MORPHOLOGICAL DIFFERENTIATION OF FLOWER BUDS IN OWN-ROOTED AND BUDDED APPLE TREES

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ABSTRACT
In the period from the year 2002 to the year 2005 the morphological differentiation of flower buds in adult own-rooted trees (produced by trench layering) and the ones budded on seedling rootstock in the apple cultivar Golden Resistant, was studied. The onset of morphological differentiation was observed in the third ten-day period of the month of June and the second ten-day period of July, with no significant differences between the own-rooted and budded plants. No differences were established both in the rate of initiation of the leaf-like appendages (bud scales, transitional leaves, true leaves and bracts) on the bud axes, and also with respect to the “critical number of appendages”. The “critical number of appendages” including bud scales, transitional leaves, true leaves and the first bract, was comparatively identical according to years (15-16 in number). The obtained information about the morphological differentiation of flower buds in the own-rooted apple trees can serve as a valuable reference point in clarifying the flower bud formation processes in the in vitro propagated apple plants which have been of particular interest lately.

Introduction
Flower bud formation in the apple occurs in a definite sequence: preliminary induction of flower bud formation, histological transformation and morphological differentiation in the course of which the bud apexes are transformed into flower appendages (2, 3). This transformation occurs only if, regarding structure, the vegetative until a given moment buds, have already been completed, meaning that the leaf-like appendages (bud scales, transitional leaves, true leaves and bracts) are initiated on the nodes of the central axis (2). The widening and slight swelling of the apex in which case it acquires a dome-like shape, is usually accepted as the first sign of the transition from the vegetative into the reproductive state of the buds (1, 5, 6, 7, 8, 9). The sum total of the number of appendages by the time of the appearance of this sign, considered to be the “critical number of appendages” (“critical number of nodes”), is indicated to be an index with a high degree of stability according to years within the range of a certain cultivar (5). Huang (6) expresses certain doubt regarding the absolute value of the “critical number of nodes”.

The studies of flower bud formation in the apple are numerous (2, 3) but they are conducted on budded trees. The literature available to us lacks data on flower bud formation in the own-rooted apple trees. The existing studies of such trees are mainly related to determining the growth characteristics and productive potentialities of the separate cultivars, regardless of the fact whether the plants are produced by the traditional methods (4) or by means of in...
vitro propagation (11, 12, 13).

The main objective of our study was to determine the time of differentiation of the flower buds of the own-rooted apple trees and its connection with the leaf-like appendages.

**Materials and Methods**

The study was conducted in the experimental field of the Agricultural University in Polvdiv, Bulgaria. The own-rooted trees of the apple cultivar Golden Resistant produced by trench layering from virus-free mother plants, whose rooted shoots were additionally cultivated for a season in a nursery, and also those budded on seedling rootstock, were planted in the autumn of the year 1985 within the range of a special experiment initiated with the purpose of testing the suitability of different cultivar-rootstock combinations for growing in intensive plantations. For the sake of monitoring the differentiation of flower buds during the period from the year 2002 to the year 2005, 6 tree-replications were selected out of the two variants, situated in a randomized block design. Samples of spurs with externally well-formed apical buds were collected annually from the beginning of the month of June until the middle of July every 3 - 4 days, and also from the middle of July to the end of August – on a weekly basis. Only spurs growing from the second year part of prolonged shoots, well-lit by the sun, were collected. In this way the eventual influence of the old wood of the composite fruit branches, which are known (7) for having earlier time of flower bud initiation, was avoided. Branches without fruit were selected to decrease the influence of fruit, which retains flower bud formation. The buds of the spurs collected from them (3 per tree on every date) were opened immediately by means of a preparatory needle under a stereomicroscope or were temporarily preserved in a fridge in a solution of formalin, 70% ethyl alcohol and concentrated acetic acid in a volumetric proportion 10:50:5.

The number of leaf-like appendages (bud scales, transitional leaves, true leaves and bracts) on the central axis of the differentiating flower buds and the visible changes in the bud apexes were recorded before and during the time of the appearance of bracts and sepals in the apical flower of the inflorescence.

