room with an identical illumination. Unfortunately, only the illumination was controlled both in SD1-4 and in SD5, and no information about other growth conditions in these rooms possibly responsible for the above effect is available. In SD3 phyllotaxis of only four shoots was transformed thus the statistical comparison of these data with the others is impossible. The mean numbers of decussate flowering nodes preceding generative transitions, i.e., the time interval from the beginning of the generative phase to the moment of phyllotactic transition, showed no statistically significant differences between experiments performed in the same conditions (LD2 and LD3). For LD1, carried out outdoors, the number of nodes was different.

DISCUSSION

Results of the experiments with various growth conditions applied to plants from three different populations have proved that the variation in phyllotaxis stability in *Anagallis arvensis* is genetically determined. Differences between the Rzaśnik population and the other two populations have been prominent in seven of eight experiments, during which plants from all the populations grew in the same environmental conditions, and the conditions differed from one experiment to another. The phyllotaxis stability has been determined on the basis of the frequency of phyllotactic transitions. In the Rzaśnik population belonging to form *carnea*, the initial decussate pattern is evidently less stable than in other populations, i.e. it is more easily transformable into other phyllotactic patterns. It is true for both the vegetative and generative phase of development. This trait can be added to that of the corolla colour being genetically determined, as both colour forms were used for studies of inheritance pattern (Nilsson 1938). Similar breeding experiments with plants exhibiting different tendency to phyllotactic transitions are planned for the future.

Yet another interesting trait supporting an assumption on the genetic basis of recorded differences is the number of vegetative or generative plastochrons preceding vegetative or generative phyllotactic transitions respectively. It has been different in different experiments, but every time similar within given population. These intervals (measured in plastochrons) showed no statistically significant differences between shoots from the same population as long as the growth conditions in subsequent experiments were the same. When they differed, however, the plastochron time of transition varied despite the same day length. This is an astonishing regularity, especially since only a fraction of all the axes exhibits transitions of the decussate pattern, even in the Rzaśnik population.

Many authors have postulated that the regularity in leaf initiation pattern requires some stable parameters of shoot apex geometry, and therefore the apex geometry has to be modified when the phyllotaxis changes (Zagórska-Marek 1987, Meinenheimer, Zagórska-Marek 1989, Green, Steele, Rennich 1996). The modification of the apex geometry may be possible due to changing growth parameters of the apex dome (Zagórska-Marek 1987), or biophysical factors like the requirement of minimal energy configuration (Green 1992, Green, Steele, Rennich 1996). Similar may be also the effect of changes in the level of growth substances in the apex (Hellendoorn, Lindermayer 1974, Veen, Lindenmayer 1977, Meinhardt 1984). In case of *Anagallis* the hypothesis should be

consequently put forward that in the Rzaśnik population characterised by less stable phyllotaxis, some factors affecting the apex geometry are more flexible than in other populations. The genetic factor controlling the stability of apex geometry both in the vegetative and generative phase of development varies among the three populations studied, since the differences in transition frequencies in both ontogenetic phases in these populations are evident in the same growth conditions.

It has already been suggested that the early initiation of floral primordia in decussate shoots of *Anagallis arvensis* affects the apex geometry (Green, Havelange, Bernier 1991, Kwiatkowska 1994). They appear already in the first plastochron of leaf primordia development and attain a size comparable with the apex dome itself still before the initiation of the next pair of leaves. Moreover, this is the second, after the pair of leaf primordia, pair of appendages produced by an apex along the same axis. It may be the reason for a relatively high probability of phyllotactic transitions in the generative phase of development, which are rather absent in the vegetative phase. This explanation is supported by two facts: at first, generative transitions may not occur at all on some shoots; secondly, if they do take place they do not occur immediately after or simultaneously with the beginning of the generative phase but after an unpredictable number of plastochrons. Whether the effect of the youngest floral primordia is of biophysical (the close vicinity of large lateral appendages), chemical (e.g. the action of growth hormones produced by the lateral primordia) character, or both, is an open question. In the Rzaśnik population some additional, genetically determined, factors affecting the stability of both generative and vegetative apices may possibly be in action. They add to the effect of floral primordia in generative apices and act alone when in the vegetative phase.

The other interesting feature of *Anagallis* phyllotaxis concerns the time of transition occurrence. As already mentioned it is relatively uniform among shoots from the same population grown in experiments with identical environmental conditions. Would that mean that depending on the conditions, the number of primordia, which are to be initiated by an apex before the phyllotaxis changes, differs? Giving an answer to this question would require a thorough analysis of the effect of light, temperature etc. on the apex growth (plastochron duration, apex diameter).

