

Comparison of photosynthetic pigment contents in stems and leaves of fruit trees: cherry, sweet cherry, common plum, and walnut tree

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ABSTRACT

The investigations were carried out on current-year, 1-, 2-, 3-year old stems and leaves of fruit trees: cherry, sweet cherry, pear, common plum, and walnut. The content of chlorophyll *a* and *b* and the sum of the carotenoids were determined.

In the examined species of trees, the content of chlorophyll increased with the age of stems, except the cherry tree, in which its content decreased. The content of chlorophyll in the current-year stems amounted from 1.6 to 2.2 mg dm⁻², in 3-year old stems it was 1.6-2.8 mg dm⁻², and in leaves its amount varied from 2.7 to 4.5 mg dm⁻². The 3-year old stems, when compared to the current-year stems, contained about 40-50% more chlorophyll in common plum, walnut and pear trees, and about 85% more in sweet cherry tree. In the cherry tree the amount of chlorophyll decreased by 15% during the time of the experiment. In comparison

with the leaves, the stems contained similar amounts of chlorophyll in sweet cherry and walnut trees, or smaller amounts in the common plum tree by about 40%, in the pear tree – by about 50%, and in the cherry trees by nearly 70%. In all the trees the value of the chlorophyll *a/b* ratio in the stems was considerably smaller in comparison with the leaves.

The content of carotenoids in the current-year stems varied from 0.5 to 1.4 mg dm⁻², and with age of the stems it increased to 0.6-1.4 mg dm⁻² in the 3-year old stems, and in the leaves it amounted from 0.9 to 1.3 mg dm⁻². Compared to the leaves, the stems contained similar amounts of carotenoids in sweet cherry and common plum trees, whereas the amount of carotenoids was higher in the leaves of the cherry and pear trees and smaller in the leaves of walnut tree.

The ratio of the content of chlorophyll to that of carotenoids oscillated from 2 to 4. In the stems and leaves of the sweet cherry and the pear trees it was similar. In the common plum and walnut trees its value in the leaves was higher in comparison with that in the stems, and in the current-year stems of the cherry and plum trees it was greater in comparison with older year groups of stems.

The results demonstrated that in most examined species bark chlorophyll content increased with the age of stems, the fact not reported in the literature before.

INTRODUCTION

The presence of photosynthetic pigments in stems of trees and shrubs is a generally observed feature. Cork which covers stems masks the existence of pigments and for that reason the presence of chlorophyll and carotenoids in bark often escapes our notice. As it has been revealed by investigations conducted on various species of lignified plants, in the young stems chlorophyll and carotenoids are present in all tissues under cork (Pilarski and Tokarz 2006, Wittmann et al. 2006), and both chloroplasts in bark (Pilarski 1990) and in parenchyma cells of the pith (van Clave et al. 1993) are photosynthetically active. With the age of stems and the increasing thickness of cork, phloem and xylem, the transmittance of irradiation to the inside of the stems decreases and gradual decay of photosynthetic pigments in deeper situated tissues can be observed. However, in species in which dead cells of cork do not accumulate, chlorophyll is present under cork in the trunks of even 100 year old trees, as it was observed in beech (Pilarski and Tokarz 2006).

The content of photosynthetic pigments in stems is directly influenced by optical properties of cork, particularly the amount and spectral composition of irradiation transmitted by cork and reaching chlorophyll cells in stems.

In the photosynthetically active range, cork transmittance increases with increasing wave length. Irradiation reaching chlorophyll cells in stem is especially

poor in the violet-blue range (400-500 nm) with greatest amount of red radiation (650-700 nm) present in its composition (Pilarski 1989, Pilarski and Tokarz 2005, Tokarz and Pilarski 2005).

