www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2022; 11(7): 917-923

www.thepharmajournal.com
Received: 09-04-2022
Accepted: 28-06-2022

Anina Jose
Department of Food Technology and nutrition, School of agriculture, Lovely Professional University, Punjab, India

Nirmal P
Department of Food Technology and nutrition, School of agriculture, Lovely Professional University, Punjab, India

Corresponding Author:
Anina Jose
Department of Food Technology and nutrition, School of agriculture, Lovely Professional University, Punjab, India

Development and Evaluation of Creamed honey added with glucose & seed crystal as nuclei

Anina Jose and Nirmal P

Abstract
The present work aimed to develop the creamed honey with the addition of glucose and seed crystal as nuclei. The effects of glucose and seed crystal on its microstructure were determined. The physico-chemical analysis was analyzed before the addition of glucose (2%, 4% and 6%) and seed crystal (5%, 7.5% and 10%) and the sample added with glucose and seed crystal were subjected to homogenization at 180 rpm, 360 rpm and 540 rpm for 10, 15 and 20 minutes and stored at 5ºC. Microstructural analysis and water activity was done on alternative days from day 0 to day 10. The influence of water activity and homogenization were also discussed. It could be observed that the samples added with 2% glucose, 540 rpm for 15 minutes, 4% glucose, 540 rpm for 15 minutes, 6% glucose, 360 rpm for 20 minutes and 6% glucose, 540 rpm for 15 minutes, 5% seed crystal, 360 rpm for 20 minutes, 7.5% seed crystal, 360 rpm for 15 minutes, 7.5% seed crystal, 540 rpm for 10 minutes and 10% seed crystal, 360 rpm for 20 minutes have more small crystals with uniform distribution with an increase in water activity.

Keywords: Crystallization, glucose, seed crystal, water activity, homogenization

1. Introduction
Honey is a viscous liquid produced by various species of honeybees having sweetness as a taste (Farooq *et al.* 2020; Alvarez-Suarez *et al.* 2014) [16, 5]. The sweetness is because of the presence of carbohydrates in honey such as fructose, glucose and sucrose. Proteins, amino acids, vitamins, minerals, flavonoids, enzymes, organic acids, and phenolic compounds are among the minor ingredients (Gündodu *et al.* 2019) [20].
Different physical properties of honey, such as colour, enzyme activity, pH, ash content, and even sweetness or flavour, vary depending on bee type, impurities in honey, and geographical origin. Honey's colour ranges from pale yellow to darker red, depending on the plant source. Honey's darkening colour is most commonly caused by temperature changes (Khan *et al.* 2018; Lawal *et al.* 2009) [23, 24].
Fruit sugar and glucose are the two major sugars content in honey of 30-44% and 25-40% respectively. Due to the lower solubility of glucose in water, it tends to crystallize first. But crystallized honey gives the impression that it got adulterated, but crystallization is an assurance that it is pure, natural in fact. So that, crystallization is a natural occurrence in honey (Amariei *et al.* 2020) [6]. Honey crystallizes at different rate depending on the honey type. Different types of honey crystallize at different rates. Honey with high F/g ratio will crystallize quite slowly, while honey with a lower F/g ratio crystallizes more rapidly (Hamdan, 2010) [22].
The crystallization process usually takes a long time, which can cause large crystals to form and uneven distribution (Zhang *et al.* 2015) [33]. When undesirable crystallization occurs during storage, the honey forms a crystalline phase on the bottom and a dark coloured liquid phase on top, making the honey unappealing to consumers. When crystallization takes place in a controlled manner, however, products like creamed honey can be made (Costa *et al.* 2015) [10]. The liquid form of honey has some issues, such as natural crystallization at room temperature, resulting in granulation. However, when it crystallizes uncontrollably, it has a coarse, gritty texture as a result of the crystallized honey (Hamdan, 2010) [22]. This type of honey is also less spreadable. As a result, transforming liquid honey to creamed honey is the appropriate solution to such issues (Abou-Shaara & Abd Elhamid, 2017) [1].
Creamed honey is made by using a controlled crystallization process to turn honey into a spreadable consistency similar to butter (Hamdan, 2010) [22]. Creamed honey production is influenced by a number of parameters, including nucleus type, honey type, storage temperature, and other honey-specific features (Abou-Shaara & Abd Elhamid, 2017) [1].

The addition of seed to liquid honey accelerates the crystallization process, resulting in a more appealing and stable product (Dettori *et al.* 2018) [11].
Out of all these factors viscosity, supersaturation, water content, sugar composition have been studied extensively by different authors. But the effect of addition of glucose, homogenization has not studied to a great extent. Therefore this work is carried out to develop creamed honey and looking at the effect of water activity in crystallization process with the addition of glucose and seed crystal as nuclei.

2. Materials and Methods

2.1 Raw materials

Honey samples were obtained from the bee farm of Lovely Professional University, Punjab and the chemicals used were of analytical grade which was procured from standard manufacturers.

