# Chromosome numbers in species of *Alchemilla* ser. *Elatae* (Rosaceae) in Turkey

# Sema Hayırlıoğlu-Ayaz & Osman Beyazoğlu

Hayırlıoğlu-Ayaz, S. & Beyazoğlu, O., Department of Biology, Faculty of Science, Karadeniz Technical University, 61080 Trabzon, Turkey

Received 29 June 1999, accepted 2 February 2000

Hayırlıoğlu-Ayaz, S. & Beyazoğlu, O. 2000: Chromosome numbers in species of *Alchemilla* ser. *Elatae* (Rosaceae) in Turkey. — *Ann. Bot. Fennici* 37: 173–182.

This paper presents the results of a karyological analysis of 17 species of *Alchemilla* L. sect. *Alchemilla* subsect. *Calycanthum* Rothm. ser. *Elatae* Rothm. The studies were carried out on plants from northeast Anatolia (Turkey). About 75% of the species have chromosome numbers 2n = 86 to 123. A few species have a lower number. The species are polyploids like the majority of species of sect. *Alchemilla* studied so far from various geographic areas.

Key words: Alchemilla, chromosome number, flora of Turkey, Rosaceae

#### INTRODUCTION

The genus *Alchemilla* L. of the family Rosaceae consists of perennial herbs with a woody rhizome and includes more than 1 000 species. The species have a mainly holarctic distribution and are distributed at various elevations in almost all types of habitats; most frequently they occur in mountains, in subalpine and alpine zones (Izmailow 1982).

In Turkey, the genus is, according to Pawlowski and Walters (1972), represented by 3 subsections and 6 series belonging to sect. *Alchemilla*. Most of them are found in northeast Anatolia.

Alchemilla, with its numerous and variable forms, is a taxonomic challenge and it has been the subject for taxonomic studies since the end of the 19th century. Buser (1894), in his revision of

the Swiss taxa, applied a very narrow species concept and described numerous forms of *Alchemilla*, treating them as distinct species. The taxonomic tradition initiated by Buser was continued by botanists such as Juzepczuk (1941), Walters (1952), Rothmaler (1944), and Pawlowski (1972).

Cytological data for *Alchemilla* are very scarce. The papers by Turesson (1957), Wegener (1967), Izmailow (1981, 1982), and Hayırlıoğlu and Beyazoğlu (1997a, 1997b, 1997c) are particularly important; Turesson (1957) determined the chromosome numbers for 19 species, Wegener (1967) for 56, Izmailow (1981, 1982) for 11, and Hayırlıoğlu and Beyazoğlu (1997a, 1997b, 1997c) for 19.

From a cytological as well as taxonomic point of view, *Alchemilla* presents a difficult subject. Izmailow (1981, 1982) reported that all studied

species collected from the Western Carpathians (Poland) are high polyploids, and in spite of the small size of the chromosomes, it is difficult to get them sufficiently to spread and to count their numbers exactly. Diploid cytotypes are as yet unknown.

Previously, the chromosome numbers of *Alchemilla* species that belong to ser. *Sericea* and ser. *Pubescentes*, distributed in northeast Anatolia were reported (Hayırlıoğlu & Beyazoğlu 1997a). The aim of this study was to investigate the chromosome numbers of 17 species of ser. *Elatae* Rothm. In the flora of Turkey, ser. *Elatae* is represented by 24 species; the remaining 7 species (*A. amonea, A. armeniaca, A. buseriana, A. hesii, A. holocycla, A. porrectidens* and *A. sciadiophylla*) are not known from northeast Anatolia.

# MATERIAL AND METHODS

#### Plant material

The *Alchemilla* species of ser. *Elatae* used in this study were collected in the mountains of northeast Anatolia (Turkey) in July and August 1994. Voucher specimens are deposited in KTÜ. The studied specimens of each species are cited in "Results" below.

#### Cytology

Actively growing root tips were used for the chromosome counts. The roots were cleaned of soil particles, then the root tips were cut off and pretreated with 0.5% colchicine for 3 h (Beyazoğlu *et al.* 1994), and then fixed in an ethanol-acetic acid (3:1) solution for at least 24 h at 4 °C. The root tips were hydrolyzed in 1 N HCl at 60 °C for 15 min and then rinsed with tap water for a minimum of 2–3 min. Staining was carried out in Feulgen for 1.5 h. Squashing was done in 45% acetic acid and the the preparations were mounted in Entellan.

