Acta Agrophysica, 2012, 19(4), 865-876

INFLUENCE OF MICROWAVE-CONVECTIVE DRYING ON CHLOROPHYLL CONTENT AND COLOUR OF HERBS

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Abstract. The aim of this study was to analyse changes in chlorophyll a and b content, *a**, *b** and chroma colour parameters during microwave-convective drying of basil, lovage, mint, oregano, parsley and rocket leaves. Drying was conducted at 40°C in air flowing perpendicular to the material layer with velocity 0.8 m s^{-1}, and at 300 W of microwave power level. The results show that drying had a significant impact on chlorophyll degradation, however the level of its losses was different for each herb species. High retention of chlorophyll a was observed in lovage and basil, and chlorophyll b retention in mint, as well. Drying did not have any significant impact on chlorophyll a content only in the case of lovage. On the other hand, chlorophyll b contents in basil, lovage and mint were statistically constant. The greatest chlorophyll a and b losses were observed in rocket leaves. Significant decreases of *a**, *b** and chroma values, as a result of drying, were observed in all herbs, except rocket leaves. The smallest changes were noted for *Apiaceae* (lovage and parsley) and rocket (*Brassicaceae*). *Lamiaceae* (oregano, mint, basil) herbs were characterised by less stable colour. The reason for colour changes was also dependent on taxonomic group. The changes of green and yellow colour resulted from significant chlorophyll a content changes in *Apiaceae* herbs. Nevertheless, chlorophyll degradation was not the only reason for colour changes in the other herbs. High relationship between degradation of chlorophyll a and *a** parameter was observed in the case of all investigated herbs.

Keywords: herbs, microwave-convective drying, chlorophyll, colour

INTRODUCTION

Herbs are believed to be among the first food products used all over the world. Already in the ancient times they were used not only to give flavour to various kinds of food, but also for medicinal purposes. Initially herbs were grown in ancient Egypt, in the Tigris valley, in China and in India, and later also in

Europe. At present herbs are extensively used in the food, pharmaceutical and cosmetics industries, and the world production of herbs is estimated at 0.5 million tons a year. In the European Union cultivations of herbs occupy an area of about 70 thousand hectares, of which 30 thousand ha in Poland, with annual production at the level of ca. 22 thousand tons (Lutomski 2000).

Herbal materials are a valuable source of numerous biologically active substances, among which chlorophylls (porphyrinic derivatives) are responsible for the colour of herbs. In land plants two main types of chlorophyll appear – chlorophyll a and b (at the ratio of 3:1). They absorb visible light in different wavelengths, which is reflected in their different colours – blue-green in the case of chlorophyll a, and yellow-green of chlorophyll b. They display low stability during drying, and their degradation results primarily from the detachment of an magnesium atom, the effect of which is the formation of pheophytins (Hutchings 1994; Elbe and Schwartz 1996). The loss of magnesium leads to a reduction of the share of green colour in favour of olive-brown, but the final colour of the materials is related to the ratio of the chlorophyll types and their various derivatives.

The method most frequently used for the drying of herbal materials is the convective technique. Unfortunately, even the application of mild process conditions (low temperature) does not guarantee the retention of a high content of chlorophylls and unchanged colour (Di Cesare *et al.* 2003, 2004, Witrowa-Rajchert *et al.* 2009, Arslan *et al.* 2010, Alibas 2010). This results from the long duration of the process and from the superficial penetration of heat. In the case of microwave drying, the absorption of radiation results from ionic conductivity (acceleration of ions in fast-changing electromagnetic field) and from dipole rotation. The effect is friction, generating heat within the whole volume of material (Schiffmann 2006). Such a method of heating ensured better retention of chlorophyll and colour in the case of basil (Yousif *et al.* 1999; Di Cesare *et al.* 2003; Witrowa-Rajchert *et al.* 2009), oregano (Yousif *et al.* 2000; Di Cesare *et al.* 2004; Witrowa-Rajchert *et al.* 2009), rosemary (Arslan and Özcan 2008), mint (Arslan *et al.* 2010) and nettle (Alibas 2010).

