CHANGES OCCURRING IN HONEY DURING RIPENING UNDER CONTROLLED CONDITIONS BASED ON α-AMYLASE ACTIVITY, ACIDITY AND 5-HYDROXYMETHYLFURFURAL CONTENT

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Summary

The aim of the study was to determine the effect of honey ripening under controlled conditions on α-amylase activity as well as on the content of free acids and 5-hydroxymethylfurfural (HMF) in selected honeys. The assessment of α-amylase activity and content of free acids was conducted on 79 samples of unripe honeys which were dehydrated in a specially designed chamber and 69 samples of honeys from the same apiaries but ripened in hives. The content of HMF was determined in 20 samples of unripe dehydrated honeys and in 20 samples of honeys obtained conventionally. During the dehydration process of unripe honeys, a characteristic tendency toward increasing enzymatic activity and acidity was observed. However, changes in these parameters were not statistically significant. Only the acidity of traditionally harvested buckwheat honey was significantly higher. There were no significant differences between the ranges of α-amylase activity and acidity between the dehydrated and in-hive ripened honeys. Statistical analysis revealed that during dehydration the content of 5-hydroxymethylfurfural in buckwheat honey samples increased significantly; nevertheless, the average content of this aldehyde did not exceed 1 mg/kg. A significantly higher content of HMF was also observed in raspberry and buckwheat honeys harvested traditionally, in comparison to the unripe and dehydrated honeys.

Keywords: honey, dehydration, α-amylase, acidity, 5-hydroxymethylfurfural (HMF).

INTRODUCTION

Previous studies confirm the feasibility of performing the process of honey dehydration under controlled conditions (Semkiw et al. 2008a). These studies also revealed that collecting unripe honey as soon as blooming of the forage plant finishes afforded greater possibilities of obtaining pure unifloral honey from each forage source (Semkiw et al., 2008b). Furthermore, the process of ripening various honey varieties under controlled conditions, provides a product with sugar content fulfills the specified quality requirements. During ripening under controlled conditions, the content of reducing sugars increases whereas the content of sucrose decreases slightly, whilst no major changes occur in polysaccharides (Semkiw et al., 2009).

The activity of α-amylase, the content of 5-hydroxymethylofurfural (HMF) and free acids are basic physicochemical parameters of honey quality. The requirements for those parameters are regulated by the Regulation of the Minister of Agriculture and Rural Development of Oct. 3, 2003 (Rozporządzenie Ministra Rolnictwa i Rozwoju Wsi z dnia 03.10.2003). The regulation concern detailed requirements for honey market quality (O. J., No. 181, item 1773 with later amendments).

The document introduces requirements in accordance with Council Directive 2001/110/EC2002 (2002). According to these standards, the minimum diastase value for bee honey is at least 8 Schade units. For honeys with naturally low enzymatic activity (e.g. citrus honeys), the value should be higher than 3 Schade units. On the other hand, the permissible acidity (of free acids) for honey should not exceed 50 meq/kg (mval/kg). The maximum content of HMF in honey may amount to 40 mg/kg. This requirement does not apply to honeys originating from tropical climatic zones, for which the maximum HMF content may reach 80 mg/kg.

The measure of α-amylase activity is the diastase number (DN), expressed in Schade units. One diastase unit is the equivalent of the activity of enzyme present in 1g of honey and capable of hydrolysing 0.01g of starch in 1h at a temperature of 40°C (Bogdanov, 1997). In the past, the determination of enzyme activity was helpful in differentiation between artificial and natural honeys (White, 1975). The measurement of diastase and invertase activity is used as an indicator of honey freshness. The activity of those enzymes diminishes during storage and heating of honey (White et al., 1964; Sancho et al., 1992; Piro et al., 1996; Skowronek et al., 1994). Furthermore, for some varieties of honey, the determination of diastase activity is a differentiating factor while defining their botanical origin. An example of this are honeys from citrus trees, characterised by a low α-amylase activity (Persano Oddo et al., 1990).