In view of the serious difficulties in karyological analysis of *Alchemilla* mentioned above, the fixation of root tips was, as a rule, repeated several times for each specimen studied. Of each pop-

ulation, 20–25 specimens for each species were collected and of these more than 15 permanent slides were prepared. The well-spread 10 metaphase plates about 10 permanent slides were photographed with an Olympus BH-2 camera and drawn from permanent slides deposited at the Department of Biology, Karadeniz Technical University, Trabzon.

#### **RESULTS**

Asterisks (\*) denote species for which chromosome numbers have not been counted prior to this report. Numbers in parentheses indicate numbers of cells with chromosome numbers as indicated (e.g., "66–75 (2)" means that 2 cells were observed to have 66–75 chromosomes).

# \*Alchemilla barbatiflora Juz.

This is an Euxine element that grows on steep slopes at 1 700–2 400 m. It has been reported from Trabzon (Zigana Dağı at 1 700 m and from N side of Soğanlı Dağı, above Çaykara at 2 000–2 200 m) by Pawlowski and Walters (1972).

2n = 109–118 (Fig. 1). Specimens: Rize, Ayder, Çamlı-hemşin, 1 450 m, 5.VII.1994, *S. Hayırlıoğlu-Ayaz 114*: 109 (1), 109–113 (4); Trabzon, Araklı, Dagbaşı, Kirazlı yay, 1 650 m, 9.VIII.1995, *S. Hayırlıoğlu-Ayaz 219*: 109–113 (1), 112–118 (3), 113 (1).

#### \*Alchemilla bornmuelleri Rothm.

This is an endemic and Iran-Turan element that grows on stream side banks at 1 200–1 800 m. It has not previously been recorded for northeast Anatolia.

2n = 64–75 (Fig. 2). Specimens: Trabzon, Zigana Pass (Tunnel), 1 800 m, 17.VII.1995, *S. Hayırlıoğlu-Ayaz 189*: 64 (6), 66–75 (2), 70–72 (4), 72 (2), 75 (1).

#### \*Alchemilla bursensis B. Pawl.

This is an endemic and Euxine element that grows in bogs under *Fagus* at 1 200–1 400 m. It has not

previously been recorded for northeast Anatolia.

2n = 96–102 (Fig. 3). Specimens: Trabzon, Sultanmurat yay, Çaykara, 1 700 m, 30.VI.1993, *S. Hayırlıoğlu-Ayaz 1*: 96 (4), 96–97 (2), 100–101 (2), 100–102 (2), 102 (2).

#### \*Alchemilla ciminensis B. Pawl.

An endemic and Iran-Turan element that grows in *Fagus* and *Picea* forests and by streams at 2 200–2 400 m. It has not previously been recorded for northeast Anatolia.

2n = 70–72 (Fig. 4). Specimens: Çoruh, Şavval Tepe above Murgul, 2 200 m, 16.VII.1993, *S. Hayırlıoğlu-Ayaz 12*: 70 (1), 70–72 (4), 72 (2), 72–76 (5).

#### \*Alchemilla erzincanensis B. Pawl.

An endemic and Iran-Turan element that grows along streams at 2 450 m. It has not previously been recorded for northeast Anatolia.

2n = 96–108 (Fig. 5). Specimens: Trabzon, Zigana Pass, 1 800 m, 19.VIII.1993, *S. Hayırlıoğlu-Ayaz 66*: 96 (2), 96–100 (3), 101–103 (1), 101–106 (2), 101–108 (3), 106 (2), 108 (1).

# \*Alchemilla hirsutiflora Rothm.

An endemic and Euxine element that grows on wet meadows and streamside banks at 1 800 m. It has not previously been recorded for northeast Anatolia.

2n = 90–102 (Fig. 6). Specimens: Trabzon, Çaykara, Akdoğan köyü, 1 800 m, 21.VIII.1993, *S. Hayırlıoğlu-Ayaz* 95: 90 (1), 92 (4), 92–102 (1), 96–100 (2), 100–102 (3).

# \*Alchemilla hirtipedicellata B. Pawl.

An Euxine element that grows on meadows of forest slopes and wet meadows at 1 100–1 350 m. It was reported from Trabzon (Fol Köy at 1 100 m) by Pawlowski and Walters (1972).

