Chapter

Hermaphroditism in *Fragaria moschata*, a Cultivated Strawberry Species Preceding the Evolution and Growing of *F.* ×*ananassa* in Europe

Hannu Ahokas

Abstract

Two cases of hermaphroditism in *Fragaria moschata* Weston, (2n = 42), are described from Finland. One of them is a mutation in a clone known as a female since 1941 but appearing as a somatic segregant in 2013. The clone also carries a recessive gene for yellow-green leaf variegation. The mutations probably date back to the airborne radioactive fallout from the fire of the Chernobyl nuclear power plant in the Soviet Union in 1986. By interspecific crosses with the diploid (2n = 14) strawberry species, *F.* ×*bifera* Duch., *Fragaria vesca* L., and *Fragaria*. *viridis* Weston, the genetic determination of sex in *F. moschata* was studied and shown to depend on different genetic mechanisms in the two hermaphroditic musk strawberries. Selected tetraploid hybrids from crosses between musk and diploid strawberry species provide a wide variation of berry flavor and may have used as home garden strawberries. Two spontaneous hybrids between *F. moschata* and *F. vesca* are shown.

Keywords: Chernobyl 1986, edaphic requirements, green strawberry, musk strawberry, mutant manifestation, redomestication, wood strawberry

1. Introduction

The musk strawberry, *Fragaria moschata* is originally a Central European species having spread as an escape from gardens in many areas [1–5]. In the Tortona municipality in North Italy, cultivation of the musk strawberry, 'Profumata di Tortona' dates historically back to the year 1411 [6 cited in 7], when in a register of the *Pieve di Garbagna*, *magiostre* or *fravole* are indicated among the local fruits and vegetables [8]. The strawberry of Tortona is a local, dioecious ecotype, and the name 'Profumata' appeared for the first time in the mid-19th century [8]. The first known cultivar name of musk strawberry was 'Le Chapiron' in Europe in 1576; the cultivar name was derived further and apparently called 'Capron' by Jean de la Quintinie in 1672 [9]. In 1773, 14 varieties of the musk strawberry were listed in England by the botanist

Richard Weston [10]. The binominal name *Fragaria moschata* valid nowadays was ascribed to Weston [11]. In 1788, the Frenchman A. N. Duchesne described the hermaphroditic musk strawberry cultivar 'Le Caperonnier royal', the origin of which was known from Brussels in 1770 [12]. With the name 'Capiton' the variant was probably introduced to cultivation in Wallonia by about the middle of the 16th century [11]. A paper recognizing a hermaphroditic musk strawberry was red in England in 1817 by Keens [13] who, however, preferred dioecious varieties. Of the 'Le Caperonnier royal' variety was probably the hermaphroditic musk strawberries with name 'Capron royal' in St. Petersburg, Russia since 1869 [14, 15]. 'Capron' was studied by Ahmadi & Bringhurst [16] in USA in 1991. The use of the cultivar names of musk strawberries was confusing in the past [11, 17].

Hegi [1] considered the distribution of *F. moschata* in the north to reach Ingria (see [18, 19]), i.e., the territory on the eastern coast of the Gulf of Finland. The archaeobotanist Terttu Lempiäinen [20] thought *F. moschata* (erroneously under the name *F. muricata*) to have occurred in the 17th on Karelian Isthmus, in the territory of Finland of the Kingdom of Sweden. The growing or semicultural choosing of *F. moschata* preceded the evolution and spread of *F.* ×*ananassa* Duch., which occurred in Europe between the latter half of the 18th and early half of the 19th century [11, 21–23]. In Swedish texts of the Nordic literature, *F. moschata* was usually called *jordgubbe*, e.g., by Kalm [24] in 1759, which term later started to mean the garden strawberry, *F.* ×*ananassa* [25]. In Finland, the growing of *F.* ×*ananassa* began in the 1840s with a few early European cultivars [26]. Growing strawberries from seeds were prompted in the past [27], which may have resulted in the development of local varieties.

The earliest introductions to Finland of cultivated *F. moschata* occurred to gardens of manor houses in the 18th century principally from Sweden, less likely from Germany, and later on, from Russia. Stocks of musk strawberries were subsequently transferred from garden to garden. To the Valamo (Валаам, Valaam in Russian) islands on the Lake Ladoga, then of the territory of Finland, *F. moschata* varieties were introduced from Dr. E. Regel's garden nursery in St. Petersburg as written in 1870 by Damaskin [28], the igumene of the Valamo monastery. Regel" [29] recommended *F. moschata* varieties 'Belle Bordelaise', 'Bloch Hautbois', 'Capron royal', 'Capron framboisé' and 'Large flat' in 1866. He listed elsewhere altogether 11 varieties of *F. moschata* in 1869 [14]. *F. moschata* also thrived in the wilderness on Valamo, in a grove, where berry forming clones were found by Dr. Med. T. Sælan in 1881 [30] shown by Sælan's specimens H 371514 – H 371516, in Botancal Museum, Luomus, Helsinki. For berries, the growing of clones of both female and male, or a hermaphroditic clone are needed.

In the latter half of the 19th century, the supply of the garden nurseries in Europe turned to sell cultivars of *F.* ×*ananassa*. As examples, the catalog of Regel" & Kessel'ring" [15] in St. Petersburg, Russia in 1879 had 11 named cultivars of *F. vesca* s.l., 10 named cultivars of *F. moschata*, and 168 cultivars of *F.* ×*ananassa* (by far the most), with *F. virginiana* Miller and *F. chiloensis* (L.) Miller cultivars pooled together. The catalog of Vilmorin-Andrieux et C^{ie} [31] in Paris, France in 1891 listed eight named cultivars of *F. vesca* s.l., one of *F. viridis*, three of *F. moschata*, one of *F. virginiana*, one of *F. chiloensis* and 46 of *F.* ×*ananassa*.

In the now ceased Soviet Union before the year 1930, musk strawberry was grown for industrial purposes, though less than *F.* ×*ananassa*, at the most, on tens of hectares in different regions assessed then in the country recovering from its inner wars. There the cultivar names 'Španka' ('Шпанка'), 'Russkaya' ('Русская') and 'Kapron' ('Капрон') were recognized [32]. The 'Capron' cultivar usually meant a hermaphroditic variety. Soviet Union exported strawberry products [32], and their ingredients probably also contained musk strawberries.

The Austrian organization Arche Noah has maintained a hermaphroditic cultivar 'Oke' of *F. moschata* [33]. Among 108 *F. moschata* plants collected in Ziegenbusch, near Dresden, Germany, six were found to be hermaphroditic [5]. *F. moschata* is clearly a trioecious species also having hermaphroditic genotypes beside females and males.