Among domestic varietal honeys, buckwheat honey (*Fagopyrum*) showed the highest α-amylase activity. In 55 samples of that honey harvested in four seasons (1960-1964), Rychlik and Fedorowska (1965) recorded α-amylase content at the level of 31.8 to 55.2 Schade units. Buckwheat honey harvested by Rybak (1986) showed α-amylase activity ranging between 41.6-73.8 Schade units. Piekut and Borawska (2000), however, recorded an average of 25.6 Schade units in buckwheat honey.

The average activity of α-amylase in honeydew ranged from 23.5 to 56.4 Schade units (Rybak, 1986; Krauze and Zalewski, 1991). The diastase level of linden honey varied between 22.4 and 55.5 Schade units (Rybak, 1986; Krauze and Zalewski, 1991; Piekut and Borawska, 2000). Heather honey (*Calluna*) had a slightly lower distase value: from 19 to 46 Schade units (Rybak, 1986; Krauze and Zalewski, 1991), red clover honey (*Trifolium pratense*) and mixed nectar and honeydew had: 47-49 Schade units, on average (Rybak, 1986) and multifloral honey had: from 16.5 to 47 Schade units (Rybak, 1986; Piekut and Borawska, 2000). In turn, rape honey and robinia honey had lower diastase levels, ranging from 12.6 to 37.5 and 15.1 to 39.9 Schade units, respectively (Rybak, 1986, Krauze and Zalewski, 1991; Piekut and Borawska, 2000).

The characteristic taste of honey results not only from the sugars present in it, but also from organic acids which greatly enrich and diversify its bouquet depending on variety (Rybak, 1985). Furthermore, the presence of acids in honey protects it from the growth of pathogenic microorganisms (White et al., 1963). The amount of acids in honey is small and ranges from 0.005 to 0.30% of dry matter (Rybak, 1985). Curyło and Rybak (1973) carried out a study on the level of free acids in standardised varietal honeys from District Apiarian Cooperatives. The results showed that deciduous honeydew had ca. 1.5 times higher acidity value in comparison to nectar honey. The acidity (for free acids) of the analysed varietal honeys ranged between 12.2 (rape) and 33.1 (linden) mval/kg. Rybak (1986) carried out an assessment of the acidity of domestic unifrolal honey originating from different apiaries. Rybak found that buckwheat honey (30.2 - 49.8 mval/kg) had the highest values. Multifloral, linden, red clover honeys as well as mixed nectar and honeydew, and deciduous honeydew had similar acidity levels, amounting to over 30 mval/kg. Honeys harvested from spring forages, such as rape -13.9; from orchards-16.5; and robinia-18.8 mval/kg. showed the lowest acidity. Krauze and Zalewski (1991) found similar results of acidity of rape, linden, and heather honeys and of honeydew to those obtained by Rybak (1986).

5-Hydroxymethylfurfural (HMF) is an heterocyclic aldehyde which forms as a result of hexoses dehydration in an acidic environment. This kind of reaction takes also place in honey, since its pH ranges between 3.3 and 4.9 (Gonnet, 1963). The content of HMF in fresh honey is very low, however it increases during storage and heating at a temperature of over 50°C (White et al., 1964; Skowronek et al., 1994; Rybak-Chmielewska and Szczęsna, 1998). An increasing content of HMF together with changes in organoleptic properties of honey occur as honey becomes darker and attains a typical aroma.

The content of HMF in fresh domestic honeys does not exceed 10 mg/kg. According to Curyło (1972) this is a very beneficial asset for it enables the storage of honey at room temperature for a long time (up to 8 years) without an increase in HMF content above the upper limit established by the Polish Standard for honey (PN-88/A-77626 „Miód pszczeli"). According to current regulations, honey should not to be stored more than 3 years. The average content of HMF in nectar varieties of honey ranges from 2.01 (linden honey) to 6.29 mg/kg (buckwheat honey), in mixed nectar and honeydew honey from 1.38 to 4.56 mg/kg, whereas in deciduous honeydew from 2.23 to 5.62 mg/kg (Szczęsna and Rybak-Chmielewska, 1999).