2n = 86–96 (Fig. 7). Specimens: Rize, Çayeli, Kaptanpaşa, Çataldere Köyü, 1 350 m, 27.VII.1996, *S. Hayırlıoğlu-Ayaz 308*: 86 (2), 93–96 (4), 96 (6).

#### Alchemilla mollis Rothm.

This species grows by streams in *Abies* and *Fagus* forests at 900–2 100 m. It has not previously been recorded for northeast Anatolia.

2n = 87–102 (Fig. 8). Specimens: Gümüşhane, Gümüşhane to Trabzon, 1 500 m, 15.VIII.1993, *S. Hayırlıoğlu-Ayaz 34*: 88–90 (2), 90–92 (2), 91 (2), 102 (2); Rize, Anzer, 2 200 m, 21.VIII.1995, *S. Hayırlıoğlu-Ayaz 212*: 87–88 (2), 90–92 (2), 102 (3). Wegener (1967) reported 2n = 102–106.

#### \*Alchemilla orduensis B. Pawl.

An endemic and Euxine element that grows on stony mountain slopes at 1 800–2 200 m. It has not previously been recorded for northeast Anatolia.

2n = 98–108 (Fig.9). Specimens: Trabzon, Zigana Dağı, on the road of Kadırga, 2 200 m, 11.VIII.1993, *S. Hayırlıo-ğlu-Ayaz* 22: 98 (3), 98–100 (4), 106 (1), 106–108 (2), 108 (2).

#### \*Alchemilla oriturcica Pawl.

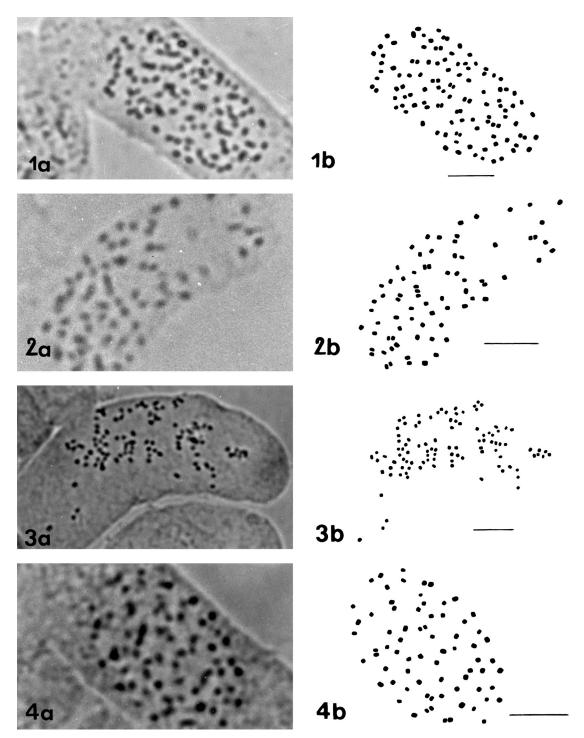
An endemic and Iran-Turan element that grows on steep rocky slopes at 2 000–2 400 m. It was reported from Trabzon (Soğanlı dağı, above Çaykara at 2 000–2 200 m) by Pawlowski and Walters (1972).

2n = 86–100 (Fig. 10). Specimens: Trabzon, Çaykara, Akdoğan Köyü, 1 400 m, 25.VIII.1993, *S. Hayırlıoğlu-Ayaz* 97: 86–90 (4), 90 (3), 104–106 (1), 106 (2).

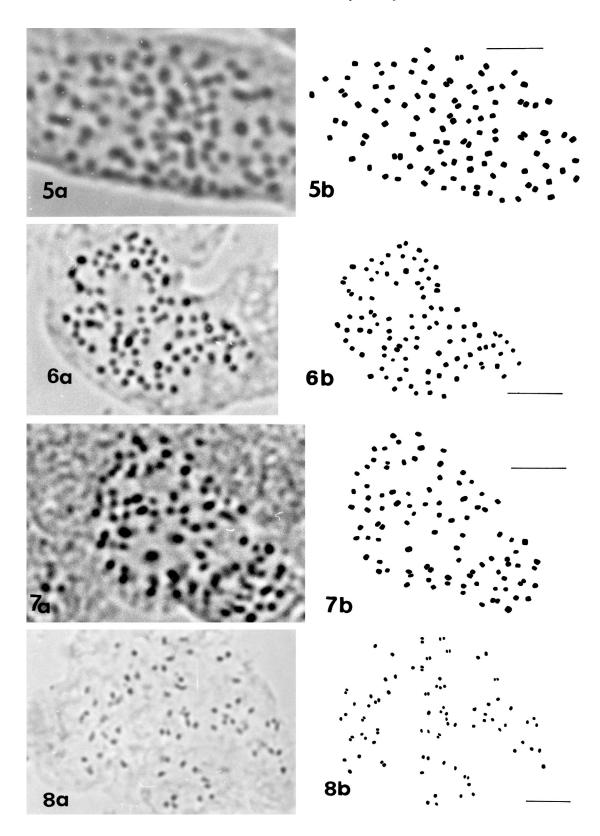
#### \*Alchemilla orthotricha Rothm.