Prof. Günter Staudt (*1926 †2008) did not believe that hermaphroditic *F. moschata* really exists [4, 12 p207]. He evidently held the concept of his teacher, Prof. Elisabeth Schiemann [34] that hermaphroditic *F. moschata*-like plants are tetraploid hybrids between *F. moschata* and *F. viridis*. Nowadays, the ancestry of the hexaploid *F. moschata* is thought to trace to three diploid species, *F. viridis*, *F. mandshurica* Staudt, and *F. vesca* [35] which are monoecious.

2. Find of two hermaphroditic *Fragaria moschata* clones in Finland

In the summer of 2012, I visited a private experimental farm in Loimaa town, SW Finland, where I to my surprise saw berry carrying plants of *F. moschata*, without any indication of separate females or males in the stand. The land owner's family had moved to the farm in 1954, when the strawberry was already there. The preceding history of the strawberry is unknown. I received a living sample of the musk strawberry, hereafter called 'Loimaa' (**Figures 1** and **2**), and deposited its herbarium specimen to Botanical Museum, Luomus (H 831495). Studying in the Museum specimens of *F. moschata* with berries, revealed a sample (H 223584) with well-formed anthers and pollen collected 13.5 km away from the experimental farm in Loimaa in 1971. This specimen and 'Loimaa' may be of the same gendre.

A surprise to a land owner in Kotka town, on the S coast of Finland, was the occurrence of berries in 2013 in an area of about 4 m^2 among about 3 ares of large strawberry plants, which had never set any berry during the observed period since

Figure 1.
The hermaphroditic 'Loimaa' of Fragaria moschata *at anthesis. June 5, 2013.*

Figure 2.
The hermaphroditic 'Loimaa' of Fragaria moschata *at maturity. June 19, 2014.*

1941. I got a message about the strange berry-set through conversations between two elderly people who met in a clinic waiting room, one being the landowner's relative, and the other my father. Visiting the farm in Kotka showed that an apparent mutation had occurred in the female clone of *F. moschata* to hermaphroditism (**Figures 3** and **4**).

Figure 3.
Occurrence of berries on a patch with flowers carrying fertile anthers in the female clone of Fragaria moschata *in Kotka, South Finland. Before the 2013 season, the musk strawberry occurrence has not made any berry since 1941, the year of the start of observation. The occurrence in the yard of the farm house has been isolated spatially from other clones of musk strawberry. July 4, 2013.*

Figure 4.
A dessicated late flower carrying fertile anthers in the anew risen hermaphrodite of Fragaria moschata *from the female clone in Kotka. The anthers have dehiscing stomia with pollen grains. See* ***Figure 3*** *for the stand already advanced to the berry maturing period on the day of the sampling, July 4, 2013.*

This mutated clone is called 'Kotka' hereafter. Its level of male fertility has appeared variable, being inferior in part of the flowers, or flowers opening at a certain point of the flowering season.

I performed crossing between 'Kotka' and 'Loimaa', and interspecific crosses of the hexaploid *F. moschata* with the diploid *F.* ×*bifera*, *F. vesca* and *F. viridis*, in order to reveal the inheritance of the sex-determining genotype in *F. moschata* in the resulting tetraploid hybrids and to study the possible use of the hybrids as cultural berries. *F.* ×*bifera* is the variably fertile hybrid between *F. vesca* and *F. viridis* [36]. Two spontaneous hybrids between *F. moschata* and *F. vesca* occurred.

The hexaploid chromosome number (2n = 42) in *F. moschata* in material from Finland has been determined in a female clone [37] and, in a male clone from Viitasaari, Central Finland (H 335838), by Ahokas (unpublished).

3. Crossing methods, seed germination and testing of self-pollination

I performed emasculation under magnification glasses in unopened flower-buds. The emasculated flowers were isolated in glassine crossing bags. Unopened flowers of the pollen parents were usually isolated in glassine crossing bags in advance to eliminated contaminating pollen. Such isolated flowers at the anthesis were directly used for pollination after the emasculated seed parent flowers were checked for the absence of anthers under magnification.

Cleaned mature seeds (achenes) were usually germinated on agar gel, where axenic conditions were tried to be attained. The seeds were surface sterilized with 0.001% $HgCl_2$ in water in tubes for 20 min, at times vortexed, rinsed with two changes of sterile water, and placed individually on sterile 1% gel of Bacto-agar (Difco, Detroit, USA) made in tap-water, with added 0.01% $CaCO_3$. After germination, the seedlings with one or two vegetative leaves were transferred to soil (**Figure 5**). The commercial organic growing soil Puutarhan Musta multa (Biolan, Eura, Finland) was usually used

Figure 5.
Axenic germination of hybrid seeds of Fragaria moschata *'Kotka'* × F. viridis *on water agar gel. The germinats are at a suitable size to be transferred to soil. Scale digits millimeters.*

for growing. The experimental cultivation was mostly done in Kirkkonummi Municipality, on the southern coast of Finland.

Inflorescences before the opening of the flower buds were isolated in glassine crossing bags to test the self-pollination ability of the flowers. If the earliest flowers had already opened, they were removed before the isolation. Crossing bags were cut to a suitable length, closed around the inflorescence stem with a paper-clip, and were often needed to be tied with a thread to a bamboo-stick for support. The opened flowers were also scored for the sex organs once or twice during the flowering period.

4. Cross between the two hermaphrodites of *Fragaria moschata*

The F_1 progeny of the cross of the hermaphrodites, *F. moschata* 'Kotka' × 'Loimaa' comprised 21 females, 10 hermaphrodites and 37 males. Pooling females and hermaphrodites together, the ratio 31: 37 would fit to a 1: 1 segregation, (χ^2 = 0.529, df = 1, p = 0.467). The offspring of the reciprocal cross has not been studied. The genetic bases of the hermaphroditism in 'Kotka' and 'Loimaa' are different.

Three berries around the 'Kotka' hermaphrodite were taken in 2015. These were evidently from female flowers open-pollinated by the near-by hermaphrodite, and as appeared later, one by *F. vesca*. The offspring of the three berries were much alike. Along with typical musk strawberries, two of the berry-series produced also variegating, morphologically distinct and weak seedlings (**Table 1**, **Figure 6**) and one exceptional seedling with 2n = 28 chromosomes, the *F. vesca* hybrid (see next sections).