The objective of the study was to examine the extent of changes in the content of chlorophyll a and b, a^* and b^* values and chroma as a result of microwave-convective drying of basil, lovage, mint, oregano, parsley and rocket leaves.

MATERIAL AND METHOD

The experimental material consisted of fresh leaves of six species of herbs: basil (*Ocimum basilicum*), lovage (*Levisticum officinale*), mint (*Mentha*), parsley (*Petroselinum crispum*), oregano (*Origanum vulgare*) and rocket (*Eruca vesicaria*). The materials came from herb plantations situated in Kraśnicza Wola, near

Grodzisk Mazowiecki. The herbs were grown in hydroponic cultures. The material was purchased in the months of October 2010 – February 2011.

Until the time of the experiment, the fresh herbs were kept at room temperature, with access to sunlight. Directly before the drying, healthy, ripe and uniform leaves were picked, without stems.

The process of drying was conducted in a laboratory microwave-convective dryer, using microwave power level of 300 W and air flow velocity and temperature of 0.8 m s^{-1} and 40ºC, respectively. Measurements of mass loss (with accuracy of 0.1 g) and temperature of the material were taken every 3 minutes. The process was continued till constant mass of product, repeating the experiment twice for each herb species.

The content of chlorophyll a and b was assayed according to the method proposed by Lichtenthaler (1987), consisting in methanol extraction of chlorophylls and spectrophotometric measurement of absorbance of the solution at various wavelengths. The content of chlorophyll a ($C_{chl(a)}$, mg·g d.m.$^{-1}$) and chlorophyll b ($C_{Chl(b)}$, mg·g d.m.$^{-1}$) was determined on the basis of the relation:

$$C_{chl(a)} = 12{,}7 \cdot A_{(663)} - 2{,}69 \cdot A_{(645)} \tag{1}$$

$$C_{chl(b)} = 22{,}9 \cdot A_{(645)} - 4{,}68 \cdot A_{(663)} \tag{2}$$

where: $A_{(663)}$, $A_{(645)}$ – absorbance at wavelengths of 663 and 645 nm, respectively.

Measurements of colour of fresh and dried leaves of all herb species in the system CIE $L^*a^*b^*$ were made with the reflection method, using a Minolta CR-300 chromameter. Fragmented leaves were placed one on top of another on a special attachment that permitted elimination of the effect of light in the room and ensured repeatable thickness of the measurement layer. The measurement was repeated 10 times, the fragmented leaves being arranged in a different way every time. Chroma, defining the ratio of the chromatic values, indicating colour intensity, was calculated on the basis of the formula (MacDougall 2002):

$$\text{Chroma } C = \sqrt{(a^*)^2 + (b^*)^2} \tag{3}$$

To determine homogeneous groups, not differing in the statistical approach (denoted with the same letters in the graphs), statistical analysis was performed, comprising Fischer's test and the Tukey HSD test. The Pearson linear correlation analysis permitted the identification of the degree and direction of correlation of the indicators studied, while two-factor analysis of variance allowed the estimation of which of the factors (process of drying or species variation) had a stronger

effect on the result of the determination. All analyses were conducted at significance level of $\alpha = 0.05$.

RESULTS AND DISCUSSION

The herbs were microwave-convective dried from the initial water content in leaves of oregano, parsley, lovage, mint, basil and rocket of 6.33, 7.96, 8.59, 8.85 11.01 and 14.21 kg H_2O·kg d.m.$^{-1}$, respectively. For all herbs the process lasted for 18 minutes, and the final water content was below 0.09 kg H_2O·kg d.m.$^{-1}$.