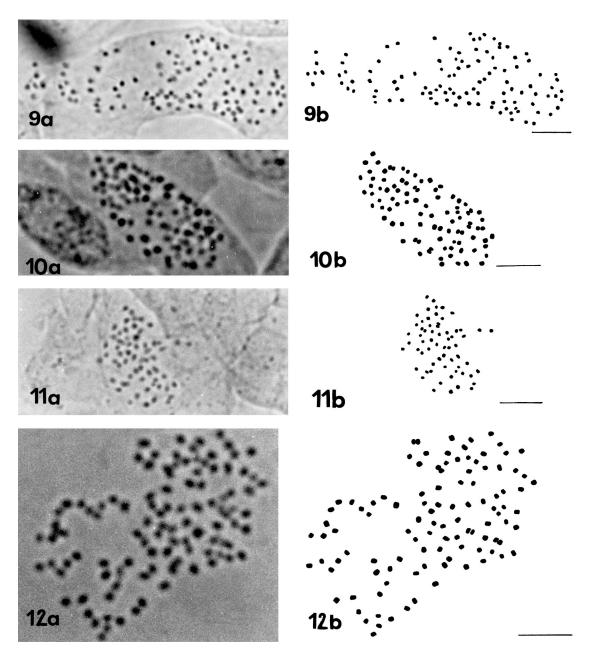
An Euxine element that grows on meadows and streamside banks at 1 700–1 800 m. It was reported from Gümüşhane (Karagöl Dağı) and Trabzon (Zigana Dağı above Hamsiköy) by Pawlowski and Walters (1972).

2n = 64 (Fig. 11). Specimens: Trabzon, Zigana Dağı, above Hamsiköy, 1 800 m, 10.VII.1994, *S. Hayırlıoğlu-Ayaz 80*: 64 (10).



**Figs. 1–8** (above and opposite). Mitotic metaphases in *Alchemilla*. The photographs (a) were taken with a BH 2 Olympus microscope. The drawings (b) were made with the aid of a drawing attachment connected to a BH 2 Olympus microscope. — 1: *A. barbatiflora*, 2n = 113. — 2: *A. bornmuelleri*, 2n = 75. — 3: *A. bursensis*, 2n = 102. — 4: *A. ciminensis*, 2n = 72. — 5: *A. erzincanensis*, 2n = 108. — 6: *A. hirsutiflora*, 2n = 100. — 7: *A. hirtipedicellata*, 2n = 96. — 8: *A. mollis*, 2n = 91. Scale bar = 10 μm.





**Figs. 9–12.** Mitotic metaphases in *Alchemilla*. The photographs (a) were taken with a BH 2 Olympus microscope. The drawings (b) were made with the aid of a drawing attachment connected to a BH 2 Olympus microscope. — 9: *A. orduensis*, 2n = 108. — 10: *A. oriturcica*, 2n = 90. — 11: A. orhotricha, 2n = 64. — 12: *A. oxysepala*, 2n = 107. Scale bar =  $10 \mu m$ .

# Alchemilla oxysepala Juz.

An Euxine element that grows on mountain meadows, forest and streamside banks at 1 600–1 800 m.

In the flora of Turkey, this species is doubtfully recorded in northeast Anatolia (Pawlowski & Walters 1972).

2n = 95-107 (Fig. 12). Specimens: Rize, Īkizdere to

Ispir, 1 600 m, 10.VII.1995, *S. Hayırlıoğlu-Ayaz 216*: 95 (2), 103–104 (3), 104–107 (1), 107 (4). Ehrenberg (1945) reported 2n = 100; Turesson (1957) reported 2n = 105–109.