5. Two spontaneous tetraploid hybrids, *Fragaria moschata* × *F. vesca*

Berries of the tetraploid strawberry found in Jokioinen, SW Finland [38] have the fragrance of *F. moschata* or *F. viridis*. At the time I found the tetraploid strawberry in

Berry no.	Females	Males	Sex unknown	Variegated leaves	Hybrid with *F. vesca*
1/3	14	13	8	3*	1
2/3	5	5	1	0	0
3/3	3	5	5	2	0
Total	22	23	14	5**	1

**One with flowers could be ranked to be female, all the other plants with variegated leaves remained unknown for the flowers.*
***The gene for variegation must be recessive, but the viability of the homozygotes is reduced and the number is less than expected, or 1/4.*

Table 1.
F_1 progenies from three berries taken from the Fragaria moschata *of the Kotka population. No hermaphrodite was observed. Before discarding, the F_1 plants were grown for two seasons during which the flowers were ranked using isolation of florescence and observing the anther development grade.*

Figure 6.
A variegating chlorotic, deformed and weak descendent grown from the seeds of an open-pollinated flower of the Kotka occurrence of Fragaria moschata *(**Table 1**). The occurrence of such mutant seedlings suggests inbreeding of heterozygosity of the allele or instability of the gene. The fact that there appeared significantly less than one forth (1: 3) of the mutants suggests them to have been deleterious with reduced viability in any stages between zygote and seedling. June 3, 2019.*

Jokioinen, explanations for its origin were open, but as the first obvious reason was the hybrid of *F. moschata* and *F. vesca*. Additional information has confirmed it to be an obvious hybrid between *F. moschata* and *F. vesca* along with the knowledge published from the genomic constitution of *F. moschata* since that. Two of its tetraploid seedling derivatives [39] or further spontaneous seedlings form the present sample of the Jokioinen hybrid (**Figures 7** and **8**). The original habitat in Jokioinen has been destroyed.

The other hybrid with *F. vesca* originates from open-pollinated female clone of *F. moschata* in Kotka. It has 28 chromosomes. Its phenotype is stout. The central leaflet overlaps partly the lateral leaflets. The leaflets are roundish. Apparent due to meiotic

Figure 7.
Under open-pollination nearly completely fertile, tetraploid hybrid of Fragaria moschata × F. vesca *originating from Jokioinen, Finland [38]. Evidently seedling of the original clone. July 5, 2021.*

irregularities it has low partial fertility of female organs under open-pollination, where there are various tetraploid hybrids around (**Figure 9**). The Kotka and Jokioinen hybrids crossed with difficulty resulting in an F_1 plant distinct in morphology from the parents (**Figure 10**).

6. Artificial hybrids, *Fragaria moschata* × *F. vesca*

An artificial hybrid of the hermaphroditic *F. moschata* 'Loimaa' × *F. vesca*. In advance isolated flowers of the *F. vesca* pollen parfent were of the spontaneous local Käpykallio clone in Kirkkonummi. *F. vesca* of Käpykallio is self-fertile, pollinating itself in isolated inflorescence. The cross gave in F_1 progeny the segregation of the hermaphroditic to male ratio of 6: 7 which is the best fit to a 1: 1 segregation among 13 examples (**Table 2**). The hermaphroditic flowers often showed partial fertility when selfed in isolation bags, which can be ascribed to meiotic irregularities of the interspecific hybrid.

7. Artificial hybrids, *Fragaria moschata* × *F. viridis*

Hybrids of *F. moschata* as the seed parent with *F. viridis* were fairly easily obtained, which I first tried with the clone of musk strawberries around the Jokioinen Manor,

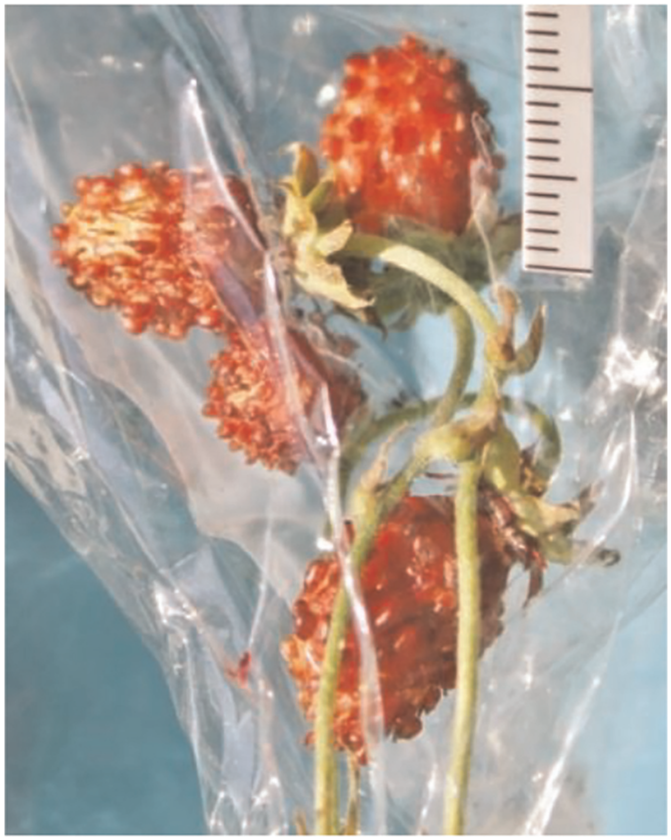

Figure 8.
Under isolation, a nearly perfect set of berries in the tetraploid hybrid of Fragaria moschata × F. vesca *originating from Jokioinen, Finland [38]. Evidently from a seedling of the original clone. The achenes are superficial and larger than those of* F. vesca*. The berries have a mild fragrance resembling that of* F. viridis *and* F. moschata*. Scale digits millimeters. July 19, 2020.*

Figure 9.
Under open-pollination, slightly fertile, tetraploid, spontaneous F_1 hybrid of Fragaria moschata *(female in Kotka)* × F. vesca*. Note the roundish leaflets with the margins of the central leaflet overlapping the laterals and the stout structure. Senescence of leaves has started along with the set of berries. July 13, 2021.*

Figure 10.
A plant of the first generation between the spontaneous hybrid of Fragaria moschata × F. vesca *from Kotka and Jokioinen. The leaf morphology does not resemble those of the parents (cf.* ***Figures 7*** *and* ***9****). Only slightly fertile under open-pollination. June 25, 2021.*

Seed parent (*F. moschata*)	Number of flowering F_1 phenotypes		
	Females	Hermaphrodites	Males
'Loimaa', hermaphrodite	0	6	7

Table 2.
F_1 progeny of the crosses of the hermaphroditic Fragaria moschata *'Loimaa' with the hermaphroditic pollen parent* F. vesca*.*

where this species had evidently been introduced in the 18th century. I obtained two plants from a single pollinated flower with a clone of *F. viridis* obtained from the native occurrence in Lemland, Alandia, Finland (Dr. Carl-Adam Hæggström's collection no. 7451, H 258115). A female and male F_1 plant resulted and proved to be interfertile (**Figure 11**). An octoploid derivative of it was nearly sterile (**Figure 12**). The genetic determining hermaphroditism in 'Loimaa' and 'Kotka' appear different with the F_1 segregation numbers (**Table 3**). The F_1 fertility with the 'Kotka' hermaphrodite was in general good both as selfed and open-pollinated (**Figures 13** and **14**). While *F. viridis* has monopodial branching pattern of the stolon, the *F. moschata* × *F. viridis* hybrids have sympodial stolon as also the *F. moschata* parent species.