2.2 Physico- chemical analysis of honey

2.2.1 Moisture

At a constant temperature near to 20 ^{0}C, the moisture content of honey samples was evaluated using Abbe's refractometer (Bogdanov, 2009) [7]. Temperature corrections to change refractive index; If Temperature > 20 ^{0}C - add 0.00023 per ^{0}C, or Temperature < 20 ^{0}C - substract 0.00023 per ^{0}C

2.2.2 Ash

The amount of ash in honey is a quality indicator. Ash was performed in muffle furnace at 550^{0}C for 5 hours following the method by (FSSAI, 2015) [18].

$$\text{Ash\%} = \frac{\text{Weight of ash}}{\text{Weight of sample}} \times 100$$

2.2.3 Water activity

AQUA LAB Water Activity Meter was used to monitor water activity (Tappi *et al.* 2019) [31].

2.2.4 Acidity & pH

Acidity was performed by titrating against NaOH, expressed in milliequivalents per kilogram honey (FSSAI, 2015) [18]. In a 100ml conical flask, 10 gm of the sample is dissolved in 75 ml of distilled water. Make a thorough mix. Using 4-6 drops of phenolpthalein indicator titrate against standard sodium hydroxide solution until pink colour remains for 10 seconds.
Determine the blank on water and the indicator to determine the volume of sodium hydroxide solution used.

$$\text{Acidity (\%)} = \frac{0.23 \times V}{M}$$

Where,
V = corrected volume of 0.05 N NaOH
M = weight in gm of the sample
pH was measured directly on the honey sample using a pH-meter (Bogdanov, 2009) [7]

2.2.5 Hydroxymethylfurfural

The UV absorbance of HMF at 550nm was used to calculate the HMF content (Bogdanov, 2009) [7]. 2 mL honey solution is pipette out into each of the two test tubes, followed by 5 mL p-toluidine solution. Pipette 1 ml water into one test tube and 1 ml Barbituric acid solution into the other, then shake both liquids. The blank is the one with the water. The reagents should be added without pausing and should take roughly 1-2 minutes to complete. The sample's absorbance is measured against a blank at 550 nm in a 1 cm cell as soon as the highest value is obtained.

$$\text{Mg/100g HMF} = \frac{\text{Absorbance} \times 19.2}{\text{thickness of layer}}$$

2.2.6 Sucrose

The capacity of the reducing sugars to decrease the copper in Fehling's solution was used to determine sucrose (FSSAI, 2015) [18].
Sucrose% by mass = [(approximate total reducing sugar% (after inversion)) – (approximate total reducing sugar% (before inversion))] × 0.95

2.2.7 Glucose & Fructose/Glucose ratio

In a weak alkaline medium, glucose percent was calculated iodimetrically, and the value was deducted from reducing sugars% to arrive at fructose% and F/g ratio (FSSAI, 2015) [18]..
Fill a 250 ml volumetric flask halfway with 2 g of sample. Transfer a 25-mL aliquot to a 250-mL iodine flask and thoroughly mix. 50 ml 0.1N iodine, 50 ml 0.2N sodium carbonate, and 50 ml 0.2N sodium bicarbonate solution. Allow for 2 hours of darkness. After acidifying with 12 mL of 25% H_2SO_4, titrate using starch as an indicator with standard sodium thiosulphate. At the same time, carry out the blank. Subtract the sample titre value from the blank titre value.

$$\text{Glucose\%} = \frac{\text{Normality of Na2S2O3 x dilution x (B} - \text{S) x 0.009005 x 100}}{\text{0.1N x weight of sample}}$$

Fructose% = Reducing sugars% - glucose%

$$\text{F/g ratio} = \frac{Fructose\%}{Glucose\%}$$

2.3 Functional analysis

2.3.1 DPPH activity

DPPH Free Radical Scavenging Activity was assayed by (Shah & Modi, 2015) [27] at 517nm using a spectrophotometer. 2 µl dpph was added to 200 µl of sample in a test tube. At 517 nm, the absorbance was measured.

$$\text{Dpph inhibition\%} = \frac{\text{Absorbance of control} - \text{absorbance of sample}}{\text{absorbance of control}} \times 100$$

2.3.2 Total phenolic content

Folin–Ciocalteu technique was used to assess total phenolic content (TPC) followed by (Pauliuc *et al.* 2020) [26] at 765nm using a UV-VIS spectrophotometer. 1 ml of sample is taken and add 0.5 ml FCR reagent and kept for 10 minutes. Then add 2 ml sodium carbonate and measured the absorbance at 765nm. TPC was measured in milligram of gallic acid per gram of dry weight (mg GAE). A gallic acid solution was used to generate a calibration curve.

2.3.3 Total flavonoid content

The technique was used to determine total flavonoid content (TFC) followed by (Pauliuc *et al.* 2020) [26] at 510nm using a spectrophotometer. Take 20 µl sample and make up to 100ml. Then take out 250 µl from the made up solution into one test tube. 100 µl sodium nitrite and kept for 5 min and add 100 µl 10% Aluminium chloride along with 1ml distilled water, kept

for 6 min and add 1000 µl 5% sodium hydroxide. Absorbance measured at 510 nm. Quercetin was used to generate a calibration curve.