Chlorophyll a content in the fresh herbs varied from 17.62±0.52 mg·g d.m.$^{-1}$ (mint) to 13.19±0.34 mg·g d.m.$^{-1}$ (oregano), while chlorophyll b content was from 6.23±0.40 mg·g d.m.$^{-1}$ (lovage) to 5.14±0.46 mg·g d.m.$^{-1}$ (basil). The application of microwave-convective drying caused a significant decrease in the content of both chlorophyll pigments (Figs. 1 and 2). After the process, the content of chlorophyll a and b in oregano was 10.42±0.28 and 4.66±0.53 mg·g d.m.$^{-1}$, respectively, in rocket – 11.06±1.24 and 4.36±0.56 mg·g d.m.$^{-1}$, in basil – 13.04±1.09 and 4.86±0.39 mg·g d.m.$^{-1}$, in parsley – 13.43±0.44 and 4.78±0.07 mg·g d.m.$^{-1}$, in lovage – 14.20±0.82 and 5.78±0.46 mg·g d.m.$^{-1}$ and in mint – 14.23±0.40 and 5.57±0.21 mg·g d.m.$^{-1}$. The process of drying had a significant effect on the variation of the results, while no statistically significant differences were observed in chlorophyll b content in most of the herb species. A greater rate of loss was observed in the case of chlorophyll a, amounting to 6-32%, whereas the degradation of chlorophyll b was at the level of 5-25%. Chlorophyll a is, therefore, less stable than chlorophyll b, which is also supported by Di Cesare *et al.* (2003, 2004) and Witrowa-Rajchert *et al.* (2009).

Both in the case of chlorophyll a and b, the highest losses were observed in rocket (32 and 25%, respectively). In turn, high stability of chlorophyll a after the drying process was characteristic of basil and lovage, and of chlorophyll b – also of mint. The retention was above 90% of the content prior to the process. On the other hand, however, only in the case of lovage the process of drying permitted the obtainment of a product with statistically unchanged content of chlorophyll a. Whereas, lack of significant effect of the process of dehydration on degradation on chlorophyll b was observed in basil, lovage and mint leaves. High retention of chlorophyll was also observed by Polak *et al.* (2009) in freeze-dried leaves of lovage, where the retention of chlorophyll a and b, depending on the process parameters applied, varied within the ranges of ca. 77-94 and 76-93%, respectively.

Dried basil displayed higher retention of chlorophyll a and b compared to oregano. The rates of retention in basil were 91 and 95%, respectively, the corresponding values for oregano being 79 and 86%. An opposite relation was observed by Witrowa-Rajchert *et al.* (2009), according to whom a greater stability of chlorophyll

a was observed in oregano. Microwave-convective drying conducted by those authors caused 90% degradation o f chlorophyll a in basil, and 17% in oregano. In the case of chlorophyll b, the losses amounted to 16 and 13%, respectively.

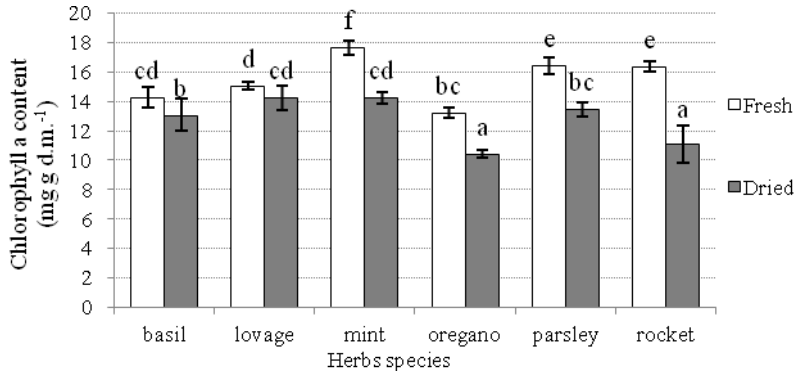

Fig. 1. Chlorophyll a content in fresh and microwave-convective dried herbs (average values with standard deviations); a, b, c, ... – the same letters mean homogeneous groups, not differing in statistical approach (α = 0.05)