The phenotype distributions of the crosses with the hermaphrodites 'Loimaa' and 'Kotka' are highly significantly different, $\chi^2 = 57.008$, df = 2, $p < 0.001$. In the 'Kotka' pedigree, the pooled female + hermaphrodite, 13 ratio to 6 males does not differ from the 67: 26 ratio of the 'Loimaa' pedigree, $\chi^2 = 0.0016$, df = 1, p = 0.968.

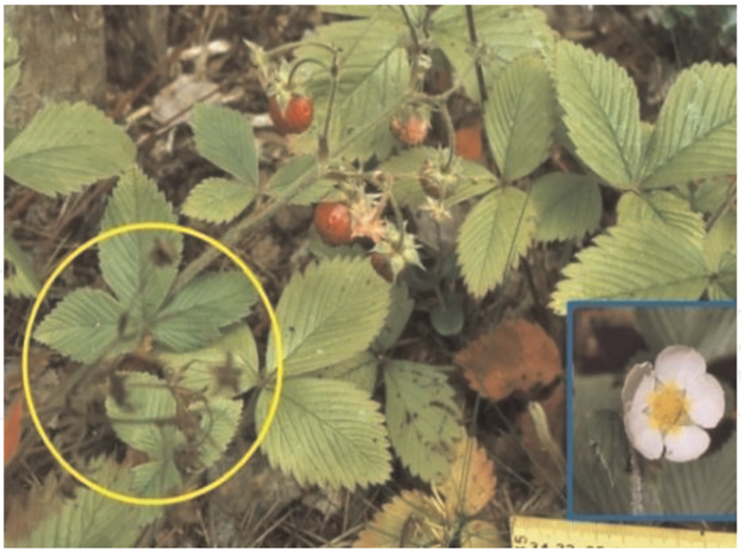

Figure 11.
The F_1 male and female plants of an artificial hybridization of Fragaria moschata *(female)* × F. viridis*. The whitered male flowers (yellow circle) have pollinated those of the female. August, 1998. The blue-framed insert shows a female flower of the hybrid on June 30, 1997. The female and male offspring of the hybrid with the hermaphroditic* F. viridis *as the pollen parent corroborate the female heterogamety in* F. moschata*.*

Figure 12.
An octoploid derivative of the tetraploid F_1 female plants of the artificial hybrid of Fragaria moschata × F. viridis*. Open-pollinated. Practically also female sterile. The polyploidy causes more roundish leaflets (cf.* ***Figure 11****) and leaves with five leaflets are frequent. The polyploidy was induced with the method described [40]. Scale 1 × 18 cm. July 19, 2021.*

Seed parent (*F. moschata*), and its flower type	Number of flowering F_1 phenotypes		
	Females	**Hermaphrodites**	**Males**
Female from Jokioinen	1	0	1
'Loimaa', hermaphrodite	0	67	26
'Kotka', hermaphrodite	10	3	6

Table 3.
F_1 progenies of the crosses of the female clone from Jokioinen, Finland and the hermaphroditic Fragaria moschata *'Loimaa' and 'Kotka' with the hermaphroditic pollen parent* F. viridis*.*

Figure 13.
Fertile, female F_1 hybrid plant of Fragaria moschata *'Kotka'* × F. viridis *forming plenty of berries by open-pollination. Under the isolation-bag the antherless female flowers did not set any berries. Scale 1 × 18 cm. July 7, 2021.*

Figure 14.
Fertile, hermaphroditic F_1 hybrid of Fragaria moschata *'Kotka'* × F. viridis *forming berries under isolation-bag. July 16, 2021.*

8. Artificial hybrids, *Fragaria moschata* × *F.* ×*bifera*

The hybrid species *F.* ×*bifera* (syn. *F. hagenbachiana*) is a morphologically variable, intermediate of its parental species, and variably fertile (**Figure 15**). The hybrid species may also include backcross derivatives with the parental species. Principally, the F_1 hybrids from the cross *F. moschata* × *F.* ×*bifera* in my experiments resembled

Figure 15.
A fairly fertile Fragaria ×bifera *(interspecific hybrid of* F. vesca *and* F. viridis*) was used as the pollen parents in crosses with* F. moschata*. The loose berries fell accidentally from the plant, while removing the metal network (mesh 12 mm) sheltering from local white tail deers, roes and moles. July 13, 2019.*

those of the *F. moschata* × *F. viridis* hybrids. The berry flavor of the hybrids is usually strong having taste of both the parents and varies from sweet to sour. The fragrance and sweetness of the delicate berries (**Figure 16**) also attract flying insects, a disadvantage in free-land growing. In *F. moschata* and its hybrids the senescence of leaves begins after flowering (**Figures 17** and **18**). Only hermaphrodites and males were obtained in F_1 (**Table 4**).

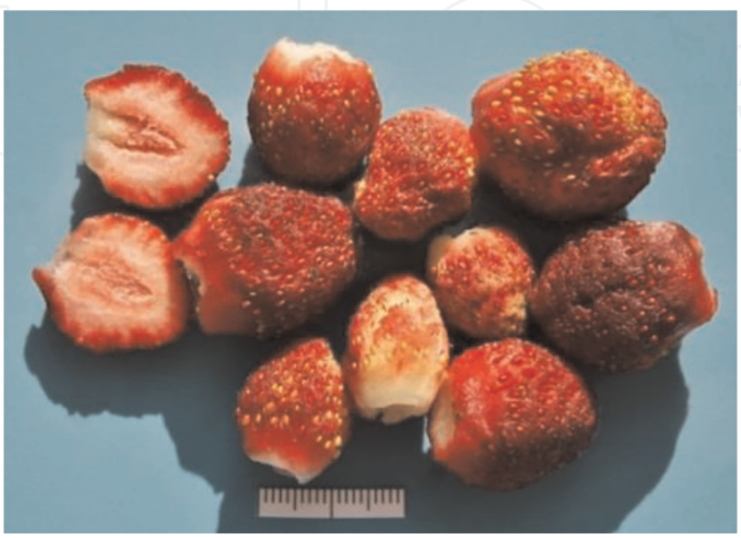

Figure 16.
Sweet and fragrant berries set with self-pollination under the tight isolation bag in the F_1 hybrid of Fragaria moschata × F. ×bifera*. Good fertility is revealed by the high seed set. Note the mode of the detachment and pinkish inner of the berry. Total fresh mass from the single inflorescence 33.5 g. Scale digits millimeters. June 19, 2020.*