The investigation carried out on the apple tree (Tokarz and Pilarski 2005) showed that transmittance of irradiation by the cork of current-year stems is equal to 30%, in 3-year old stems it decreases to 23%. The content of chlorophyll and the proportions of chlorophyll *a/b* at that time remained on similar level, whereas the content of carotenoids increased with the age of stems. In lilac, the transmittance of cork decreased with the age of the stems, from 17% in the current-year stems to 10% in 3-year old stems. During that time the content of chlorophyll and carotenoids decreased by about 25%, the ratio of chlorophyll *a/b* was slightly diminished and photochemical activity of chloroplasts isolated from the bark decreased. Nevertheless, chloroplasts from 3-year old stems still demonstrated 60% of the activity of chloroplasts from the current-year stems.

Optical properties of stems depend on plant species. Investigation carried out on the fruit trees: pear, cherry, sweet cherry, common plum, and walnut have shown the presence of considerable differences between them (Pilarski and Tokarz 2005). The transmittance of irradiation by the cork of the current-year stems oscillated from 14% in the sweet cherry tree and common plum tree, 20% in the cherry tree and pear tree to 26% in the walnut tree, and in the 3-year old stem it decreased to 15% in the pear tree, 10% in the cherry tree, common plum tree, and walnut tree, and only 6% in the sweet cherry tree.

Irradiation, its intensity and spectral composition, have a direct effect on the amount and composition of photosynthetic pigments, and these in turn have a direct effect on photosynthetic activity of stems. Stems are photosynthetically active during the whole year and, as it has been demonstrated by investigations carried out on various tree species, their proportion in the carbon balance of the tree may be considerable. During the leafless period, particularly before the development of leaves and the resulting great demand for assimilates in spring, they are the only source of assimilates from the current production (Pilarski 1990). In fruit farming the density of planting, the pruning and forming of the trees guarantee considerably greater access of irradiation to the chlorophyll layer of the bark in comparison c.g. with forest complexes, and therefore photosynthetic activity of the stems of fruit trees may be of greater importance in the carbon balance of a plant in comparison with the forest trees.

The aim of the investigations was to compare the content of photosynthetic pigments in the leaves and stems of various age in fruit trees, which are of considerable economic importance in moderate climate.

MATERIAL AND METHODS

The experiments were conducted on current-year, 1-, 2-, 3-year old stems and the leaves of the following fruit trees: sweet cherry (*Prunus avium* L. 'Hedelfinger'), cherry (*Prunus cerasus* L. 'Łutówka'), pear (*Pirus communis* L. 'Konferencja'), common plum (*Prunus domestica* L. 'Węgierka Zwykła') and walnut trees (*Juglans regia* L.). Materials were collected from five trees growing in the Garlica Murowana Pomology Experimental Station near Kraków. The content of photosynthetic pigments in the bark of stems of various age and in leaves was determined in July, 2005.

The content of photosynthetic pigments was determined in 80% acetone by the method described by Wellburn (1994). The content of chlorophyll *a* and *b* and the sum of carotenoids in the bark of stems and the leaves were determined, and the obtained results were presented in mg dm^{-2} of the surface.

The measurements were carried out in five replicates on material collected from various trees and the results were presented as arithmetic means with the standard deviation.

RESULTS

Fig. 1 shows the results obtained on sweet cherry tree. With the age of stems the increase in the content of chlorophyll *a* and *b* in the bark was observed. It was particularly distinct in the 3-year old one. In the examined period the content of chlorophyll *a* increased by 70% and that of chlorophyll *b* – by 100%, and the total content of chlorophyll increased by 80%. In comparison with the stems, the content of chlorophyll *a* in the leaves was greater, but the content of chlorophyll *b* was smaller than in 3-year old stems and similar to its content in the younger stems. Total content of chlorophyll *a+b* in the leaves was similar to its content in the 3-year old stems (2.7 mg dm^{-2}) and distinctly higher in comparison with the younger age groups of stems ($1.55\text{-}2 \text{ mg dm}^{-2}$). The ratio of the content of chlorophyll *a/b* in the stems shows the tendency to decrease with the age of stems from 2.1 in the current-year stems to about 1.7 in 2- and 3-year old ones. In the leaves it was higher to 3.1. The content of carotenoids in the stems also showed the tendency to increase with the age ($0.49\text{-}0.78 \text{ mg dm}^{-2}$) and in the leaves it was similar to their content in 3-year old stems. The calculated ratio of the total content of chlorophyll to that of carotenoids was 3.1-3.5 and there were no essential differences between the particular year groups of stems also in comparison with the leaves.