A course of the transition from decussate phyllotaxis to spiral Fibonacci pattern in vegetative and generative shoots of *Anagallis arvensis* is strikingly different. Vegetative transitions are more gradual than generative; significant changes in leaf spacing are manifested already in the last two-leaved node of the decussate pattern. Moreover, in the course of vegetative transitions, atypical wings which are not connected with any leaf margin are much more common than in the course of generative transitions. Also the vertical leaf spacing during vegetative transitions is less regular than during generative. This may possibly result from the instability of apex geometry in vegetative shoots from the Rzaśnik population. All these facts might suggest the existence of at least two factors, not a single one, affecting an apex stability in *Anagallis*. One of them would cause slow changes in vegetative apices, and would add to the influence of floral primordia (the second factor) in the generative phase of development.

The frequent wings not connected with leaf margins in vegetative *Anagallis* shoots suggest that the autonomy of wings in regard to leaves in this species must be greater than De Sloover (1958) interpretation of decussate shoots implies. Atypical wings pass along an elongated internode of the

transition zone always within the wider of the two circumferential sectors between the leaves of the subsequent pseudowhorl. This sector is limited by two typical wings, both of them connected with leaf margins. Possibly then the bulking of stem surface may occur independently from the leaf primordia growth in these places where enough space is available between leaf primordia. In an elongating shoot portion this space is bordered by wings connected with margins of these leaves. The splitting of wings, again in the wider circumferential stem sector in the transition zone, may be a manifestation of the gradual character of vegetative transitions. The possible interpretation of this phenomenon is that within the transition zone the stem surface available for wings slowly increases acropetally along a primordial internode.

Although not often reported, examples of intraspecific variation of phyllotaxis are already known from literature. Flowers of various *Magnolia acuminata* trees show different spectra of phyllotactic patterns in their gynoeceia (Zagórska-Marek 1994). Arborescent species, however, do not allow easy performance of growth experiments and therefore the hypothesis on the genetic versus environmental factors controlling the variation cannot be so readily verified. *Lycopodium annotinum* populations from different regions (Finland and Poland) also have different spectra of "microphyllotactic" patterns (Gola unpublished data). In this case environmental conditions may play a main role. A better example of intraspecific variation could be delivered by capitula of cultivars and wild forms of *Asteraceae* (Battjes 1994). However, these differences in phyllotaxis may be rather the result of developmental irregularities or even fasciations (Gola unpublished data) more common in cultivars than in wild specimens. Compared with these examples *Anagallis* occurs to be a good model plant for the investigation on the intraspecific phyllotaxis variation, encouraging further studies possibly also at the molecular level.

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STRESZCZENIE

WEWNĄTRZGATUNKOWA ZMIENNOŚĆ STABILNOŚCI FILOTAKSJ U *ANAGALLIS ARVENSIS*

Charakterystyczną cechą *Anagallis arvensis* jest występowanie jakościowych zmian wzoru ulistnienia w generatywnej fazie rozwoju. W fazie wegetatywnej zmiany te najczęściej nie występują. Wyjątek stanowi unikalna populacja *Anagallis arvensis*, należąca do różowo kwitnącej formy *carnea*, na pędach której transformacje filotaksji występują często w obu fazach rozwoju. Rośliny należące do tej populacji, oprócz częstych transformacji wzoru ulistnienia w wegetatywnej fazie rozwoju, charakteryzują się także większą niż w populacjach kontrolnych (należących do formy *arvensis*) częstością występowania zmian filotaksji w fazie generatywnej. Tłumaczy się to genetycznie uwarunkowaną niestabilnością geometrii wierzchołka pędu w tej właśnie populacji. Jak wykazano eksperymentalnie, czas upływający od rozpoczęcia danej fazy rozwoju do wystąpienia transformacji filotaksji zależy od warunków uprawy roślin. Przebieg transformacji wegetatywnych różni się od transformacji generatywnych. Transformacje wegetatywne są bardziej stopniowe; zmiany dywergencji w trakcie transformacji wegetatywnej są wcześniejsze niż w przypadku transformacji generatywnych. Różnice dotyczą także osiowego rozmieszczenia liści w przejściowej strefie pędu, oraz wzoru tworzonego przez nasady liści i przez skrzydełka widoczne na powierzchni łodygi.

SŁOWA KLUCZOWE: *Anagallis arvensis*, transformacje filotaksji, zmienność wewnątrzgatunkowa.