Figure 17.
An F_1 hybrid of Fragaria moschata *('Loimaa')* × F. ×bifera *at anthesis. The leaves stay dark-green up to flowering. June 16, 2021.*

Figure 18.
The F_1 hybrid of Fragaria moschata × F. ×bifera *of* ***Figure 17*** *at maturity. The senescence of the leaves begins at the berry set. July 13, 2021.*

Seed parent (*F. moschata*)	Number of flowering F_1 phenotypes		
	Females	Hermaphrodites	Males
'Loimaa', hermaphrodite	0	31*	18

**Including three partially fertile inflorescences in isolation bags.*

Table 4.
F_1 progenies of the crosses of the hermaphroditic Fragaria moschata *'Loimaa' with the hermaphroditic pollen parent* F. ×bifera*.*

The 31: 18 ratio has a poor fit to 1: 1, χ^2 = 3.449, df = 1, p = 0.063. The 31: 18 ratio does not differ significant from the 67: 26 ratio of the *F. moschata* 'Loimaa' × *F. viridis* progeny (**Table 3**), χ^2 = 0.782, df = 1, p = 0.376.

9. Edaphic requirements of *Fragaria moschata* in natural habitat

The soil analysis of the habitat of *F. moschata*, from the site where the species has survived over 220 years in Hämeenkylä, Kouvola, SE Finland (**Figure 19**) shows a relatively high Ca content about the K and Mg contents (**Table 5**). The survival of *F. viridis* and the hybrid species *F.* ×*bifera* on their northern limit of distribution in southernmost Finland are highly dependent on a high Ca content of the soil in the habitat, where *F. vesca* is indifferent, but *F. vesca* also turns dependent on high Ca in its northern limit of distribution [42]. Another important issue is the high Ca: K ration of the soil in the habitats of *F. viridis*, while the *F.* ×*bifera* habitats are intermediate between those of *F. vesca* and *F. viridis* [42].

In garden strawberry (*F.* ×*ananassa*) a deficiency of Ca in emerging leaves causes leaf tip-burn [43]. A high availability of K in the soil may decrease Ca mobility in the strawberry plant, and result in tip-burn [44]. A further reason for tip-burn can be boron deficiency [45]. In the populations surviving in the wildness, I have not observed tip-burn disorder in *F. moschata* in the natural populations, but in cultivation for years in limited soil bags, tip-burn may occur. The incipient tip-burn in *F. moschata* may deform the leaflet shape from the typical rhomboid to that of *F.* ×*ananassa* (**Figure 20**).

Figure 19.
A female clone of Fragaria moschata *which has survived* in situ *over 220 years, see Munsterhjelm 1799–1801 [41].* F. moschata *is practically the only field layer species on the site in the highly shadowed deciduous grove in Hämeenkylä, Kouvola, Finland. Photography needed the use of flash on a sunny day. June 11, 2020.*

Soil type	Fine-sandy moraine Rich in organic
Conductivity	0.7 mS/cm
pH	5.4
Ca	1100 mg/l
P	8.8 mg/l
K	160 mg/l
Mg	170 mg/l
S	11 mg/l
B	0.6 mg/l
Cu	2.9 mg/l
Mn	120 mg/l
Zn	16 mg/l
Cation exchange capacity	13 cmol/kg
Ca/cat. exch. cap.	42%
K/cat. exch. cap.	3%
Mg/cat. exch. cap.	11%
Na/cat. exch. cap.	2%

Table 5.
Results of the topsoil analysis of the Hämeenkylä site in Kouvola, Finland, where F. moschata *has survived over 220 years, see Munsterhjelm 1799–1801 [41]. Sampling date Aug. 14, 2020. The analysis was purchased from Viljavuuspalvelu oy Eurofins (Mikkeli, Finland); analysis no. 20–00079100 named Mansikka-2.*

Figure 20.
Grown in limited soil bags, an incipient tip-burn disorder in leaflets of Fragaria moschata *with healthy leaves of the neighboring plant on the left. The affected leaves start to resemble those of a garden strawberry* F. ×ananassa*. A male descendent grown from the seeds of an open pollinated flower of the Kotka occurrence (**Table 1**). The seed parent plant must have been a female clone pollinated by the 'Kotka' hermaphroditic mutant. June 13, 2020.*

The foliage of *F. moschata* and its hybrids start to senesce when the berries grow. These plants have a tendency in the autumn to make over-wintering, dark-green leaves characterized with short petioles about half those of the summer leaves.

10. Discussion

Ahmadi and Bringhurst [16] reported having received only hermaphroditic descendants (24 plants) from the selfing of the hermaphroditic cultivar 'Capron' (*F. moschata*), while the cross between the female 'Perfumata' × hermaphroditic 'Capron' resulted in 12 females and 10 males and no hermaphroditic. This 1: 1 ratio was confirmed in the 9-ploid hybrids, 74 females and 72 males. The female *F. moschata* (Jokioinen) × hermaphroditic *F. viridis* cross (**Table 3**) also fits the heterogametic nature of female plants.

The hermaphroditism seems to lower yield in octoploid strawberry species compared with dioecious female clones [46, 47]. With three endemics American octoploid strawberries, also the sex of the plant was found to significantly affect the microbiomes on the flowers [48]. In environments, where both female and male clones of *F. moschata* are frequent enough for cross pollination, the fitness of female clones may be higher than that of hermaphrodites. In the margins of the sparse distribution of *F. moschata* with only one sex attainable on the place, this species relays on vegetative spread. The vegetative spread has also been promoted by human activities. The hermaphroditic *F. moschata* (syn. *F. elatior*) variety 'La Grange' could have sterile flowers from 7 to 90%, with an average of 52% of fertile flowers as written by Longworth in 1846 [49 cited in 46]. The recently arisen hermaphroditic mutant 'Kotka' of musk strawberry resembles the properties of 'La Grange' having variably perfect male fertility.