The results obtained on a sour cherry tree are shown in Fig. 2. In this case the content of chlorophyll decreased with the age of the stems and the reduction in the examined period was equal to about 15%. In the leaves, chlorophyll *a* content was

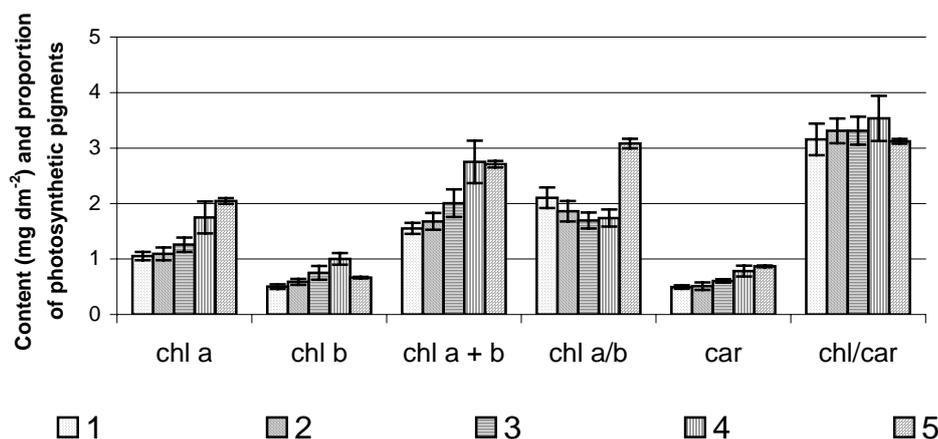


Fig. 1. The content of photosynthetic pigments (mg dm^{-2}) and their proportions in the stems and leaves of a sweet cherry tree. 1 – stem of current-year, 2 – stem of 1-year old, 3 – stem of 2-year old, 4 – stem of 3-year old, 5 – leaf

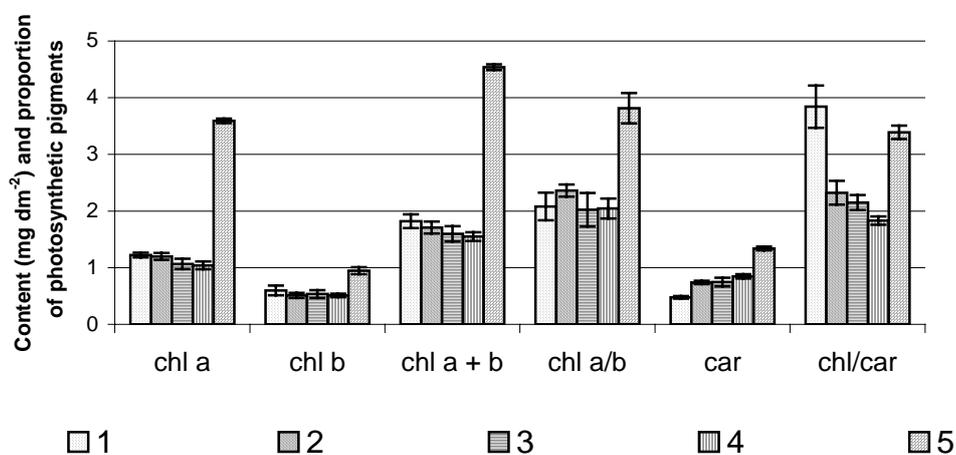


Fig. 2. The content of photosynthetic pigments (mg dm^{-2}) and their proportions in the stems and leaves of a sour cherry tree. 1 – stem of current-year, 2 – stem of 1-year old, 3 – stem of 2-year old, 4 – stem of 3-year old, 5 – leaf