Italian studies confirmed that the content of HMF in fresh and non-adulterated honeys was low (Fini and Sabatini. 1972), whereas Vorwohl (1976) confirmed the natural, higher content of HMF in honeys originating from a tropical climate.

USA scientists analyzed, 481 honeys received from beekeepers and 41 samples obtained directly from honeycombs in order to assess the content of HMF. In the honey received from beekeepers, the content of HMF ranged from 0 to 136 mg/kg. The high HMF contents indicated adulteration of honeys by artificial inverted sugar from acidic hydrolysis of saccharose. The content of 5-hydroxymethylfurfural in the samples collected directly from combs ranged between 0 and 9.2 mg/kg, with an average of 2.7 mg/kg, (White and Siciliano, 1980).

The aim of the present study was to determine the effect of outside hive honey ripening on α-amylase activity and the contents of free acids and 5-hydroxymethylfurfural in selected honeys.

MATERIAL AND METHODS

The studies were conducted in 2004-2006 at the Apiculture Division of the Research Institute of Pomology and Floriculture in Puławy, Poland. The honey samples for the study were collected at apiaries run by the Institute and at commercial apiaries located across the country (Semkiw et al., 2008b).

From each honeybee colony stationed at a defined forage site, two honey samples were collected: one from unripe honey and another from ripe honey. Each sample was ca. 200 ml in volume. A sample of unripe honey was harvested by removing one comb with uncapped honey from the colony while nectar flow from the principal forage was still in progress. The comb was centrifuged in a special cassette protected with a polyethylene bag. In this way it was possible to harvest only honey from one frame during centrifuging. The collected sample was dehydrated. A sample of in-hive ripened honey was collected from the same colony as that of the unripe honey. The sample was collected from capped honeycomb following the same procedure as for the unripe honey (Semkiw et al., 2008a).

The Phadebas method according to Bogdanov et al. (1997) determined the activity of α-amylase expressed as a diastase number (DN). The principle of the method consists in determining the content of starch hydrolysed by α-amylase, contained in a specified amount of honey. Instead of natural starch, a synthetic substitute in the form of Phadebas tablets was used for analyses. The acidity was determined by the potentiometric method (Bogdanov et al., 1997). The method is based on titration of honey solution with 0.1M sodium hydroxide to pH 8.3. The determination was performed in duplicate and the results were expressed as a mean of two parallel determinations differing by less than 0.1 ml of sodium hydroxide used. In the case of greater differences, the analysis was repeated. The result obtained was expressed as mval/kg. The content of 5- hydroxymethylfurfural (HMF) was assessed by the HPLC method according to Bogdanov et al. (1997), with modification of the Bee Products Testing Laboratory. The content of HMF in honey was calculated by using an external standard method.

The results of individual parameters of unripe dehydrated honeys and naturally ripened honeys were assessed using one way analysis of variance by DUNCAN test at a significance level of $\alpha = 0.05$.

The α-amylase activity and the content of free acids were determined for 79 samples of unripe honeys, 79 samples of dehydrated honeys and 69 samples of in-hive ripening honeys. The analysis of 5-hydroxymethylfurfural was conducted in 20 samples of unripe dehydrated honeys and in 20 samples of in-hive ripening honeys.

RESULTS

The enzymatic activity of the analysed unripe honeys, expressed as diastase number in Schade units, ranged from 9.18 to 90.75 Schade units, while for dehydrated honeys it ranged from 9.72 to 96.50 Schade units and for in-hive ripened honey from 9.12 to 90.70 Schade units. Buckwheat honey had the highest value of the diastase level among the unripe and dehydrated honeys. Multifloral and heather honey also had a high α-amylase activity But α-amylase activity was low in linden, raspberry, robinia, borage and rape honey (Tab. 1).