In most octoploid *Fragaria* taxa, a 13 kb sequence that occurred in all females and never in males was called sex-determining region (SDR). The SDR cassettes revealed a history of repeated translocation [50]. This also raises the hypothesis that the sex mutation from female to hermaphroditic ('Kotka') could be caused by a transposition. The probability for an occurrence of a transposon in the Kotka female clone is high, because transposable element sequences make up about 36% of the total genome assembly in the octoploid strawberry [51], and variation in the location of the sex-determining genes within the homoeologous chromosome group VI was shown in the North-American octoploid *F. virginiana* [52].

Thinly variegating leaves (**Figure 6**) occurred in five descendants of the open-pollinated *F. moschata* in Kotka, where the female clone was evidently back-crossed by its male-fertile hermaphroditic mutant. The variegation can be an expression of an unstable gene and could be caused by the movement of a transposing element, which may have been activated by an environmental mutagen. Already in 1955, Darrow [53] concluded that "The evidence is that variegation is due to a frequently mutating or unstable gene and that the tendency to mutate is inherited as recessive" and "A similar variegation has been obtained when seeds have been irradiated". The Kotka site was subjected to high doses of radioactive airborne fallout from the fire of the Chernobyl nuclear power plant in the Soviet Union in April–May of 1986. In Kotka, on June 6, 1986, a hot or high-activity particle apparently caused dysfunction in a monitor of radioactivity [54]. About five months after the beginning of the Chernobyl fallout, on October 1, 1986, in the Kotka area, the external dose rate was up to 0.159 μSv/h and the estimated ^{137}Cs surface activity up to 45 kBq/m^2 [55]. The mutating rate was high in parts of Finland after airborne fallout radioactivity from Chernobyl since the 1986 April, because one could make autoradiograms on Kodak X-OMAT AR films with fresh grass [54], and because of the occurrences of the hot particles [56]. On some sites, tens of such hot particles occurred on a 1-dm^2 area of land [54]. In a highly radioactively polluted area 32 km NW of the site of the 'Kotka' strawberry [55], three mutants occurred in less than 200 wheat plants grown for other research purposes in the field soil in the 1986 season, and were detected in subsequent seasons and generations [57]. On a tested site, radioactivity dose rate (mR/h) was 50 higher on the soil surfaces than at the 1-m level still on May 28, 1986 [54].

The 1986 radioactive airborne fallout from Chernobyl may have induced genetic changes in vegetative, on land surface lying strawberry stolon tissue, later somatically segregating into the phenotype like the hermaphroditism or, after inbreeding, occurring in seedlings like the variegation. Somatic segregation of polyploidy from stolon was shown in the polyploidization technique derived for strawberries ([40], figure 1d). There under the action of colchicine on cell division, polyploidized cells and cell-groups are numerous. Instead, a radiation-induced mutation on a given gene per cell is extremely rare, and it takes years that a viable mutated cell-line forms a mutant-carrying strawberry stolon, if ever. A recessive mutant needs a meiotic division to manifest, like the variegation gene (**Figure 6**).

The hybrids between *F. moschata* and *F. ×bifera*, *F. vesca* or *F. viridis* provide a wide variety of berry flavors and sweetness. They could be added as flavors mixed to other berries and berry products. The musk strawberries 'Capron Royal' and 'Profumata di Tortona' were among the highest aromatic 17 cultivars, 15 of which were modern *F. ×ananassa* cultivars [58]. Studying the aroma of two sources of musk strawberries, Ferreira et al. [59] detected more than 100 distinctive volatile compounds by GC–MS.

During some seasons, berries of musk strawberry may be infected with the fungus *Spherotheca macularis* f. sp. *fragariae*. Environmental factors like drought, high proportion of sunshine and amount of UV may control the occurrence of the infection. There may be hope to recombine resistance towards the fungus from *F. viridis* or *F. vesca* strains.

F. viridis is self-incompatible [60] and needs another genotype of incompatibility to pollinate. The self-incompatibility apparently increases chance of *F. viridis* to be pollinated with *F. vesca* where *F. viridis* is marginal and rare. The self-incompatibility is not inherited to the *F. moschata* × *F. viridis* hybrids. The self-incompatibility also gives up in the autotetraploid form of *F. viridis* studied with the polyploidization technique [40] which permits to test the single genotype on diploid and tetraploid level (unpublished).

11. Hybrids referred to as *Fragaria ×neglecta* and *F. ×intermedia*

The *F. neglecta* (*F. ×neglecta* Lindem.) hybrids were reported from Austria in 1896 [61]. *F. neglecta* was, however, described as a variant of *F. viridis* in 1865 [1, 62]. The dioecious *F. moschata* female pollinated with the hermaphroditic *F. viridis* gave tetraploid female and male descendants in a 1: 1 ratio [63]. Bors & Sullivan [64] obtained mostly tetraploids, some hexaploids and one mixoploid (4x/6x) among the offspring from the crosses between *F. moschata* and *F. viridis*. Their tetraploid hybrids were evidently fertile.

In 1841 v. Bach [65] described *β. intermedia* as a variant of *F. moschata* (syn. *F. elatior*). *F.* ×*intermedia* (Bach) Beck has later been considered as the hybrid between *F. moschata* and *F. vesca* [1, 61]. *F.* ×*intermedia* hybrids were collected by V. Feráková and their tetraploidy determined by A. Murin in Slovakia [66]. The unexpected chromosome numbers published for *F. moschata* by Lippert [67] might have been counted from interspecific hybrids with the diploid strawberries and their derivatives, though the plants were morphologically of the *F. moschata* type. Odd polyploidy levels, penta- and heptaploidy have been detected in spontaneous *F. moschata* × *F. vesca* hybrids in Europe [68, 69]. Evolutionary trials are ongoing with the trioecious *F. moschata*.