almost 3 times greater and that of chlorophyll *b* – two times greater in comparison with the stems. The total content of chlorophyll in the stems was reduced from 1.8

mg dm⁻² in the current-year stems to 1.5 mg dm⁻² in the 3-year old ones, and in the leaves its amount was 4.5 mg dm⁻². The ratio of the content of chlorophyll *a/b* in the stems ranged from 2.0 to 2.3, depending on the stem's age, and in the leaves this ratio was greater, amounting to 3.8. Unlike in the case of chlorophyll, the content of carotenoids increased with the age of stems in the examined period by 80%, from 0.50 mg dm⁻² in the current-year stems to 0.85 mg dm⁻² in 3-year old stems, and in the leaves it was about two times greater in comparison with the stems (1.34 mg dm⁻²). The ratio of chlorophyll to carotenoids in the current-year stems was equal to 3.9 and decreased with the age of stems to 2.3 in the 2-year old stems to 1.9 in 3-year old ones. In the leaves its value was 3.5.

The results obtained on a pear tree are given in Fig. 3. In the stems of this species chlorophyll *a* and *b* and their sum content showed an increasing tendency with the stems age, and in the examined period the increase in their content was equal to about 15%. In the leaves, the content of chlorophyll *a* was about two times greater and that of chlorophyll *b* by about 35% higher in comparison with the stems. The ratio of chlorophyll *a/b* in the stems was similar, amounting to about 2.3 whereas in the leaves it was equal to 3.9. The content of carotenoids in the stems in the examined year groups was similar – about 0.5 mg dm⁻², while in the leaves it was two times higher – 1.1 mg dm⁻². The content of chlorophyll in comparison with the content of carotenoids was 3.5-4 times greater and there were no essential differences in the value of this ratio between the year groups of the stems and the leaves.

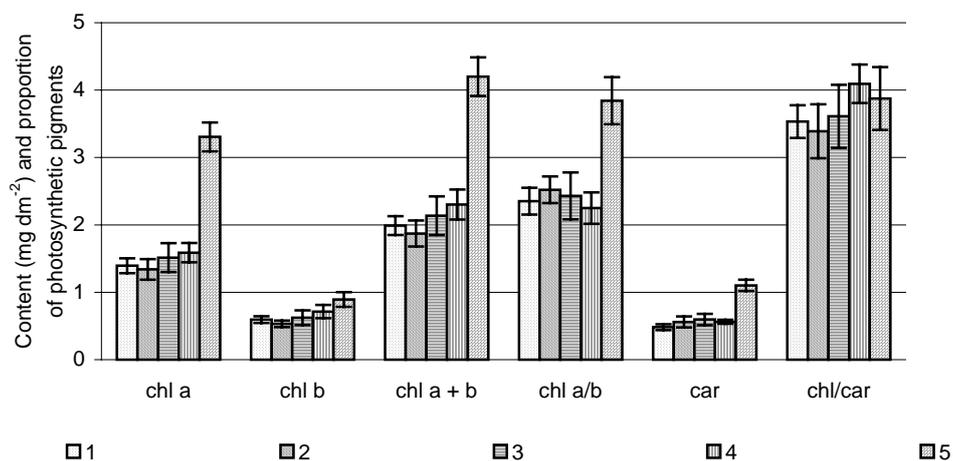


Fig. 3. The content of photosynthetic pigments (mg dm⁻²) and their proportions in the stems and leaves of a pear tree. 1 – stem of current-year, 2 – stem of 1-year old, 3 – stem of 2-year old, 4 – stem of 3-year old, 5 – leaf