# *Alchemilla persica* Rothm.

An Iran-Turan element that grows by streams at 1 850–2 800 m. It has not previously been recorded for Northeast Anatolia.

2n = 96–106 (Fig. 13). Specimens: Trabzon, Zigana Pass, 1 800 m, 20.VII.1993, *S. Hayırlıoğlu-Ayaz 15*: 96 (2), 96–104 (1), 104 (2); Trabzon, Araklı, Dağbaşı, Kirazlı yay., 1 650 m, 20.VII.1996, *S. Hayırlıoğlu-Ayaz 300*: 96–104 (2), 102 (1), 106 (4). Wegener (1967) reported 2n = 101–106.

#### \*Alchemilla sintenisii Rothm.

An endemic and Euxine element that grows in Alpine meadows at 1 800 m. It was recorded from Gümüşhane (Karagöl Dağı) by Pawlowski and Walters (1972).

2n = 94–108 (Fig. 14). Specimens: Trabzon, Zigana Tunnel and its around, 1 800 m, 7.VII.1995, *S. Hayırlıoğlu-Ayaz 199*: 94 (2), 94–96 (2), 94–98 (3), 106 (3).

#### \*Alchemilla stricta Rothm.

This may be a Caucasian element that grows on marshy ground by lakes and streams and in *Pinus* forest at 1 400–2 300 m. *Alchemilla undecimloba* Juz. may be identical with this species according to the flora of Turkey (Pawlowski & Walters 1972). It has not previously been recorded for northeast Anatolia.

2n = 66–100 (Fig. 15). Specimens: Rize, Cimil, Başköy, 2 300 m, 11.VIII.1995, *S. Hayırlıoğlu-Ayaz 302*: 82 (2), 82–86 (1), 90–98 (2), 100 (3); Trabzon, Çaykara, Sultanmurat yay., 2 100 m, 11.VII.1993, *S. Hayırlıoğlu-Ayaz 8*: 66 (3), 67–68 (2), 70–75 (2), 82 (2), 97–98 (1).

# \*Alchemilla tiryalensis B.Pawl.

An endemic and Euxine element that grows on rocky igneous slopes at 2 300–2 700 m. It has been reported from Çoruh Tiryal Dağı, above Murgul,

2 300 m and Şavval Tepe above Murgul, 2 700 m, by Pawlowski and Walters (1972).

2n = 97–123 (Fig. 16). Specimens: Çoruh, Tiryal Dağı above Murgul, 2 500 m, 4.VIII.1996, *S. Hayırlıoğlu-Ayaz 303*: 97 (3), 100 (1), 100–107 (2), 107 (2), 114–119 (1), 119–123 (3).

# \*Alchemilla ziganadagensis B. Pawl.

An endemic and Euxine element that grows in *Picea orientalis* forests and woodlands at 1 700–1 750 m. It was recorded from Trabzon (Zigana Dağı supra Hamsiköy) by Pawlowski and Walters (1972).

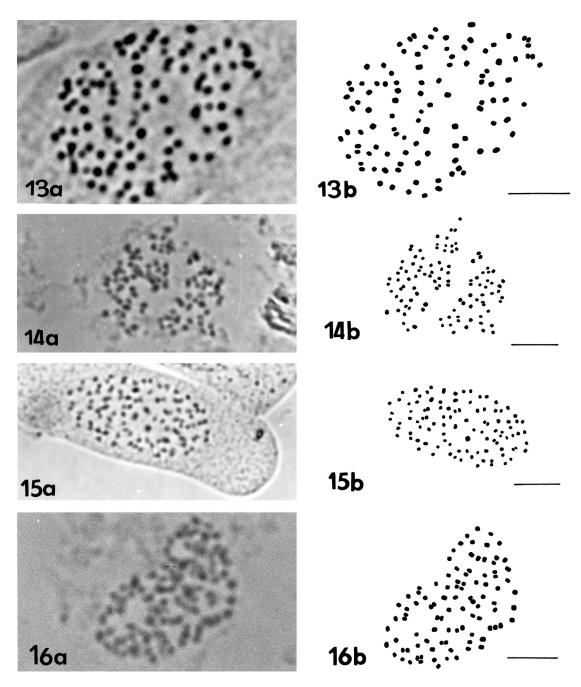
2n = 72 (Fig. 17). Specimens: Trabzon, Zigana Dağı, 1 700 m, 19.VII.1994, *S. Hayırlıoğlu-Ayaz 83*: 72 (11).