During the dehydration process of unripe honey, there was a characteristic tendency for an increasing enzymatic activity. The values of diastase number (being a measure of this activity) increased from one to several Schade units depending on honey variety. However, the changes were not statistically significant. Honeys with high α-amylase activity, e.g. buckwheat and heather honey, exhibited a larger increase in the diastase number as a result of dehydration in comparison to honeys with low activity of that enzyme, e.g. robinia and raspberry honeys.

There were few differences (statistically insignificant for most varieties) between dehydrated honeys and those obtained traditionally. A higher diastase activity was recorded for dehydrated deciduous honeydew, heather, robinia, linden and buckwheat honey, in comparison to the conventionally harvested honeys. In the case of the former honeys, the differences were considerable. On the other hand, there was a lower α-amylase activity of dehydrated honeys compared to the conventional ones was for mixed nectar and honeydew, rapeseed, raspberry, borage and multifloral honey. The highest diastase activity, both among dehydrated and conventional honeys, was determined in buckwheat honey. High α-amylase activity was also observed for multifloral honey. The diastase number of mixed nectar and honeydew, honeydew and heather honey were on the average, slightly more than 30 Schade units. Among all varieties of conventionally harvested honeys, the lowest α-amylase activity was in linden, robinia and raspberry honeys.

The acidity of the unripe honeys ranged from 10.00 to 44.20 mval/kg, that of the dehydrated honeys from 10.00 to 48.70 mval/kg, while the values recorded for the harvested honey which ripened in hives was from 11.60 to 48.60 mval/kg (Tab. 2).

Out of all the unripe honeys, buckwheat and multifloral honeys had the highest mean acidity. Heather honey had a slightly lower mean acidity. Rape, borage honeys and deciduous honeydew had the lowest mean acidity. The process of dehydration caused a small increase in acidity, however, the differences noticed were not statistically different. For most of the varieties, the differences accounted for ca. 1 mval/kg.

Table 1

A comparison of the changes in α – amylase activity in unifloral honeys proceeding upon artificial dehydration compared to in-hive ripening honeys (Schade units)

Honey variety	Unripe honeys			Dehydrated honeys		In-hive ripening honeys		
	Number of samples (n)	Range min-max	Mean	Range min-max	Mean	Number of samples (n)	Range min-max	Mean
Mixed nectar and honeydew	10	12.41-78.60	**29.90 a**	13.02-69.36	**31.07 a**	10	12.79-69.96	**31.22a**
Deciduous honeydew	5	33.04-43.06	**35.94 a**	34.14-49.06	**38.74 a**	5	24.60-38.92	**32.36 a**
Rape	10	11.16-27.18	**16.20 a**	11.83-27.59	**19.16 a**	9	11.83-42.44	**23.47 a**
Robinia	5	9.26-21.12	**15.81 a**	10.45-23.50	**17.62 a**	4	12.98-18.64	**15.51 a**
Raspberry	10	9.18-25.71	**15.26 a**	9.72-26.00	**15.89 a**	8	10.56-38.74	**16.34 a**
Linden	10	10.35-26.90	**15.01 a**	10.48-27.60	**16.27 a**	8	9.12-28.97	**15.49 a**
Buckwheat	8	49.10-90.75	**68.23 a**	48.89-96.50	**73.00 a**	6	56.03-90.70	**72.01 a**
Borage	5	13.14-21.96	**16.78 a**	15.91-25.30	**19.49 a**	5	15.71-33.10	**21.23 a**
Heather	6	14.83-63.72	**38.85 a**	18.90-70.50	**44.48 a**	4	29.82-53.76	**37.19 a**
Multifloral	10	12.01-60.31	**44.44 a**	12.37-74.88	**53.26 a**	10	19.98-86.65	**53.92 a**

The same letter a in rows denotes, that the differences between means are not statistically significant