The results obtained on a plum tree are shown in Fig. 4. With the age of the stems, the increasing tendency in the content of chlorophyll *a* and *b* and the carotenoids was observed. This increase continued till the stems were 2-years old and the content of chlorophyll increased by about 19% and that of carotenoids by 57%. The leaves contained about 2 times more chlorophyll *a*, while the amount of chlorophyll *b* was similar to that in the stems. The ratio of chlorophyll *a/b* in the stems was alike and equal to about 2.7, and in the leaves it was 3.8. The content of carotenoids in the stems and leaves was similar. The value of the proportion of the content of chlorophyll to that of carotenoids in the current-year stems was 3.4 and in the older year groups of stems it was evidently smaller – about 2.5, whereas in the leaves it was the highest – 4.1.

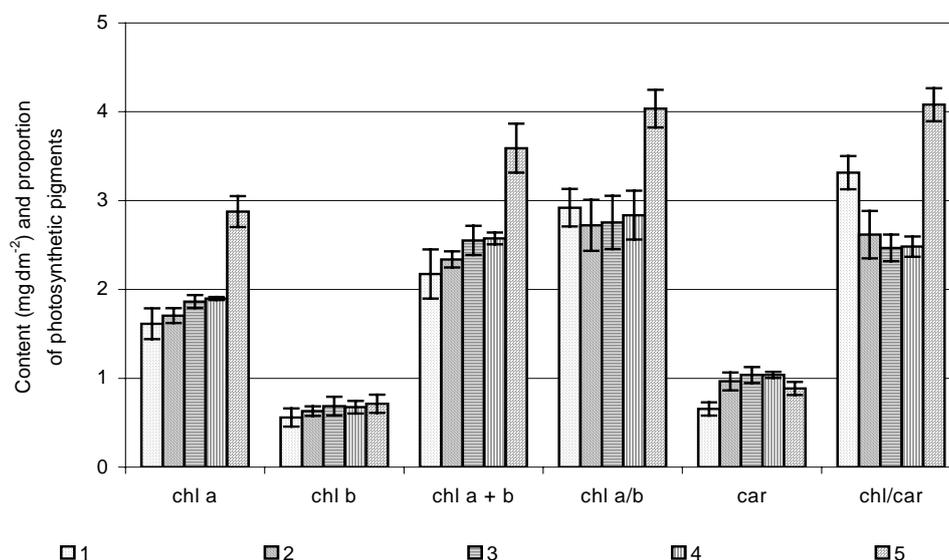


Fig. 4. The content of photosynthetic pigments (mg dm^{-2}) and their proportions in the stems and leaves of a common plum tree. 1 – stem of current-year, 2 – stem of 1-year old, 3 – stem of 2-year old, 4 – stem of 3-year old, 5 – leaf

Also in the walnut tree (Fig. 5), the content of chlorophyll *a* in the bark increased by about 40%, and that of chlorophyll *b* – by about 60%, and its total content – by 45% with the age of the stems. In the leaves, the content of chlorophyll *a* was by 10% higher, whereas that of chlorophyll *b* by about 50% lower, and its total content was by 10% lower in comparison with the bark of 3-year old stems. The ratio of chlorophyll *a/b* in the stems changed little with the stems age and it amounted to 1.8-2.2, whereas in the leaves it was much higher, being equal to 4.3. The content of carotenoids increased with the age of stems from 1.0 mg dm^{-2} in the current-year stems to 1.4 mg dm^{-2} in the 3-year old ones, and in

the leaves the amount of carotenoids was about 0.9 mg dm^{-2} . The ratio of the content of chlorophyll to that of carotenoids in the stems was similar, equal to about 2, and in the leaves it was equal to 2.9.