2.4 Preparation of creamed honey

2.4.1 Intermediate batch for seed crystals

Previously crystallized honey ⟶ Ground to get fineness ⟶ Add fresh honey (10-20 times of its weight) ⟶ Mix thoroughly ⟶ Store at 5ºC

2.4.2 Creamed honey

Before adding the seed, the honey sample was maintained at room temperature. For seeding, glucose monohydrate and seed crystal were used.

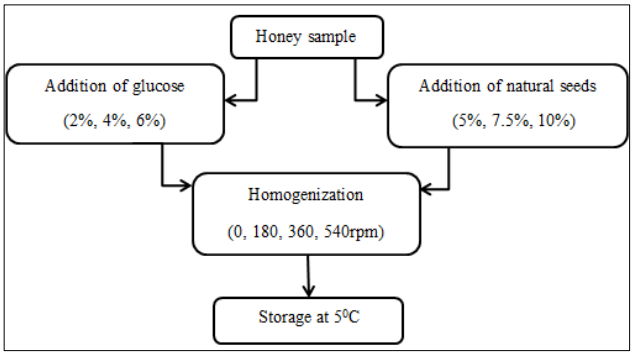

Fig 1: Preparation of creamed honey

20 ml of liquid honey was poured into thirty four 100 ml beakers each. Three levels of glucose were added at 2.0, 6.0, and 8.0% (w/w) in 20 g of liquid honey and three levels of seed crystal were added at 5.0, 7.5, and 10% (w/w) in 20 g of liquid honey. Then it underwent to homogenize at 180, 360, and 540 rpm for 10, 15, and 20 minutes at room temperature. After homogenizing the samples were shifted to glass bottles and stored at 5^0C for crystallization process. The analysis was done for 10 days on alternative days.

2.5 Microstructure analysis

2.5.1 Light microscope

Samples were microstructurally analyzed by light microscope at room temperature with a magnification of 10x (Suriwong *et al.* 2020; costa *et al.* 2015; Conforti *et al.* 2007) [30, 10, 9].

3. Result and Discussion

3.1 Physico-chemical analysis

The honey sample was taken for the physico-chemical analysis and their results are shown in Table 1.

3.1.1 Ash content in honey

Honey's ash content serves as a quality indication for geographic origin. Honey has a low ash content, which is determined by the chemical components of nectar, which varies depending on the botanical sources used in honey production. It can range from 0.02 to 1.0 percent, with a maximum of 0.6 percent legally authorized for honey from floral sources (El Sohaimy *et al.* 2015; Felsner *et al.* 2004) [13, 17]. The sample's ash content was within the codex permitted range (0.6-1.2 g/100 g), that is 0.393% of ash content was present in honey (Codex Alimentarius, 2001) [8].

3.1.2 Moisture content in honey

Moisture is one of the most important characteristics of honey. Honey's resistance to fermentation and granulation is determined by the quantity of water it contains. Honey with high water content ferments quickly (Singh *et al.* 2018) [28]. The water present in honey affects physical qualities such as viscosity, rheological properties, crystallization, and so on. Honey has a longer shelf life with lower moisture limits (20%) (Codex Alimentarius, 2001) [8] And the moisture content is 17.6%, which is within this range. Temperature and relative humidity of the atmosphere, however, have an impact.

3.1.3 Sugars in honey

Honey's sugar content is used to identify the botanical origin of samples. Glucose, fructose, and sucrose are the three main sugars contained in honey, but their amounts vary. Glucose and sucrose percentage were 30.45% and 5.217% respectively. Therefore, fructose was calculated by subtracting reducing sugars percent and glucose given 34.75%. The results indicated that fructose and glucose are the most prevalent sugars in honey samples.

3.1.4 F/G ratio and G/W ratio

The F/G ratio is the most important factor in honey crystallization time. Venir *et al.* (2010) [32] found that samples with a ratio more than 1.58 had no tendency to crystallize, while those with a value greater than 1.33 crystallize slowly (Dobre *et al.* 2012) [12]. However, samples with a ratio slightly over 1 crystallize quickly (Escuredo *et al.* 2014; Venir *et al.* 2010; Smanalieva and Senge, 2009) [15, 32, 29]. Escuredo *et al.* (2014) [15] discovered that honey with a lower F/G ratio and less water content crystallizes faster. The result obtained for F/G ratio was 1.14. So this sample was categorized under fast crystallization. Honey's F/G ratio is also influenced by its botanical source.

The G/W ratio influences the rate of glucose crystallization. Crystallization occurs faster when glucose levels are high and water levels are low. Honey samples with a G/w ratio of less than 1.7 crystallize slowly, but those with a ratio of greater than 2.0 crystallize fast and entirely (Escuredo *et al.* 2014; Dobre *et al.* 2012) [15, 12]. The sample included 17.6 percent water, and the glucose/water ratio was 1.80. As a result, the sample's G/W ratio was close to 2, and it crystallized fast.