Table 2

Changes in acidity in varietal honeys proceeding upon artificial dehydration in respect of changes in values of that parameter in the in-hive ripening honeys (mval/kg)

Honey variety	Unripe honeys			Dehydrated honeys		In-hive ripening honeys		
	Number of samples (n)	Range min-max	Mean	Range min-max	Mean	Number of samples (n)	Range min-max	Mean
Mixed nectar and honeydew	10	10.50-44.20	**21.47 a**	11.00-48.70	**23.99 a**	10	15.70-48.60	**25.10 a**
Deciduous honeydew	5	13.10-18.00	**16.38 a**	14.30-21.10	**16.68 a**	5	17.00-18.30	**17.56 a**
Rape	10	11.30-20.80	**14.42 a**	12.70-21.60	**15.61 a**	9	12.70-20.00	**16.83 a**
Robinia	5	10.00-27.00	**19.20 a**	10.00-29.60	**19.90 a**	4	12.00-21.00	**15.43 a**
Raspberry	10	13.00-27.10	**18.90 a**	12.70-28.00	**18.97 a**	8	15.50-28.00	**19.90 a**
Linden	10	10.80-26.00	**19.76 a**	11.20-26.00	**19.98 a**	8	11.60-26.00	**20.35 a**
Buckwheat	8	26.50-31.20	**29.21 a**	29.30-32.90	**30.74 a**	6	26.70-43.80	**34.50 b**
Borage	5	14.20-17.70	**15.98 a**	15.00-17.80	**16.36 a**	5	15.00-18.00	**16.82 a**
Heather	6	14.50-35.00	**26.93 a**	16.40-36.10	**28.02 a**	4	18.90-35.00	**24.55 a**
Multifloral	10	18.70-43.80	**28.97 a**	20.50-46.00	**32.11 a**	10	21.80-38.60	**27.93 a**

Different letters in rows a and b denote statistically significant differences at $p \leq 0.05$

The increase by 2 mval/ kg on average was noted for mixed nectar and honeydew and by ca. 3 mval/kg for multifloral honey. Raspberry honey had the lowest increase - 0.07 mval/kg.

For most varieties, the conventional way of ripening in hive resulted in further, but not statistically significant, changes in acidity. The exception was buckwheat honey harvested conventionally with a significantly higher acidity recorded in comparison to unripe and dehydrated honeys. An opposite dependency was revealed for robinia, heather and multifloral honeys. The dehydrated honey, along with buckwheat and multifloral conventional honeys, had the highest acidity. Heather honey and mixed nectar and honeydew had slightly lower average values. As in the unripe honeys, rape, borage honeys and deciduous honeydew had the lowest acidity.

The analyses of HMF content were performed for four varietal honeys. The HMF content in all analysed samples ranged from 0.07 to 2.80 mg/kg (Tab. 3). Unripe honeys - 0.63 mg/kg had the lowest average content of HMF-0.63 mg/kg, whereas the highest was for conventionally harvested honeys - 0.98 mg/kg. The average content of HMF in the unripe honeys (from robinia, buckwheat and raspberry) did not exceed 0.5 mg/kg, except for linden honey in which it accounted for 1.46 mg/kg.

The statistical analysis of results revealed that during dehydration the content of 5-hydroxymethylfurfural increased significantly only in the samples of buckwheat honey. However, the average content of that aldehyde did not exceed 1 mg/kg. The content of HMF in the samples of raspberry and buckwheat honey harvested conventionally was statistically higher than in the unripe and dehydrated honeys. There were no statistically significant differences in mean HMF contents between unripe, dehydrated and in-hive ripened honey.