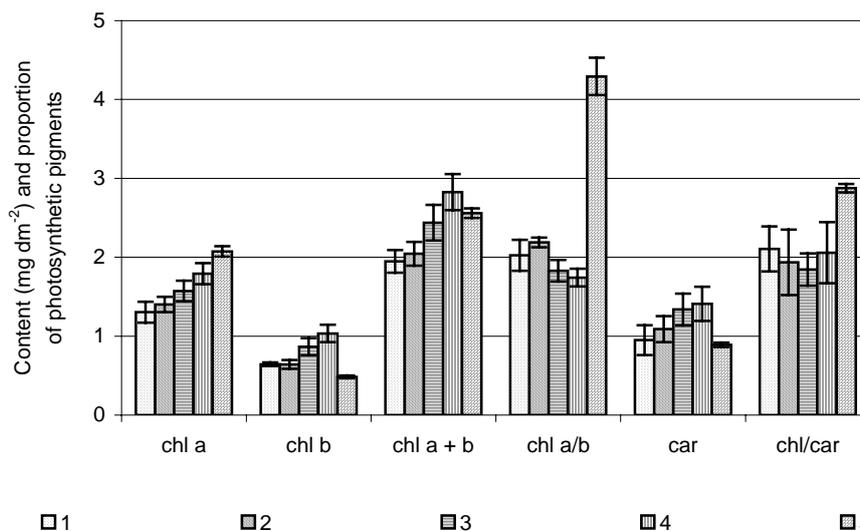


Fig. 5. The content of photosynthetic pigments (mg dm^{-2}) and their proportions in the stems and leaves of a walnut tree. 1 – stem of current-year, 2 – stem of 1-year old, 3 – stem of 2-year old, 4 – stem of 3-year old, 5 – leaf

The presented results show that in the examined species of trees the content of chlorophyll increases with the age of the stems, except for sour cherry tree, in which it's content decreased with the stems age. In the stems, the total content of chlorophyll oscillated from 1.7 to 2.8 mg dm^{-2} , and in the leaves from 2.7 to 4.5 mg dm^{-2} . The stems contained similar amounts of chlorophyll as the leaves of the sweet cherry and the walnut trees or smaller amounts – in the plum tree by about 30%, in the pear tree by about 50%, and in the sour cherry tree – almost by 60%. In all trees the value of the chl *a/b* ratio in the stems was lower in comparison with the leaves. In all species the content of carotenoids increased with the age of the stems. In the current-year stems it oscillated from 0.5 to 1.0 mg dm^{-2} , and in the 3-year old ones it increased from 0.6 to 1.4 mg dm^{-2} . In the leaves it amounted from 0.9 to 1.3 mg dm^{-2} . In comparison with the leaves, the stems of the sweet cherry and the plum trees contained similar amounts of carotenoids and their number was greater in the leaves of sour cherry and pear trees and smaller in the leaves of walnut tree.

The ratio of the content of chlorophyll and that of carotenoids oscillated from 2 and 4, and varied. It was similar in the stems of various age and in the leaves of

sweet cherry and walnut trees. In the latter, it was similar in the stems, but higher in the leaves. In the current-year stems of the sour cherry tree its value was higher in comparison with that of the leaves, but in the older year groups of the stems it was only half as high in comparison with the current-year stems.

DISCUSSION

In young stems the photosynthetic pigments are present in all tissues, except cork, but their distribution may vary. In *Euonymus europea* (Szujko-Lacza et al. 1972) they were located in ca 90% in the bark, in ca 8% in the xylem and ca 2% in the core, but in the beech tree 50% of chlorophyll was found in the bark, 30% in the xylem and 20% in the core (Larcher et al. 1988). In the core, the photosynthetic pigments are located only in the thin layer of cells adhering to the xylem, e.g. in lilac, or are presented in all its cells, e.g. in the beech tree, as it was revealed by the photographs of chlorophyll fluorescence (Pilarski and Tokarz 2006, Berveiller et al. 2007).