3.1.5 Water activity (a_w)

Honey's water activity varies from 0.5 to 0.65. Water activity of the sample was around 0.550 and less susceptible to fermentation as their water activity is about ≤ 0.61. 0.62 is the minimum water activity required for osmotolerant yeast activity (Abramovic *et al.* 2008) [2].

3.1.6 pH and acidity

The honey sample's pH values were analyzed, and the results revealed that the honey was acidic (pH 4.08) and within the usual range (pH 3.40–6.10), suggesting that it was fresh (Codex Alimentarius, 2001) [8].

Honey's overall acidity is influenced by the presence of organic acids, particularly gluconic acid and its ester, as well as inorganic ions such as phosphate and chloride ions. The sample's acidity was 0.0480 percent. The lack of undesirable fermentation is shown by the low total acidity value (Ahmida *et al.* 2012) [3]. Honey's high acidity is caused by the fermentation of carbohydrates in the honey into organic acid, which gives honey its flavour and resistance against microbial

degradation (Bogdanov, 2009) [7]. It might also indicate that the honey samples contain a lot of minerals (El-Metwally, 2015; Mohammed and Babiker, 2009) [14, 25].

3.1.7 Hydroxymethylfurfural (HMF)

Dehydration of hexose sugars in acid media results in the formation of HMF. The HMF content in honey is affected by temperature, heating time during processing, storage conditions, honey product ageing, and flora sources. HMF indicates if honey is fresh, whether it has been overheated, and how long it has been stored in bad circumstances (Ahmida *et al.* 2012) [3]. The HMF of the sample was 17.664 mg/kg which comes within the range of IHC about 3.7-22.0. This indicates the sample is fresh, not overheated and stored in good conditions.

Table 1: Physico-chemical analysis of honey

Honey Property	Results
Ash (%)	0.393±0.003
Moisture (%)	17.6±0.115
pH	4.08±0.034
Acidity (%)	0.0480±0.001
Sucrose (%)	5.217±0.034
Glucose (%)	30.45±0.220
Fructose (%)	34.75±0.402
F/g ratio	1.14± 0.011
G/w ratio	1.80± 0.138
Water activity	0.550±0.001
HMF (mg/kg)	17.664±0.267

Three replicates (n = 3) were used to calculate the means ± standard deviation

Sample added with glucose which was kept in refrigerator at 5^0C was analyzed at alternative days from day 0 to day 10. Visually on day 0, (fig 2) sample with 2% glucose homogenized at 540 rpm for 15 minutes, 4% glucose homogenized at 360 rpm for 15 minutes and 6% glucose homogenized at 360 rpm for 15 minutes, 540 rpm for 20, 15 minutes showed creamy colour. Other samples still the same colour of honey. On day 10, (fig 3) all the samples showed creamy colour except 4% glucose homogenized with 180 rpm for 10 minutes and sample with 6% glucose homogenized with 180 rpm for 15 minutes showed separation of honey and crystals.

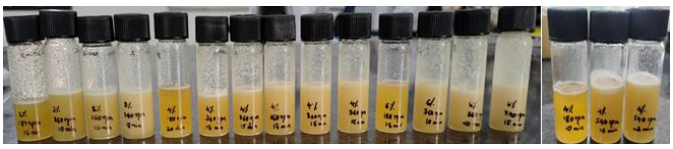

Fig 2: Day 0 - sample added with glucose

Fig 3: Day 10 - sample added with glucose

On day 0, (figure 4) sample added with 7.5% seed crystal homogenized at 540 rpm for 20 minutes and the sample added with 10% seed crystal homogenized at 540 rpm at 15 minutes showed a slight creamy colour. Other samples have a slight change in colour from the actual honey colour.

On day 10, (figure 5) all samples have a light creamy colour than previous days except sample added with 7.5% seed crystal homogenized at 540 rpm for 20 minutes and the sample added with 10% seed crystal homogenized at 540 rpm at 15 minutes showed more creamy colour.

Fig 4: Day 0 - sample added with seed crystal

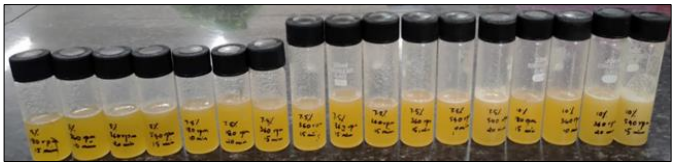

Fig 5: Day 10 - sample added with seed crystal

3.2 Functional analysis of honey

3.2.1 Total phenolic content

TPC (Total Phenol Content) is a quick and easy approach to determine how much phenol is in honey. TPC was said to be sensitive enough to calculate total phenol levels in honey samples (Al *et al.* 2009) [4]. Total phenolic, total flavonoid, and DPPH methods have been used to indicate that these compounds are primarily responsible for honey's antioxidant potential. The total phenolic content (TPC) of honey samples was found to be 93.5±0.063 mg GAE/100 g honey on average. The observed value is consistent with Gul & Pehlivan (2018) [19].