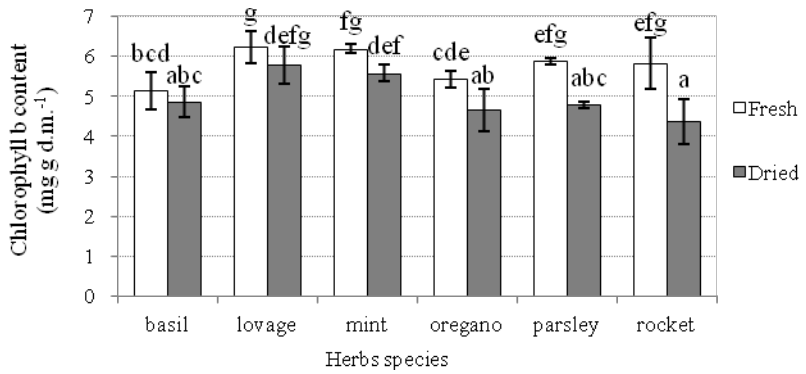

Fig. 2. Chlorophyll b content in fresh and microwave-convective dried herbs (average values with standard deviations); a, b, c, ... – the same letters mean homogeneous groups, not differing in statistical approach (α = 0.05)

Analysis of the obtained results does not permit the formulation of clear conclusions concerning the differences among the species in terms of the content of the two main types of chlorophyll. On the one hand, a distinct deterioration of the quality of rocket was observed after the process of drying, while dried lovage and basil were characterised by high retention of chlorophyll. However, within a sin-

gle family of herbs the stability of green pigments was varied. In the case of *Apiaceae* (which includes parsley and lovage) and *Lamiaceae* (basil, mint and oregano), retention of chlorophyll a varied within the ranges of 82-94 and 79-91%, respectively, while the corresponding values for chlorophyll b were 81-93 and 86-95%.

In the case of green leaves of plants, the most important index is parameter a^* of green-red colour. Changes in the values of that parameter as a result of drying of the herbs are presented in Figure 3. Green colour values (a^*) of fresh basil, lovage, mint, oregano, parsley and rocket were –10.50, –9.49, –8.49, –5.10, –10.43 and –8.78, respectively. The dried herbs were characterised by a notable change of green colour in favour of red, and the results of the determinations were significantly affected both by the species and by the drying process itself. Lovage retained the green colour to the greatest degree, while oregano was the least stable in this respect. The change of green colour was largely related with the structural similarity within a systematic family of herbs. Herbs from the family *Apiaceae* and rocket were characterised by a high tolerance to process conditions. The retention of parameter a^* in lovage, parsley and rocket amounted to 86, 75 and 73%, respectively. Lower stability was characteristic of oregano, basil and mint, belonging to the family *Lamiaceae*, in which the values of parameter a^* after the drying process were 17, 44 and 65% of the initial values, respectively. Significant changes in the colour of oregano and basil, subjected to freeze, convective and microwave drying, were also observed by Witrowa-Rajchert *et al*. (2009). They found a 50% increase in the value of a^* as a result of drying of basil, irrespective of the drying technique applied. The results of their study confirmed also the significant decrease of the share of green colour in oregano, to a level close to the value of –1, as was the case in the study reported here. In turn, retention of the value of parameter a^* amounting to 67-97% was observed by Alibas (2010) in microwave-convective dried nettle at various process parameters.

The change of green colour was largely related with the loss of chlorophyll. It was found that greater retention of chlorophyll a was accompanied by lesser change of green colour in the direction of red. The coefficient of correlation between the content of chlorophyll a and the colour parameter a^* determined for all the herbs ($r = –0.74$) indicates a strong inversely proportional relation, which was also supported by Witrowa-Rajchert *et al*. (2009). The correlation coefficients determined by those authors were $r = –0.98$ and $r = –0.94$, respectively, for basil and oregano dried with various methods.

The taxonomic classification of the herbs to particular families played a particular role in the relation between green colour and the content of chlorophyll a. Decrease in the content of chlorophyll a had no significant effect on the green colour change in the case of the family *Lamiaceae* (basil, mint, oregano). Though

the correlation coefficient within the family of herbs was $r = -0.78$, the level of the p-Value was higher (p-Value = 0.07) than the adopted level of significance ($\alpha = 0.05$). Whereas, a very strong negative correlation ($r = -0.98$) was noted in the case of lovage and parsley (*Apiaceae*), in which the change in the content of chlorophyll a had a significant effect on green colour (p-Value = 0.02).