Investigations concerning chlorophyll content in the stems and leaves have shown that in the stems its content may be only slightly smaller in comparison with the leaves, e.g. in *Populus tremula* (Pearson and Lawrence 1958, Barr and Potter 1974), or when compared with the leaves, the stems contain about 1/3 less chlorophyll, e.g. in *Syringa vulgaris* (Pilarski 1984, 1999), *Ilex aquifolium* (Schmidt et al. 2000), and even by 2/3 less, e.g. in *Fagus sylvatica* (Pfanzen and Aschan 2001). In most investigations the effect of the age of stems was not taken into consideration. In the investigation carried out on *Syringa vulgaris* (Pilarski 1984, 1999) it was demonstrated that with the age of stems, chlorophyll content in the bark of the stems decreases, but in the apple tree the reduction of the amount of chlorophyll in the 3-year old stems in comparison with the younger age groups of the stems was not observed (Tokarz and Pilarski 2005).

The present investigations have confirmed the difference between the tree species: the content of chlorophyll in the stems, in comparison with that in the leaves, was similar in the sweet cherry tree and walnut tree, smaller by about 30% in the plum tree, ca 50% in pear tree and by ca 70% in sour cherry tree. With the age of the stems, according to the authors' investigations, the content of chlorophyll decreased only in the sour cherry tree, similarly as it was in the case of lilac (Pilarski 1984). In the other examined species, the content of chlorophyll increased and in the sweet cherry tree, in which the increase was the greatest, in the examined period the 3-year old stems contained ca 80% more chlorophyll in comparison with the current-year stems. Such a case has not been reported in the literature. The ratio of chlorophyll *a/b* in the stems of the trees examined by the authors was similar to the earlier reports carried out on lignified (Larcher et al.

1988) and herbaceous plants (Oelmüller and Mohl 1985). In the stems, their value is considerably smaller in comparison with the leaves as a result of relatively greater content of chlorophyll *b*. This is due to lower irradiation reaching the chloroplasts in the stems of lignified plants, expanded antennae absorbing the penetrating irradiation and smaller density of chlorophyll, since in the stems it is situated in a considerably thicker layer of cells. With the age of the stems, a reduction in the value of the ratio of chlorophyll *a/b* is noticed due to increasing content of chlorophyll *b* in comparison with chlorophyll *a*.

Additionally, the increasing content of carotenoids with the age of the stems indicates their important role in the absorption of the penetrating irradiation. In the stems of herbaceous plants, the protective role of carotenoids against photodestruction of chlorophyll through a xanthophyll cycle was demonstrated (Oelmüller and Mohl 1985). In the stems of plants lignified as a result of small amount of the penetrating irradiation they play an important role in its absorption and transmission into chlorophyll (Young et al. 1997, Nobel 2005), and only in autumn, when leaves have dropped, they can protect against photoinhibition (Solhaug and Haugen 1998, Damesin 2003, Manetas 2004). In all examined trees, the diminishing amount of irradiation transmitted to the chlorophyll layer of the bark was accompanied by the increase of the content of carotenoids. These results are an indication of their importance increasing with the age of the stems. A comparison of the content of photosynthetic pigments with the optical properties of the stems of the species of the fruit trees investigated by the authors (Pilarski and Tokarz 2005) shows that there are no explicit dependences between the transmission of irradiation PAR by the cork and the content of photosynthetic pigments. In the pear, sour cherry and walnut trees the transmission of irradiation by the current-year cork was 20-25% and the bark contained 1.02-1.99 mg dm⁻² of chlorophyll and the content of carotenoids amounted from 0.48-0.95 mg dm⁻². In the sweet cherry tree the transmission was lower: 14%, and the bark contained less chlorophyll: 1.55 mg dm⁻² and 0.5 mg dm⁻² of carotenoids, however in the plum tree the transmission was also equal to 14%, while the amount of chlorophyll was 2.17 mg dm⁻² and that of carotenoids was 0.66 mg dm⁻².