3.2.2 Total flavonoid content

The mean value of triplicate tests was used to calculate total flavonoid content (TFC), which was represented as milligram of quercetin equivalent (QAE) per g of honey. The flavonoids content was 53.8±0.010 mg QAE/ 100g of honey. Hagr *et al.* (2017) [21]'s study also shows flavonoid content of 55.14 ± 1.09 mg QAE/ 100g which is similar to the current study.

3.2.3 DPPH Activity

The DPPH activity of the honey samples differs significantly (Gül & Pehlivan, 2018) [19]. The picture depicts an antioxidant test based on the decrease of the (DPPH) standard curve. The absorbance at 517nm as a function of ascorbic acid concentration was used to determine the degree of inhibition in (percent). The antioxidant activity observed in the honey is 55.5±0.0560% which agrees with the values obtained by Harg *et al.* (2017) [21].

Table 2: TPC, TFC, DPPH analysis of honey

Functional properties	Results
Total phenolic content(TPC) (mg/100g GAF)	93.5±0.063
Total flavonoid content(TFC) (mg/100g QE)	53.8±0.010
DPPH Activity(%)	55.5±0.0560

3.3 Water Activity of creamed honey

Table 3 and table 4 shows the water activity of sample added with glucose and sample added with seed crystal respectively. The Initial water activity of honey was 0.550. After the preparation of creamed honey with the addition of two

different nuclei (glucose and seed crystal) under varying conditions such as homogenizing rate (180, 360, and 540 rpm), and time (10, 15, and 20 minutes), there was a slight change in water activity. In sample added with glucose, water activity varied in between 0.551-0.593 and in sample added with seed crystal, water activity varied in between 0.523-0.546. According to Abramovic *et al*. (2008) [2], Water activity increases during crystallization, although the moisture content remains constant. Crystallization reduces the concentration of glucose in the liquid phase, causing a rise in the honey's water activity, so that the water activity was increased from the initial value. This indicates the presence of crystals in the sample. a_w values grew progressively as the amount of crystalline glucose increased. The observed values were agreed with International honey commission (IHC).

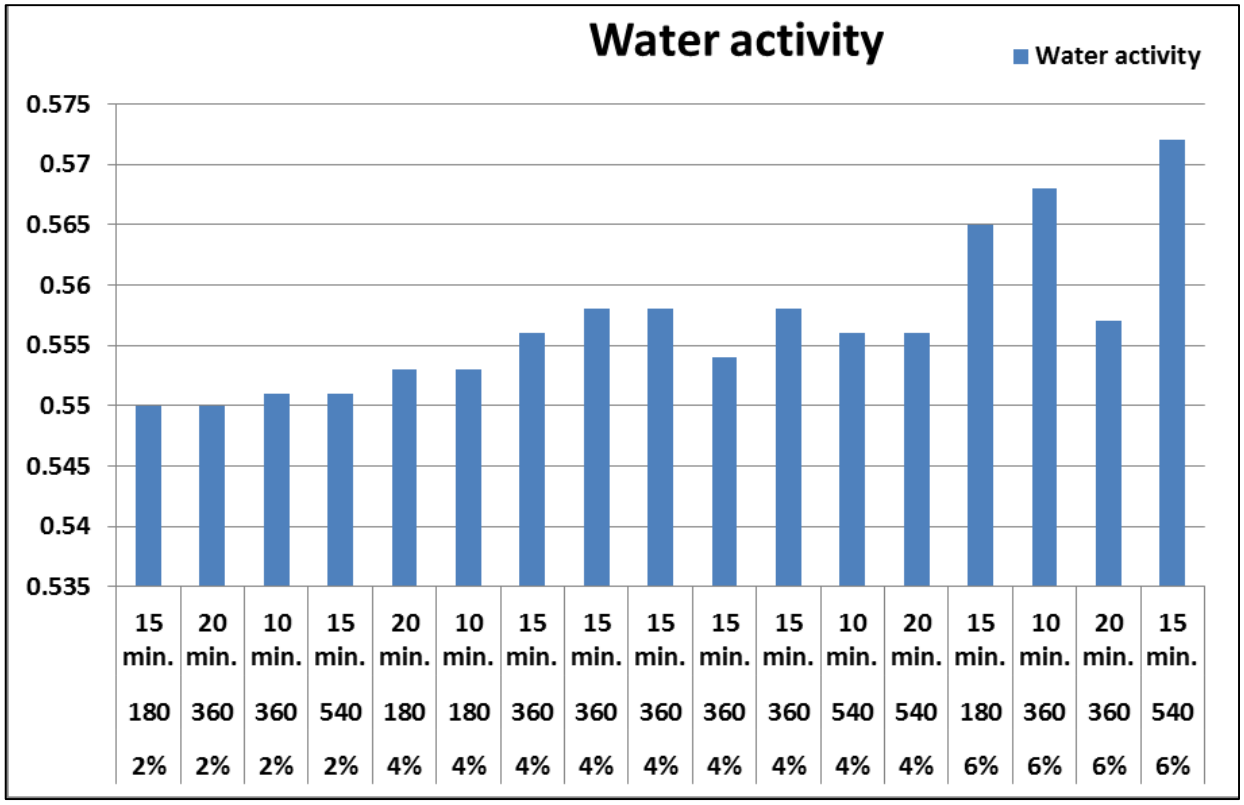

Fig 6: Water activity of creamed honey (glucose)