#### **DISCUSSION**

Former karyological studies of *Alchemilla* species belonging to sect. *Alchemilla* have revealed that they are represented by a series of very high polyploid cytotypes with the chromosome numbers ranging from 64 to ca. 224. About 75% of the species have chromosome numbers of 2n = 96-110 (Turesson 1957, Wegener 1967, Izmailow 1981).

Karyological data of ser. *Elatae* are still scarce. Some species were previously studied by Wegener (1967) from the Alps, Caucacus and a Botanical Garden in Austria, and by Ehrenberg (1945) and Turesson (1957) from the Alps. Wegener (1967) reported the chromosome numbers of *A. mollis* (2n = 102-106) and *A. persica* (2n = 101-106) of ser. *Elatae*. Ehrenberg (1945) was the first to report the chromosome number of *A. oxysepala* (2n = 100). Later, Turesson (1957) reported the chromosome number of *A. oxysepala* as 2n = 105-109.

Comparison of reports by Wegener (1967), Ehrenberg (1945) and Turesson (1957) with the results of the present study shows that specimens collected from the Alps and Caucasus and from northeast Anatolia have different chromosome numbers. The diverging karyological results might result from intraspecific karyological differentiation. Such a differentiation has been found in *Al*-

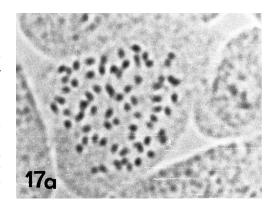


**Figs. 13–16.** Mitotic metaphases in *Alchemilla*. The photographs (a) were taken with a BH 2 Olympus microscope. The drawings (b) were made with the aid of a drawing attachment connected to a BH 2 Olympus microscope. — 13: *A. persica*, 2n = 104. — 14: *A. sintenisii*, 2n = 106. — 15: *A. stricta*, 2n = 100. — 16: *A. tiryalensis*, 2n = 97. Scale bar = 10  $\mu$ m.

chemilla alpina (2n = 137-144 in Scandinavia, 2n = 119-122 in the Alps according to Turesson 1958; 2n = 119-129 in the central Alps according

to Wegener 1967). Similarly, Izmailow (1982) reported that specimens collected from various geographical regions have different chromosome

Fig. 17. Mitotic metaphases in *Alchemilla ziganadagensis*, 2n=72. Scale bar =  $10 \mu m$ . The photograph (a) was taken with a BH 2 Olympus microscope. The drawing (b) was made with the aid of a drawing attachment connected to a BH 2 Olympus microscope.





numbers.

The investigations of Turesson (1957) and Wegener (1967) showed that that 75% of species of sect. Brevicaulon have 2n = 102-106. Information of genome, chromosome structure and karyogram are necessary in understanding the relationships of Alchemilla species. A taxonomic classification of certain species of Alchemilla based on their chromosome numbers results in 4 main groups (Wegener 1967). In the first group, the chromosome number is not more than 2n = 110, and in the second group it is 2n = 119-132. In the third and fourth groups, the chromosome numbers are higher than in the other two groups, which clearly makes a difference between two groups (joining 1 and 2, and 3 and 4). The species studied from northeast Anatolia belonging to the ser. Sericeae, Pubescentes and Elatae (in the present study) represent the first group.

Different series with the same chromosome numbers can occur in a single group. Thus, series are classified according to their morphological characters. Although the species are classified by a few certain morphological characters, a group may not be well distinguished morphologically. Alchemilla orthotricha is similar to A. ziganadagensis according to flora of Turkey (Pawlowski & Walters 1972). Both species are distinguished by a few morphological features. Our results showed that A. orthotricha and A. ziganadagensis have the chromosome numbers 2n = 64 and 2n =72, respectively. We suggest that these numbers can be used as additional data to support morphological separation of the species. Although the results of cytological studies have not been very important in the taxonomy of Alchemilla, cytological data can have taxonomic value, as this example shows.