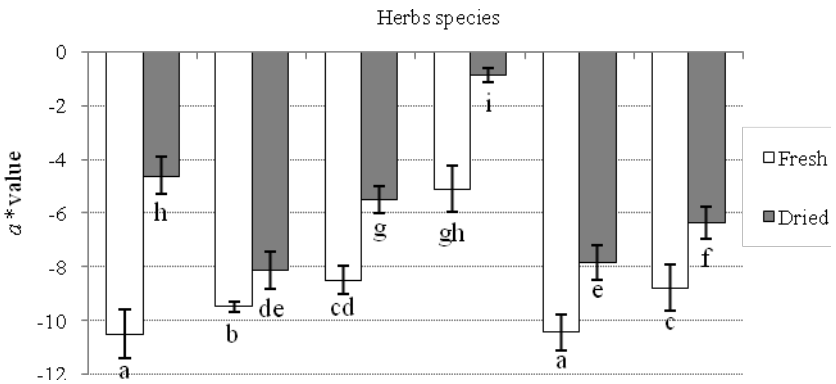

Fig. 3. Values of *a** parameter of fresh and microwave-convective dried herbs (average values with standard deviations); a, b, c, ... – the same letters mean homogeneous groups, not differing in statistical approach (α = 0.05)

The microwave-convective drying process caused also a significant change in the yellow colour (Fig. 4). In the case of lovage, mint, parsley, basil and oregano the values of parameter *b** decreased by 10, 13, 25, 38 and 50%, respectively, which was due to a greater extent to the drying, than related with the herb species. However, in both cases the effect was not statistically significant. Analogous changes in the yellow colour were observed during drying or rosemary (Arslan and Özcan 2008), coriander (Sarimeseli 2011) and mint (Arslan *et al.* 2010). Yellow colour loss of microwave-convective dried nettle varied from 2 to 27%, depending on the process parameters applied (Alibas 2010).

In the case of rocket, an increase in the level of yellow colour after the drying process was observed. That was related with a brightening of the colour and with high degradation of chlorophylls, as compared to the other herbal materials. The loss of a considerable part of green pigments could, in turn, cause that other pigments, e.g. carotenoids, became more visible. Carotenoids are presented in chloroplasts (located mainly in leaves), together with chlorophylls. Rocket contains considerable amounts of carotenoids. According to Michalczyk and Macura (2008), their content in fresh rocket leaves is above 20 mg $(100\ g)^{-1}$. Moreover, Polak *et al.* (2009) demonstrated that losses of chlorophyll pigments are greater

than losses of carotenoids, which brought them to the conclusion that this causes a colour change from the green-blue to the green-yellow. A similar increase in the level of the yellow colour was observed by Demir *et al*. (2004) in bay leaves dried with various methods (convective, exposed to sunlight and in a darkened room). The value of parameter *b** in their study was from 18 to as much as 25-fold higher than in fresh bay leaves.

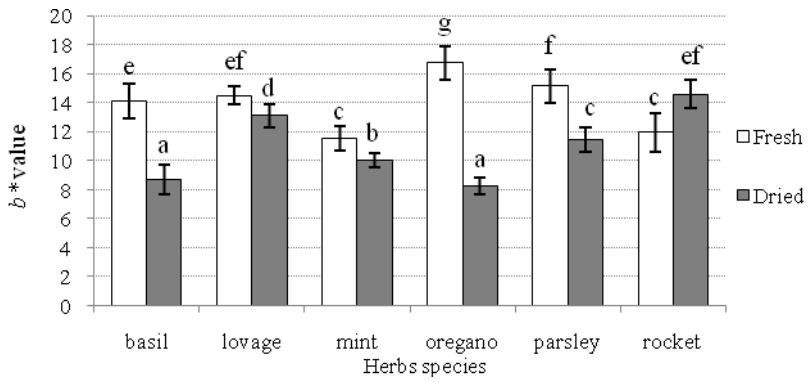

Fig. 4. Values of *b** parameter of fresh and microwave-convective dried herbs (average values with standard deviations); a, b, c, ... – the same letters mean homogeneous groups, not differing in statistical approach (α = 0.05)

Analysis of the relationship between the chlorophylls content and the values of the yellow colour parameter of basil, mint and oregano (*Lamiaceae*) did not reveal any significant correlation (*r* below 0.40). Whereas, in the case of the family *Apiaceae* (lovage, parsley) decrease in the value of the yellow colour resulted from a decrease in the content of chlorophyll a (r = 0.95).