**Results and Discussion**

The rate of increasing of the number of leaf-like appendages on the bud axes from the middle of the month of June and further on, was comparatively constant until the appearance of the first bract and the first sepal appendages on the apical flower of the inflorescence (Fig. 1). The first bract (Fig. 2) was observed towards the end of the first half of the month of July in the year 2002 and 2003 and 2 – 3 weeks earlier (during the last ten-day period of the month of June) in the next two years. The period from the termination of full bloom to the appearance of the first bract was approximately identical (from 67 to 78 days) in the year 2003, 2004 and 2005. The only exception was the year 2002 during which the indicated period was 100 days. In that year the phenophase of flowering occurred unusually early and continually (data are not presented) because of the warmer weather in the second half of winter. As early as the time of flowering, the warming was followed by lowering of the 24-h temperatures and frequent rainfalls. Besides, the sum of effective temperatures specified over the basic temperature of 10 degrees C (9) from the end of full bloom to the appearance of the first bract, was 870 degrees C. During the following year (2003) this sum was approximately identical, while in the year 2004 and 2005 it was considerably smaller (from 526 to 617).

When observing the apical meristem in the course of our experiment, we registered its different states which we could consider the first sign of bud transition from the
Fig. 1. Rate of production of leaf-like appendages in differentiating flower buds in the apple cv. “Golden Resistant”.

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vegetative into the reproductive state, manifested by the widening and slight swelling of the meristem. However, at hand was a real possibility of introducing an element of subjectivity in specifying this sign. Taking into consideration this and the standpoint expressed by other authors (10), that with altering the state of the buds, the apical meristem is in a critical state precisely during the period after the appearance of the last true leaf and before the appearance of the first bract, we accepted the appearance of the first bract, referring it to the “critical number of appendages”, as a reliable visible sign of the onset of the morphological differentiation. Thereby completed, the “critical number of nodes” in our study was a comparatively constant value according to years – 16,22 and 16,27 in the year 2002 and from 15,00 to 15,67 in the next three years. The number of appendages formed until the appearance of the sepals of the apical flower, was from 17 to 17,80.

In the three out of four years of study, the onset of the morphological differentiation was observed 10-11 weeks after the termination of full bloom, a period reported by other authors as well (8). There was a longer period in the year 2002, characterized by unusually early and prolonged flowering caused by the great deviations in the meteorological conditions. As far as the sum of effective temperatures is concerned during the indicated periods of time, it is difficult to make an exact explanation of the differences according to years, taking into consideration the viewpoint of McArtney et al. (9) according to which it is also possible for the temporary changes in air temperatures, difficult to establish in heat accumulation during the comparatively long period from flowering to morphological differentiation, to interfere with the development of flower buds.

The own-rooted and budded trees of the cultivar included in our study, had an almost identical rate of initiation of leaf-like

Fig. 2. Longitudinal bud section, showing domed apex (a), first bract (b) and upper true leaf (c). The other leaf-like appendages removed.
appendages on the central axis of the differentiating flower buds, and formed almost the same “critical number of appendages” before the onset of the morphological differentiation. The lack of significant differences in the “critical number of appendages” is also established in examining the influence of rootstocks differing in their growth vigour, on flower bud formation in the trees of the cultivar Starkspur Supreme Delicious (5). It could be assumed that the type of root system of the apple trees is of no particular importance for the term of the onset of the morphological differentiation of the flower buds.

Conclusions
The obtained results give us grounds for the so-called “critical number of appendages” including bud scales, transitional leaves, true leaves and the first bract, all formed on the nodes of the central axis of the differentiating flower buds in the apple, to be accepted as a reliable index for determining the onset of morphological differentiation. In the cultivar Golden Resistant this number is 15-16, no matter whether the trees are own-rooted or budded.

Excluding the years of unusual deviations in the meteorological conditions for a particular region, the number of days from the termination of full bloom to the formation of the first bract, can be used as a reference point index for prognosticating the onset of morphological differentiation of buds. This period for the cultivar Golden Resistant is 10-11 weeks.

Taking into consideration the lack of information about flower bud formation in the own-rooted apple trees, it could be assumed that the results obtained in this study are of original nature. They can be of considerable importance for studying the stages of flower bud formation in the in vitro propagated plants which have been of particular interest lately.

REFERENCES