The transmission of irradiation reduced nearly by about 50% in the 3-year old stems in comparison with the current-year stems and it was accompanied by a considerable increase of chlorophyll in the pear, sweet cherry and plum trees, but by a distinct decrease in the sour cherry tree. In the sweet cherry tree, the PAR transmission in the 3-year old stems was the smallest among the examined species and it amounted to 6%, whereas the content of chlorophyll was the highest. This observation may also suggest that increased amount of chlorophyll facilitates the observation of the diminishing irradiation. Present day orchards are planned so as to make the best possible use of the available space. Thus, the surface of the stems appears to be important as it may represent, e.g. in the case of apple tree, from 30

to 70% of the entire photosynthetically active surface (Tokarz, unpublished). The additional energetic profit deriving from photosynthetically efficient surface of the stems of trees and bushes may represent, according to various authors, from a few to some ten to twenty percent (Kharauk et al. 1995, Pilarski 1999).

The demonstrated results indicate that the stems of the examined fruit trees contain considerable amounts of photosynthetic pigments, both in the current- and 3-year old stems. They are also the evidence of potential photosynthetic possibilities of the bark of stems of pear, sweet cherry, sour cherry, plum and walnut trees. The amounts of chlorophyll in some species, similar to its content in the leaves, and the surface of stems on the trees allow to assume that the participation of stems in the photosynthetic bounding of CO₂ by a tree may be considerable. At the present stage of the investigations, however, it is not possible to determine the extent of this participation.

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PORÓWNANIE ZAWARTOŚCI BARWNIKÓW FOTOSYNTETYCZNYCH W PĘDACH I LIŚCIACH DRZEW OWOCOWYCH: CZEREŚNI, WIŚNI, GRUSZY, ŚLIWY I ORZECHA WŁOSKIEGO

Streszczenie: Badania przeprowadzono na liściach, pędach bieżącego rocznika i 1 - 3-letnich pędach czereśni, wiśni, gruszy, śliwy i orzecha włoskiego. Oznaczano zawartość chlorofilu *a* i *b* oraz sumę karotenoidów.

W badanych gatunkach drzew z wiekiem pędów zwiększała się zawartość chlorofilu, za wyjątkiem wiśni, u której jego zawartość malała. Zawartość chlorofilu w pędach bieżącego rocznika wynosiła od 1,6 do 2,2 mg dm⁻², w pędach 3-letnich 1,6-2,8 mg dm⁻², a w liściach od 2,7 do 4,5 mg dm⁻². Pędy 3-letnie w porównaniu z pędami bieżącego rocznika zawierały o około 40-50% więcej chlorofilu u śliwy, orzecha i gruszy i o 85% więcej u czereśni, a u wiśni w tym czasie ilość chlorofilu zmniejszyła się o 15%. Pędy w porównaniu z liśćmi zawierały zbliżone ilości chlorofilu w przypadku czereśni i orzecha lub mniejsze ilości – u śliwy o około 40%, u gruszy o około 50%, a u wiśni prawie o 70%. U wszystkich drzew wartość stosunku chl *a/b* w pędach była znacznie mniejsza w porównaniu z liśćmi.

Zawartość karotenoidów w pędach bieżącego rocznika wahała się od 0,5 do 1,0 mg dm⁻² i z wiekiem pędów zwiększała się do 0,6-1,4 mg dm⁻² w pędach 3-letnich, a w liściach wynosiła od 0,9 do 1,3 mg dm⁻². Pędy w porównaniu z liśćmi zawierały podobne ilości karotenoidów u czereśni i śliwy natomiast więcej było ich w liściach u wiśni i gruszy, a mniej w liściach orzecha.

Stosunek zawartości chlorofilu do karotenoidów wahał się pomiędzy 2 a 4. W pędach i w liściach u czereśni i gruszy był podobny w pędach i liściach. U śliwy i orzecha w liściach miał wyższe wartości w porównaniu z pędami, a u wiśni i śliwy w pędach bieżącego rocznika był większy w porównaniu ze starszymi rocznikami pędów.

Przeprowadzone badania wykazały, że zawartość barwników chlorofilowych w pędach większości badanych gatunków wzrastała z wiekiem, co nie było dotychczas obserwowane.