Author details

Hannu Ahokas
Helsinki, Finland

*Address all correspondence to: hannu.ahokas0@saunalahti.fi

References

[1] Hegi G. *Fragária* L. Erdbeere. Illustrierte Flora von Mittel-Europa. 1923;**4**(2):895-907

[2] Lee DV. Early history of the strawberry. In: Darrow GM, editor. The Strawberry. New York, Chicago, San Francisco: Holt, Rinehart, Winston; 1966. pp. 15-23. Also available from http://specialcollections.nal.usda.gov//speccoll/collectionsguide/darrow/Darrow_TheStrawberry.pdf

[3] Hultén E, Fries M. Atlas of North European Vascular Plants. Vol II-Vol III. Königstein: Koeltz Scientific Books; 1986

[4] Staudt G. Strawberry biogeography, genetics and systematics. Acta Horticulturae. 2009;**842**:71-83

[5] Lesemann SS, Bölke N, Tejeda Fernández CE, Buschmann S, Gerischer U, Neinhuis C, et al. Fruit diversity in a natural European habitat of *Fragaria moschata* Weston. Acta Horticulturae. 2017;**1156**: 215-222

[6] Bergaglio A. Il porto della fragole. Amministrazione Comunade di Tortona. 2007

[7] Negri AS, Allegra D, Simoni L, Rusconi F, Tonelli C, Espen L, et al. Comparative analysis of fruit aroma patterns in the domesticated wild strawberries "Profumata di Tortona" (*F. moschata*) and "Regina delle Valli" (*F. vesca*). Frontiers in Plant Science. 2015;**6**(56):1. fpls-06-00056.pdf

[8] Fragola Profumata di Tortona. L'Azienda Agricola Biologica "La Carcassola". https://www.carcassola.it/fragola_profumata_di_tortona.html [accessed on September 24, 2021]

[9] Reich L, Arlein VH. Uncommon Fruits for every Garden. 2nd ed. Portland, London: Timber Press; 2008

[10] Weston R. Tracts on Practical Agriculture and Gardening. 2nd ed. London: S. Hooper; 1773. https://books.google.com

[11] Wilhelm S, Sagen JE. A History of the Strawberry from Ancient Gardens to Modern Markets. Berkeley: University of California, Division of Agricultural Sciences; 1974

[12] Staudt G, Duchesne AN, Les dessins d AN. Duchesne pour son Histoire naturelle des fraisiers. A. N. Duchesne's drawings for his Histoire naturelle des fraisiers. Paris: Publications Scientifiques du Museum; 2003

[13] Keens M. On the cultivation of strawberries, in the open ground. Transactions of the Horticultural Society of London. 1818;**2**:392-397

[14] Regel E. Versuche über die Erdbeersorten im petersburger Klima. Gartenflora. 1869;**18**:172-179

[15] Регелъ ЭЛ, Кессельрингъ Я. 1880 пятнадцатый списокъ сортамъ плодовыхъ, древесныхъ, кустарныхъ, и многолътнихъ растенiй. Catalog von Obstsorten, Ziersträuchern und Stauden des Pomologischen Gartens und der Baumschulen. S.-Peterburg'; 1879

[16] Ahmadi H, Bringhurst RS. Genetics of sex expression in *Fragaria* species. American Journal of Botany. 1991;**78**: 504-514

[17] Goeschke F. Das Buch der Erdbeeren. 2nd ed. Berlin: Paul Parey; 1888

[18] Ruprecht FJ. Flora Ingrica. Vol. 1. Petropol, Riga, Lipsia: Eggers et soc., Sam. Schmidt, Leopold Voss; 1860

[19] Meinshausen KF. Flora Ingrica. St. Petersburg; 1878

[20] Lempiäinen T. Notes on the plants of Johannes municipality in the Karelian isthmus (Russia). Iskos. 2008; **16**:212-214

[21] Solms-Laubach Hz. Ueber unsere Erdbeeren und ihre Geschichte. Botanische Zeitung. 1907;**65**(1 Abt):46-76

[22] Staudt G. Die Entstehung und Geschichte der groβfrüchtigen Gartenerdbeeren *Fragaria ×ananassa* Duch. Der Züchter. 1962;**31**:212-218

[23] Lee DV. The strawberry from Chile. Duchesne and his work. Early breeding in Europe. In: Darrow GM, editor. The Strawberry. New York, Chicago, San Francisco: Holt, Rinehart, Winston; 1966. pp. 24-84. Also available from: http://specialcollections.nal.usda.gov//speccoll/collectionsguide/darrow/Darrow_The_Strawberry.pdf

[24] Kalm P. Kortt berättelse om de publique planteringar jag skiött och hwad wid de samma blifuit tilgiort, sedan till riksens höglofl. ständers manufactur-contoir min sidsta ödmiuka berättelse blef ingifwen. In: Leinberg KG, editor. Bidrag till kännedomen af vårt land. Vol. 4. Jyväskylä: J. Länkeläs förlag; 1888. pp. 73-90

[25] Hylander N. Linné, Duchesne och smultronen. Linnæus, Duchesne and strawberries. Svenska Linné-Sällskapets Årsskrift. 1945;**28**:17-40 – 95-97

[26] Anonymous. Om Huwitus. Åbo Tidningar. 1850;**73**(72):1-3

[27] Heikel BW. Hedelmä- ja marjaviljelyn opas. Helsinki: Otava; 1908

[28] Дамаскин. Садовство и огородничество на о. Валаамъ. Вѣстникъ Императорскаго Россійкаго Общества Садоводства. 1870;**10**:224-234

[29] Регелъ Э. Земляника. Вѣстникъ Императорскаго Россійкаго Общества Садоводства. 1866;**6**:104-126

[30] Hjelt H. Conspectus florae fennicae. Acta Societatis pro Fauna et Flora Fennica. 1915-1919;**41**(1):1-502

[31] Vilmorin-Andrieux et Cie. Les plantes potagères. 2nd ed. Paris: Vilmorin-Andrieux et Cie marchands grainiers; 1891

[32] Шарина Н. Промышленная культура земляники и клубники в СССР. Bulletin of Applied Botany, of Genetics and Plant-Breeding. 1930-1931;**25**(5):197-229

[33] Wachsmuth B. Von Monats-, Wald- und Moschus-Erdbeeren. Gartenpraxis. 2009;**35**(4):20-28

[34] Schiemann E. Artkreuzungen bei *Fragaria*. II. Zeitschrift für Induktive Abstammungs- und Vererbungslehre. 1937;**73**:375-390

[35] Kamneva OK, Syring J, Liston A, Rosenberg NA. Evaluating allopolyploid origins in strawberries (*Fragaria*) using haplotypes generated from target capture sequencing. BMC Evolutionary Biology. 2017;**17**(180):1-19 additional files

[36] Staudt G, DiMeglio LM, Davis TM, Gerstberger P. *Fragaria ×bifera* Duch.: Origin and taxonomy. Botanische Jahrbücher für Systematik, Pflanzengeschichte und Pflanzengeographie. 2003;**125**:53-72

[37] Evans WD. Guelph SO1 synthetic octoploid strawberry breeding clone. HortScience. 1982;**17**:833-834

[38] Ahokas H. Spontaneous tetraploidy in strawberry (*Fragaria* sp., Rosaceae). Nordic Journal of Botany. 1999;**19**:227-234

[39] Antonius K, Ahokas H. Flow-cytometric determination of polyploidy level in spontaneous clones of strawberries. Hereditas. 1996;**124**:285