One of the more important parameters defining the share of the chromatic parameters (*a** and *b**) in the overall perception of colour is colour saturation or chroma (C). That parameter permits the determination of the strength of response to the hue of a colour in a qualitative manner, through interpretation of its intensity and depth (Gozdecka 2006). The less saturated the hue, the more it appears "bleached", pale, which is reflected in strong positive correlation between the brightness and saturation of colour (r = 0.74). The drying process is significantly responsible for the change of saturation (p-Value = 0.04), while the effect of the species of the herbs was not significant.

The initial level of achromaticity varied within the range of 14.32 ± 0.97 (mint) – 18.40 ± 1.26 (parsley). In turn, as reported by Witrowa-Rajchert *et al.* (2009), colour saturation reached the values of 29.0 in basil and 21.1 in oregano.

The extent of changes in chroma values of basil, lovage, mint, oregano, parsley and rocket in microwave-convective drying was presented in Figure 5. The decrease in the chroma values, compared to the fresh herbs, was the least observable in materials belonging to the family *Apiaceae* and amounted to 11% in lovage and 25% in parsley. Whereas, *Lamiaceae* (basil, mint, oregano) were characterised by a greater extent of changes, on average by 25%, which gave lower values of chroma, by 20, 44 and 52%, respectively, compared to the fresh leaves of mint, basil and oregano. Highly similar results were obtained in the case of basil and oregano studied by Witrowa-Rajchert *et al.* (2009). After microwave drying of those herbs, their chroma values decreased by ca. 50 and 53%, respectively. In turn, good retention of colour were characteristic of nettle (Alibas 2010) and coriander (Sarimeseli 2011). Their mean chroma values after microwave-convective drying amounted to 81 and 98%, respectively, relative to fresh leaves.

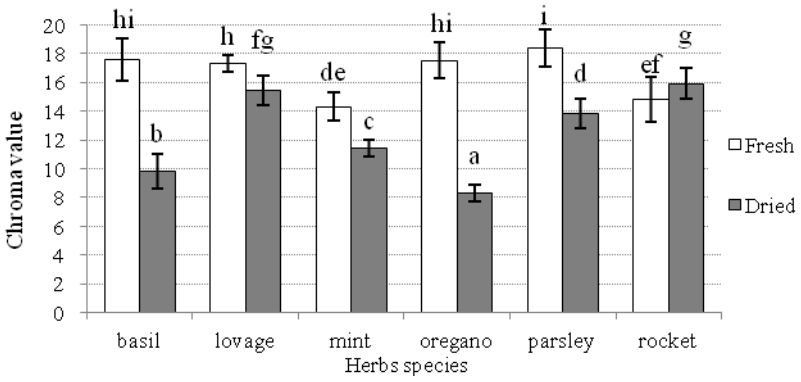

Fig. 5. Chroma values in fresh and microwave-convective dried herbs (average values with standard deviations); a, b, c, ... – the same letters mean homogeneous groups, not differing in statistical approach ($\alpha = 0.05$)

The increase of chroma value of dried rocket resulted from increase in the level of yellow colour (Figs. 4 and 5). The increase of chroma value was 7%, at a 22% increase in the value of parameter *b**. In turn, Demir *et al.* (2004) observed notably greater changes when drying bay leaves with various methods. The average increase in chroma value was 20-fold, while that of the yellow colour parameter – about 22-fold.

Lovage, in which chlorophyll retention was the highest, was characterised by the greatest colour stability. Generally, it can be stated that the change of colour of the herbs was dependent on their taxonomic classification. The very strong correlation in the case of *Apiaceae* (lovage, parsley) between the colour parame-

ters *a**, *b** and chroma value and the content of chlorophyll a permits the conclusion that it was chlorophyll a degradation that contributed to the greatest extent to the colour change. This means that in the case of the remaining herbs decrease in the content of green pigments is not the only cause of colour change. Probably, also important in this respect are the reactions of enzymatic and non-enzymatic browning (Witrowa-Rajchert *et al*. 2009) or the degree of carotenoids degradation (Polak *et al*. 2009), taking place during drying.