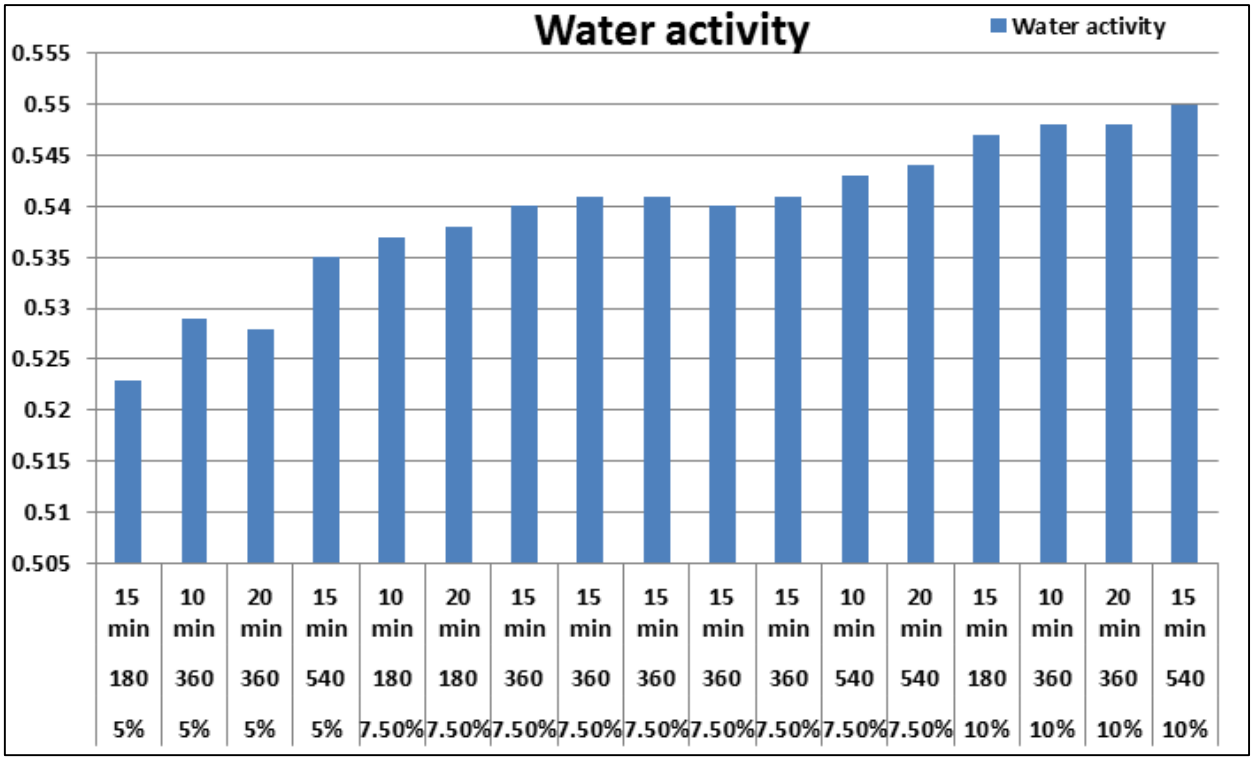

Fig 7: Water activity of creamed honey (Seed crystal)

Table 3: Water activity of sample added with glucose

Glucose%	RPM	Time	Water activity
2%	180 rpm	15 min.	0.550±0.006
2%	360 rpm	20 min.	0.550±0.005
2%	360 rpm	10 min.	0.551±0.007
2%	540 rpm	15 min.	0.551±0.004
4%	180 rpm	20 min.	0.553±0.005
4%	180 rpm	10 min.	0.553±0.004
4%	360 rpm	15 min.	0.556±0.001
4%	360 rpm	15 min.	0.558±0.001
4%	360 rpm	15 min.	0.558±0.002
4%	360 rpm	15 min.	0.554±0.001
4%	360 rpm	15 min.	0.558±0.003
4%	540 rpm	10 min.	0.556±0.006
4%	540 rpm	20 min.	0.556±0.004
6%	180 rpm	15 min.	0.565±0.003
6%	360 rpm	10 min.	0.568±0.002
6%	360 rpm	20 min.	0.557±0.002
6%	540 rpm	15 min.	0.572±0.004