In connection with diffuculties in an exact determination of chromosome numbers, the basic number as well as the degree of polyploidy in *Alchemilla* remain problems. Gentscheff and Gustafsson (1940), as well as Gudjonsson (1941), suggested x=7 is the basic number in *Rosoideae*. Later, Löve and Löve (1948) and Raven (1975) have suggested the basic number in *Alchemilla* is x=8. The occurrence of the same basic number in the related genus *Aphanes* as well as the chromosome number 2n=64 reported for some *Alchemilla* species are in favour of ther latter opinion. Nevertheless, knowledge of the cytology of *Alchemilla* is yet too insufficient to determine the basic number with certainty (Wegener 1967).

#### REFERENCES

Beyazoğlu, O., Hayırlıoğlu, S. & Ayaz, F. A. 1994: Karyotype analysis of *Aconitum orientale* and *Aconitum nasutum.* — *Turkish J. Bot.* 18: 493–495.

Buser, R. 1894: Zur Kenntnis der Schweizerischen Alchemillen. — Ber. Schweiz. Bot. Ges. 4: 41–80.

Ehrenberg, L. 1945: Kromosomtalen hos några kärlväxter.
— *Bot. Not.* 4: 430–437.

Gentscheft, G. & Gustafsson, A. 1940: Parthenogenesis and pseudogamy in *Potentilla*. — *Bot. Not.* 40: 109–132.

Gudjonsson, G. 1941: Om *Aphanes* L. og *A. microcarpa* (Boiss. et Reut.) Rothm. og deres udbredelse i Danmark. — *Bot. Tidskr.* 45: 352–370.

Hayırlıoğlu, S. & Beyazoğlu, O. 1997a: Chromosome numbers in species of *Alchemilla* L. belong to the series *Sericeae* Bus and *Pubescentes* Bus. (section *Alchemilla* Rothm.) in Turkey. — *Caryologia* 50: 77–84.

Hayırlıoğlu, S. & Beyazoğlu, O. 1997b: New chromosome

- numbers in *Alchemilla* L. (Rosaceae) from Turkey. *Willdenowia* 27: 191–194.
- Hayırlıoğlu, S. & Beyazoğlu, O. 1997c: The chromosome numbers of five new *Alchemilla L*. (Rosaceae) records for Turkey. — *Turkish J. Bot*. 21: 385–389.
- Izmailow, R. 1981: Karyological studies in species of Alchemilla L. from the series Calycinae Bus. (section Brevicaulon Rothm.). Acta Biol. Cracov. Ser. Bot. 23: 117–130.
- Izmailow, R. 1982: Further karyological studies in species of Alchemilla L. from the series Calycinae Bus. (section Brevicaulon Rothm.). — Acta Biol. Cracov. Ser. Bot. 24: 127–141.
- Juzepczuk, S.-W. 1941: Alchemilla L. In: Komarow, W. L. (ed.), Flora of the U.S.S.R 10: 216–301. Izdatel'stvo Akademii Nauk SSR, Moskva–Leningrad.
- Löve, A. & Löve, D. 1948: Chromosome numbers of northern plant species. University of Applied Sciences,
   Department of Agriculture, Reports, Reykjavik. 67 pp.
   Löve, A. & Löve, D. 1975: Cytotaxonomical atlas of the

- arctic flora. J. Cramer, Leutershausen, pp. 341–343.
   Pawlovski, B. 1972: De Alchemillis Turcicis subsectionibus
   Chirophyllum Rothm. et Calycanthum Rothm. sectionis
   Alchemilla. Fragm. Flor. Geobot. 18: 3–44.
- Pawlovski, B. & Walters, S. M. 1972: Alchemilla L. In: Davis, P. H. (ed.), Flora of Turkey and the East Aegean Islands 4: 80–105. Edinburgh Univ. Press, Edinburgh.
- Raven, P. H. 1975: The bases of angiosperm phylogeny: cytology. — Ann. Missouri Bot. Garden 62: 724–764.
- Rothmaler, W. 1944: Zur nomenklatur der Europäischen Alchemilla-Arten. — Svensk Bot. Tidskr. 38: 102–112.
- Turesson, G. 1957: Variation in the apomictic microspecies of *Alchemilla vulgaris* L. III. Geographical distribution and chromosome number. — *Bot. Not.* 110: 413–422.
- Walters, S. M. 1952: Alchemilla subcrenata Buser in Britain. — Watsonia 2: 277–278.
- Wegener, K. A. 1967: Chromosomenzahlen aus wurzelspitzen von *Alchemilla*-Arten (der sektionen *Pentaphyllon* Rothm. und *Brevicaulon* Rothm.). *Biol. Zentralbl.* 81: 771–792.