[40] Ahokas H. A method to induce polyploids from adult strawberry plants, *Fragaria* spp., avoiding tissue culture *in vitro*. Plant Breeding. 1998;**117**: 500-502

[41] Munsterhjelm JC. Jacobina Charlotta Munsterhjelms dagböcker 1799-1801. In: Lönnqvist B, editor. Helsinki (Helsingfors) Svenska litteratursällskapet i Finland: Folklivsstudier 7. 1970. pp. 1-149

[42] Ahokas H. Factors controlling the north-edge distribution of *Fragaria viridis* and its *F. vesca* hybrids in South Finland. Acta Horticulturae. 2002;**567**: 385-388

[43] Guttridge CG, Bradfield EG, Holder R. Dependence of calcium transport into strawberry leaves on positive pressure in the xylem. Annals of Botany. 1981;**48**:473-480

[44] Chiu TF, Bould C. Effects of calcium and potassium on ^{45}Ca mobility, growth and nutritional disorders of strawberry plants (*Fragaria* spp.). Journal of Horticultural Science. 1976;**51**:525-531

[45] Maas JL, editor. Compendium of Strawberry Diseases. 2nd ed. St. Paul: APS Press; 1987

[46] Darrow GM. The importance of sex in the strawberry. Journal of Heredity. 1925;**16**:193-204

[47] Stahler MM, Luby JJ, Ascher PD. Comparative yield of female and hermaphroditic *Fragaria virgniana* germplasm collected in Minnesota and Wisconsin. In: Dale A, Luby JJ, editors. The Strawberry into the 21st Century. Portland: Timber Press; 1991. pp. 104-105

[48] Wei N, Ashman T-L. The effects of host species and sexual dimorphism differ among root, leaf and flower microbiomes of wild strawberries *in situ*. Scientific Reports. 2018;**8**:5195. DOI: 10.1038/s41598-018-23518-9

[49] Longworth N. Report on the cultivation of the strawberry. Report of Transactions of the Cincinnati Horticultural Society. 1846;**4**:59-61

[50] Tennessen JA, Wei N, Straub SCK, Govindarajulu R, Liston A, Ashman T-L. Repeated translocation of a gene cassette drives sex-chromosome turnover in strawberries. PLoS Biology. 2018;**16**(8): 1-22, e2006062, supporting material. DOI: 10.1371/journal.pbio.2006062

[51] Edger PP, Poorten TJ, Van Buren R, Hardigan MA, Colle M, McKain MR, et al. Origin and evolution of the octoploid strawberry genome. Nature Genetics. 2019;**51**:541-547. www.nature.com/naturegenetics

[52] Wei N, Govindarajulu R, Tennessen JA, Liston A, Ashman T-L. Genetic mapping and phylogenetic analysis reveal interspecific variation in sex chromosomes of the Virginian strawberry. Journal of Heredity. 2017; **108**:731-739. DOI: 10.1093/jhered/esx077

[53] Darrow GM. Leaf variegation in strawberry - a review. Plant Disease Reporter. 1955;**39**:363-370

[54] Rytömaa T, Servomaa K, Toivonen H. Tšernobylin ydinvoimalaturmassa syntyneet kuumat hiukkaset: Mahdollinen vaara

terveydelle Suomessa. Chernobyl reactor accident and radioactive particles found in Finland. Duodecim. 1986;**102**: 1190-1196

[55] Arvela H, Blomqvist L, Lemmelä H, Savolainen AL, Sarkkula S. Environmental gamma radiation measurements in Finland and the influence of meteorological conditions after the Chernobyl accident in 1986. In: STUK-A65. Helsinki: Finnish Centre for Radiation and Nuclear Safety; 1987. pp. 1-40

[56] Toivonen H, Servomaa K, Rytömaa T. Aerosols from Chernobyl: Particle characteristics and health implications. In: Philipsborn H, Steinhäusler F, editors. Hot Particles from the Chernobyl Fallout. Vol. 16. Theuern: Schriftenreihe des Bergbau- und Industriemuseum; 1988. pp. 97-105

[57] Ahokas H. Vehnämutantteja syntymävuosi 1986, Tsernobyl-vuosi. Wheat mutants, generated in 1986, the year of Chernobyl. Luonnon Tutkija. 1989;**93**:26-27

[58] Urruty L, Giraudel J-L, Lek S, Roudeillac P, Montury M. Assessment of strawberry aroma through SPME/GC and ANN methods. Classification and discrimination of varieties. Journal of Agricultural and Food Chemistry. 2002; **50**:3129-3136

[59] Ferreira V, Pet'ka J, Leitner E, Parameswaran B. Musk strawberries: The flavour of a formerly famous fruit reassessed. Flavour and Fragrance Journal. 2012;**27**:273-279

[60] Staudt G. Zur Selbststerilität von *Fragaria viridis* Duch. Die Naturwissenschaften. 1952;**39**:572

[61] Halácsy Ev. Flora von Niederösterreich. Prag, Wien, Leipzig: F. Tempsky, G. Freytag; 1896

[62] Lindemann Ev. Ueber eine sehr verbreitete und bisher verkannte Erdbeereart, *Fragaria neglecta*. Bulletin de la Société Impériale des Naturalistes de Moscou. 1865;**38**:218-220

[63] Schiemann E. Über die Geschlechtsvererbung bei *Fragaria*. Berichte der Deutschen Botanischen Gesellschaft. 1938;**56**(5):161-162

[64] Bors RH, Sullivan JA. Interspecific hybridization of *Fragaria moschata* with two diploid species, *F. nubicola* and *F. viridis*. Euphytica. 2005; **143**:201-207. DOI: 10.1007/s10681-005-3496-9

[65] Bach Mv. Bemerkungen über einige der selteneren Pflanzen des Mittelrheines. Flora. 1841;**45**:715-720

[66] Májovský J. Index of chromosome numbers of Slovakian flora. Acta Facultatis rerum naturalium Universitatis Comenianae - Botanica. 1976;**25**:1-18

[67] Lippert W. Morphologische und zytologische Untersuchung an *Fragaria*, insbesondere an *Fragaria moschata* in Bayern. Botanische Jahrbücher für Systematik, Pflanzengeschichte und Pflanzengeographie. 1985;**107**:195-202

[68] Nosrati H, Price AH, Gerstberger P, Wilcock CC. Identification of a natural allopentaploid hybrid *Fragaria* (Rosaceae), new to Europe. New Journal of Botany. 2011;**1**:88-92

[69] Nosrati H, Price AH, Gerstberger P, Wilcock CC. The first report of an alloheptaploid from the genus *Fragaria* (Rosaceae). New Journal of Botany. 2013;**3**:205-209