Table 4: Water activity of sample added with seed crystal

Seed crystal%	RPM	Time	Water activity
5%	180 rpm	15 min.	0.523±0.001
5%	360 rpm	10 min.	0.529±0.001
5%	360 rpm	20 min.	0.528±0.001
5%	540 rpm	15 min.	0.535±0.001
7.5%	180 rpm	10 min.	0.537±0.001
7.5%	180 rpm	20 min.	0.538±0.001
7.5%	360 rpm	15 min.	0.54±0.001
7.5%	360 rpm	15 min.	0.541±0.001
7.5%	360 rpm	15 min.	0.541±0.0009
7.5%	360 rpm	15 min.	0.540±0.001
7.5%	360 rpm	15 min.	0.541±0.001
7.5%	540 rpm	10 min.	0.543±0.002
7.5%	540 rpm	20 min.	0.544±0.005
10%	180 rpm	15 min.	0.547±0.003
10%	360 rpm	10 min.	0.548±0.005
10%	360 rpm	20 min.	0.548±0.004
10%	540 rpm	15 min.	0.550±0.001

3.4 Microstructure analysis

3.4.1 Sample added with glucose

The samples were microscopically analyzed from day 0 to day 10 (alternative days) of the sample added with glucose. On day 0, in most of the samples showed presence of small crystals and on day 2, 4, 6 started the formation of large elongated crystals in the sample with a larger crystal growth in all the samples. On day 10, there was a reduction in the size of the crystals in sample with 2% glucose, 540rpm for 15 minutes, 4% glucose, 540 rpm for 15 minutes, 6% glucose, 360rpm for 20 minutes and 6% glucose, 540rpm for 15 minutes.

3.4.2 Sample added with seed crystal

The samples were microscopically analyzed from day 0 to day 10 (alternative days) of the sample added with seed crystal. On day 0, in most of the samples showed presence of small crystals and on day 2, 4, 6 started forming needle like shapes and distributed evenly in most of the samples. On day 10, uniform fine needle shaped crystals were distributed throughout the sample with 5% seed crystal, 360 rpm for 20 minutes, 7.5% seed crystal, 360 rpm for 15 minutes, 7.5% seed crystal, 540 rpm for 10 minutes and 10% seed crystal, 360 rpm for 20 minutes).

4. Conclusion

The addition of glucose & seed crystals to honey and water activity influenced the crystallization rate, resulted in crystal formation during crystallization. The small crystals' microstructural examination revealed that they were in hexa-pentogonal shape in the sample with 2% glucose, 540rpm for 15 minutes, 4% glucose, 540 rpm for 15 minutes, 6% glucose, 360rpm for 20 minutes and 6% glucose, 540rpm for 15 minutes and needle like shape in the sample with 5% seed crystal, 360 rpm for 20 minutes, 7.5% seed crystal, 360 rpm for 15 minutes, 7.5% seed crystal, 540 rpm for 10 minutes and 10% seed crystal, 360 rpm for 20 minutes with an increased homogenization speed and water activity than fresh honey.

5. Reference

1. Abou-Shaara, Hossam Elhamid, Abeer. Scientific Note: Effects of Nuclei Type on Characteristics of Creamed Honey. Journal of Apiculture. 2017;32:59-62. 10.17519/apiculture.2017.04.32.1.59.
2. Abramovič H, Jamnik M, Burkan L, Kač M. Water activity and water content in Slovenian honeys. Food Control. 2008 Nov 1;19(11):1086-90.
3. Ahmida NH, Elagori M, Agha A, Elwerfali S, Ahmida MH. Physicochemical, heavy metals and phenolic compounds analysis of Libyan honey samples collected from Benghazi during 2009-10.
4. Al ML, Daniel D, Moise A, Bobis O, Laslo L, Bogdanov S. Physico-chemical and bioactive properties of different floral origin honeys from Romania. Food Chemistry. 2009;112(4):863-867.
5. Alvarez-Suarez JM, Gasparrini M, Forbes-Hernández TY, Mazzoni L, Giampieri F. The composition and biological activity of honey: a focus on Manuka honey. Foods. 2014 Jul 21;3(3):420-32.
6. Amariei S, Norocel L, Scripcă LA. An innovative method for preventing honey crystallization. Innovative Food Science & Emerging Technologies. 2020 Dec 1;66:102481.
7. Bogdanov, S. Harmonized methods of the International Honey Commission. Retrieved April 26, 2021, from https://www.ihc-platform.net/ihcmethods2009.pdf.
8. Codex Alimentarius. Revised Codex Standard for Honey, Standards and Standard Methods Codex Alimentarius Commission FAO/OMS 2001.
9. Conforti PA, Lupano CE, Malacalza NH, Arias V, Castells CB. Crystallization of honey at−20 C. International Journal of Food Properties. 2006 Apr 1;9(1):99-107.
10. Costa LC, Kaspchak E, Queiroz MB, Almeida MM, Quast E, Quast LB. Influence of temperature and homogenization on honey crystallization. Brazilian Journal of Food Technology. 2015 Apr;18:155-61.
11. Dettori A, Tappi S, Piana L, Dalla Rosa M, Rocculi P. Kinetic of induced honey crystallization and related evolution of structural and physical properties. LWT. 2018 Sep 1;95:333-8.
12. Dobre I, Georgescu LA, Alexe P, Escuredo O, Seijo MC. Rheological behavior of different honey types from Romania. Food Research International. 2012 Nov 1;49(1):126-32.
13. El Sohaimy SA, Masry SH, Shehata MG. Physicochemical characteristics of honey from different origins. Annals of Agricultural Sciences. 2015 Dec