CONCLUSIONS

1. Degradation of chlorophylls and colour changes were observed as a result of microwave-convective drying of herbs. The changes were related to the herb species, and materials belonging to a single family were characterised by similar stability. Herbs from the family *Apiaceae* (lovage, parsley) were characterised by the highest retention of colour parameters and high retention of chlorophylls, compared to the remaining herbs.
2. In the course of the drying process there was a decrease in the level of green and yellow colour, and also a decrease in the chroma value of most of the herbs. Only in the case of rocket an increase was noted in the yellow colour parameter and chroma value, which indicates the occurrence of a different type of transformations in that material, probably related with structural differences.
3. Very strong negative correlation was demonstrated between the colour parameter *a** and the content of chlorophyll a, and a strong positive correlation between colour parameter *b** and chroma value and the content of chlorophyll a, but only in the case of herbs from the family *Apiaceae*. This indicates that the colour change in lovage and parsley is due mainly to the degradation of chlorophyll a and, additionally, to the occurrence of other reactions changing the colour of the remaining herbs.
4. Microwave-convective drying of herbs proved to a technique permitting the obtainment of dried material of high quality, providing that the process parameters adopted in this study are applied. Additionally, the very short time of the process can be an economic factor and incentive for the application of that method of drying on an industrial scale. Nevertheless, the degree of retention of biologically active compounds and the extent of colour change in the course of drying are related to the herb species, hence the necessity of studying a large array of materials belonging to various taxonomic groups.

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WPŁYW SUSZENIA MIKROFALOWO-KONWEKCYJNEGO NA ZAWARTOŚĆ CHLOROFILU ORAZ BARWĘ ZIÓŁ

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Streszczenie. Celem pracy była analiza zmian zawartości chlorofilu a i b, współczynnika *a** i *b** oraz nasycenia barwy w trakcie suszenia mikrofalowo-konwekcyjnego liści bazylii, lubczyku, mięty ogrodowej, oregano, pietruszki oraz rukoli. Suszenie prowadzono w temperaturze powietrza 40°C, przepływającego poprzecznie do warstwy materiału z prędkością 0,8 $m \cdot s^{-1}$ i przy mocy mikrofal 300 W. Wykazano, że proces suszenia w istotny sposób wpłynął na degradację barwników chlorofilowych, a jej stopień był zróżnicowany dla poszczególnych gatunków ziół. Wysoką retencję chlorofilu a zaobserwowano w lubczyku i bazylii, natomiast chlorofilu b również w mięcie. Jedynie w przypadku lubczyku suszenie nie wpłynęło w sposób istotny na wynik oznaczenia chlorofilu a, natomiast zawartość chlorofilu b nie uległa znaczącym zmianom w bazylii, lubczyku oraz mięcie. W przypadku obu typów barwników chlorofilowych największe straty zaobserwowano w rukoli. Efektem procesu suszenia było istotne zmniejszenie udziału barwy zielonej, żółtej oraz nasycenia wszystkich ziół, za wyjątkiem rukoli. Najmniejsze zmiany dotyczyły surowców z rodziny *Apiaceae* (lubczyk i pietruszka) oraz rukoli (*Brassicaceae*), natomiast mniej stabilną barwą charakteryzowały się zioła z rodziny *Lamiaceae* (oregano, mięta, bazylia). Również przyczyna zmiany barwy uzależniona była od przynależności roślin do grupy systematycznej. Przesunięcie współrzędnych barwy zielonej i żółtej wynikało ze znaczących zmian zawartości chlorofilu a w ziołach należących do *Apiaceae*. Natomiast w przypadku pozostałych ziół degradacja chlorofilu nie była jedynym czynnikiem wpływającym na zmianę barwy. W przypadku wszystkich badanych ziół stwierdzono istnienie silnego związku pomiędzy degradacją chlorofilu a i współczynnikiem barwy *a**.

Słowa kluczowe: zioła, suszenie mikrofalowo-konwekcyjne, chlorofil, barwa