DISCUSSION

The diastase number, being a measure of α-activity, exceeded 8 units in all analysed samples of honey before dehydration. This means that it met the requirements stipulated by standards (Council Directive 2002, Codex Alimentarius Commission 2001, the Regulation of the Ministry of Agriculture and Rural Development considering the market quality of honey 2003). The process of dehydration of unripe honeys increased the diastase level by 1 or more Schade units. However, the changes were not statistically significant. These results confirm observations of Klemarewski (1976) who found that the value of the diastase activity increased during honey ripening and its moisture content decreased based on an analysis of rape and robinia honeys. On the other hand, this author

Table 3

Changes in the contents of 5-hydroxymethylfurfural in varietal honeys proceeding upon artificial dehydration compared to in-hive ripening honeys (mg/kg)

Honey variety	Unripe honeys			Dehydrated honeys		In-hive ripening honeys	
	Number of samples (n)	Range min-max	Mean	Range min-max	Mean	Range min-max	Mean
Robinia	5	0.07 - 0.70	**0.45a**	0.21 - 0.94	**0.37 a**	0.30 - 0.98	**0.54 a**
Raspberry	5	0.07 - 0.70	**0.35 a**	0.25 - 0.80	**0.53 a**	0.30 - 1.00	**0.80 b**
Linden	5	0.10 - 1.90	**1.28 a**	0.30 - 1.90	**1.26 a**	0.10 - 2.80	**1.03 a**
Buckwheat	5	0.10 - 0.70	**0.42 a**	0.70 - 1.10	**0.90 b**	0.80 - 2.40	**1.46 c**
	20	0.07 - 1.90	**0.63a**	0.21 - 1.90	**0.76a**	0.10 - 2.80	**0.98a**

Different letters in rows a and b denote statistically significant differences at $p \leq 0.05$

recorded an opposite dependency for buckwheat honey. In that study, the diastase number was the highest in the freshly collected nectar (73.4 Schade units), whereas in the uncapped honey it amounted to ca. 65.9; and was the lowest in ripe honey at ca. 64.2 Schade units, on average. Such dependencies were, however, not confirmed in our experiment, because during the ripening of buckwheat honey under controlled conditions the diastase number increased by ca. 5 Schade units.

In the reported study, the highest diastase activity was for buckwheat honey, both the unripe, dehydrated and conventional versions. The values were comparable to results obtained by Rybak (1986), however, they were much higher than those reported by Rychlik and Fedorowska (1965) and also by Piekut and Borawska (2000). High α-amylase activity was also recorded in heather and multifloral honeys as well as deciduous honeydew. The results obtained in the study were in accordance to those presented by Rybak (1986). In the case of heather honey, the α-amylase activity was higher than that reported by Krauze and Zalewski (1991) The diastase activity of multifloral honey was higher than that determined by Piekut and Borawska (2000). The differences could result from the fact that the authors were analysing honeys obtained from the market which might have been heated in order to solubilize, which could decrease the activity of α-amylase.

The low diastase number of linden honey reported in the present study differed substantially from the results obtained by Rybak (1986), Krauze and Zalewski (1991) as well as Piekut and Borawska (2000). However, the value was close to that recorded by Persano Oddo et al. (1990 and 1995). The reason for the low diastase activity in the analysed linden honey was probably the intensive nectar-bearing of linden in the course of the study. A high daily increase of nectar into hives prevented bees from enriching it with the sufficient quantity of enzymes.

The permissible acidity of honey should not exceed 50 meq/kg (mval/kg). The analysed varietal honeys, before and after dehydration, as well as the conventionally harvested honeys met that requirement. It was also recorded that honeys from spring and early summer forages were characterised by lower acidity compared to those from later forages, which was in accordance with the observations made by Curyło and Rybak (1973), Rybak (1986) as well as Krauze and Zalewski (1991). The acidity of rape honey determined by Rybak (1986) was 13.9 mval/kg, on average, whereas in the current study it amounted to ca. 16 mval/ kg. The acidity of linden honey was ca. slightly higher than 30 mval/kg, as reported by Curyło and Rybak (1973), Rybak (1986) as well as Krauze and Zalewski (1991), whereas in the current study it amounted to ca. 20 mval/kg.