1;60(2):279-87.
14. EL-Metwally AAE. Factors Affecting the Physical and Chemical Characteristics of Egyptian Beehoney. Ph. D. Thesis, Fac. Agric. Cairo Univ, 2015, 320p.
15. Escuredo O, Dobre I, Fernández-González M, Seijo MC. Contribution of botanical origin and sugar composition of honeys on the crystallization phenomenon. Food chemistry. 2014 Apr 15;149:84-90.
16. Farooq R, Majid S, Hanif A, Ashraf A, Khan A. Different Types of Honey and Their Properties. In Therapeutic Applications of Honey and its Phytochemicals. Springer, Singapore. 2020, 261-278.
17. Felsner ML, Cano CB, Matos JR, Almeida-Muradian LB, Bruns RE. Optimization of thermo gravimetric analysis of ash content in honey. Journal of the Brazilian Chemical Society. 2004;15:797-802.
18. FSSAI (Manual of methods of analysis of foods (beverages and confectioneries) - A Laboratory Manual 4. Food safety and standards authority of India, Ministry of health and family welfare, Government of India, New Delhi, India, 2015.
19. Gül A, Pehlivan T. Antioxidant activities of some monofloral honey types produced across Turkey. Saudi journal of biological sciences. 2018 Sep 1;25(6):1056-65.
20. Gündoğdu E, Çakmakçı S, Şat İG. An overview of honey: its composition, nutritional and functional properties. J. Food Sci. Eng. 2019;9:10-4.
21. Hagr TE, Mirghani ME, Elnour AA, Bkharsa BE. Antioxidant capacity and sugar content of honey from Blue Nile State, Sudan. International Food Research Journal. 2017 Dec 2, 24.
22. Hamdan K. Crystallization of honey. Bee World. 2010 Jan 1;87(4):71-4.
23. Khan SU, Anjum SI, Rahman K, Ansari MJ, Khan WU, Kamal S *et al.* Honey: Single food stuff comprises many drugs. Saudi journal of biological sciences. 2018 Feb 1;25(2):320-5.
24. Lawal RA, Lawal AK, Adekalu JB. Physico-chemical studies on adulteration of honey in Nigeria. Pakistan journal of biological sciences: PJBS. 2009 Aug 1;12(15):1080-4.
25. Mohammed SA, Babiker EE. Protein structure, physicochemical properties and mineral composition of Apis mellifera honey samples of different floral origin. Australian Journal of Basic and Applied Sciences. 2009;3(3):2477-83.
26. Pauliuc D, Dranca F, Oroian M. Antioxidant activity, total phenolic content, individual phenolics and physicochemical parameters suitability for Romanian honey authentication. Foods. 2020 Mar 8;9(3):306.
27. Shah P, Modi HA. Comparative study of DPPH, ABTS and FRAP assays for determination of antioxidant activity. Int. J. Res. Appl. Sci. Eng. Technol. 2015;3(6):636-41.
28. Singh I, Singh S. Honey moisture reduction and its quality. J Food Sci Technol. 2018 Oct;55(10):3861-3871. doi: 10.1007/s13197-018-3341-5. Epub 2018 Jul 24. PMID: 30228384; PMCID: PMC6133847.
29. Smanalieva J, Senge B. Analytical and rheological investigations into selected unifloral German honey. European Food Research and Technology. 2009 May;229(1):107-13.
30. Suriwong V, Jaturonglumlert S, Varith J, Narkprasom K, Nitatwichit C. Crystallisation behaviour of sunflower and longan honey with glucose addition by absorbance measurement. International Food Research Journal. 2020 Aug 1;27(4).
31. Tappi S, Laghi L, Dettori A, Piana L, Ragni L, Rocculi P. Investigation of water state during induced crystallization of honey. Food chemistry. 2019 Oct 1;294:260-6.
32. Venir E, Spaziani M, Maltini E. Crystallization in Tarassaco Italian honey studied by DSC. Food chemistry. 2010 Sep 15;122(2):410-5.
33. Zhang Z, Sun DW, Zhu Z, Cheng L. Enhancement of Crystallization Processes by Power Ultrasound: Current State-of-the-Art and Research Advances. Comprehensive Reviews in Food Science and Food Safety. 2015;14(4):303-316. doi:10.1111/1541-4337.12132