An important criterion of the quality assessment of dehydrated honeys was the content of 5-hydroxymethylfurfural. The content of that compound increases in processed honeys, especially during heating. The analysed honeys fully satisfied the requirements concerning the maximum permissible content of HMF of both Council Directive (2002) and Codex Alimentarius Commission (2001). Among all samples, the maximum content of HMF was determined in linden honey and it amounted to 2.8 mg/kg, whereas the acceptable level of this aldehyde in honey is stipulated at 40 mg/kg. The other samples were characterised by a very low HMF content, not exceeding 1.5 mg/kg, on average. The obtained results differ from data reported in domestic studies to date. According to Szczęsna and Rybak-Chmielewska (1999), the content of HMF in domestic nectar honeys ranged from 2.01 (average of linden honeys) to 6.29 mg/kg (average of buckwheat honeys). The results of the current studies were, presumably, influenced by the fact that the honeys were analysed directly after harvesting.

The results of this and the authors'

previous studies (Semkiw et al., 2008a, Semkiw et al., 2008b; Semkiw et al., 2009) confirm that the process of artificial ripening, carried out according to the suggested procedure, does not bear negative affects on the quality of honey. Furthermore, these studies encouraged the Department of Apiary Technology, Division of Apiculture, to construct an appliance to enable dehydration of honey on a commercial scale.

CONCLUSIONS

1. The ripening process of different varieties of honey under controlled conditions enable obtaining a product with α-amylase activity, acidity and a content of 5-hydroxymethylfurfural, satisfying the specified quality requirements.
2. The ripening process conducted following the presented procedure does not elicit any substantial changes in the parameters which characterise the physicochemical properties of honey compared to the same varieties of in-hive ripened honeys.

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ZMIANY ZACHODZĄCE W MIODZIE PODCZAS DOJRZEWANIA W KONTROLOWANYCH WARUNKACH NA PODSTAWIE AKTYWNOŚCI α-AMYLAZY, KWASOWOŚCI I ZAWARTOŚCI 5-HYDROKSYMETYLOFURFURALU

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S t r e s z c z e n i e

Celem badań było określenie wpływu dojrzewania miodu w kontrolowanych warunkach na aktywność α-amylazy oraz zawartość wolnych kwasów i 5-hydroksymetylofurfuralu. Badania aktywności α-amylazy oraz zawartości wolnych kwasów przeprowadzono na 79 próbkach miodów niedojrzałych, które poddano odwadnianiu w przygotowanej do tego celu komorze oraz na 69 próbkach miodów z tych samych pasiek ale dojrzewających w ulach. Zawartość HMF oznaczano w 20 próbkach miodów niedojrzałych i odwadnianych oraz w 20 próbkach miodów pozyskanych konwencjonalnie. Podczas procesu odwadniania niedojrzałych miodów wystąpiła charakterystyczna tendencja zwiększania się ich aktywności enzymatycznej i kwasowości, jednakże zmiany tych parametrów nie były istotne statystycznie. Tylko w miodzie gryczanym pozyskanym w sposób tradycyjny oznaczono istotnie wyższą kwasowość. Nie stwierdzono natomiast istotnych różnic w zakresie aktywności α-amylazy oraz kwasowości pomiędzy miodami odwadnianymi, a dojrzewającymi w ulach. Analiza statystyczna wykazała, że podczas odwadniania istotnie zwiększyła się zawartość 5-hydroksymetylofurfuralu w próbkach miodu gryczanego, aczkolwiek średnia zawartość tego aldehydu nie przekroczyła 1 mg/kg. Stwierdzono także istotnie wyższą zawartość HMF w próbkach miodu malinowego i gryczanego pozyskanych w sposób konwencjonalny, w porównaniu do miodów niedojrzałych i odwadnianych.

Słowa kluczowe: miód, dehydratacja, α-amylaza, kwasowość, 5-hydroksymetylofurfural (HMF).