Physicochemical properties, sensory acceptability, and antioxidant activity of chocolate bar fortified by solid lipid nanoparticles of gallic acid

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ABSTRACT
The chocolate bar is a processed cocoa product that is rich in polyphenols, but polyphenols decrease during processing. Gallic acid encapsulated in the form of solid lipid nanoparticles (SLN) can be a solution for the fortification of phenolic compounds in chocolate bars. This study aimed to obtain the right concentration of SLN-gallic acid in the manufacture of chocolate bars. SLN was made by the double emulsion method and added fat rich in Monoacylglycerol (MAG) and Diacylglycerol (DAG) from coconut stearin as an emulsifier as well as a solid lipid matrix. The concentrations of SLN-gallic acid used for fortification were 0%, 2.5%, 5%, and 7.5%. The results showed that the addition of SLN-gallic acid affected the characteristics of physicochemical, sensorial, and antioxidant activity of chocolate bars. The higher the concentration of the addition of gallic acid SLN increased the total phenolic and antioxidant activity. However, the characteristics of color, texture, and organoleptic properties were decreased. The microstructure of the chocolate bar did not show a significant difference, namely the spherulite needlelike crystal. The best treatment was obtained by adding 5% of SLN-gallic acid on chocolate bars with an 

Introduction

Chocolate is known as a food processed from cocoa beans with a high polyphenols content, but the polyphenols are damaged, oxidized, and their bioavailability is decreased due to processing.\(^1\) Cocoa beans undergo a multistep process starting from fermentation, drying, roasting, grinding, conching, and tempering, which have the potential to reduce polyphenol levels.\(^2,3\) The progressive decrease in phenolic content generally occurs in the roasting process.\(^4\)

The instability of phenolic compounds in products has become one of the concerns in the food industry.\(^5\) Gallic acid is one of the phenolic compounds in chocolate bars and belongs to the natural polyphenol group of phenolic acid types, which are hydrophilic and have high antioxidant activity, have anti-bacterial and anti-browning properties, and can prevent lipid oxidation.\(^6\) However, gallic acid has the disadvantages of poor absorption, rapid elimination, and instability.

One way to improve the bioavailability of phenolic compounds is by applying nanotechnology. In this decade, nanotechnology has become a renewable technology with particles measuring 1–100 nm with the principle of maintaining the protection of hydrophobic and hydrophilic compounds so that...
their characteristics, stability, and availability can be improved, especially in the delivery system. \cite{7,8} Solid Lipid Nanoparticles (SLN) is a carrier system that has a crystallized lipid core with a dispersed phase in the form of a mixture of solid lipids – bioactive compounds stabilized by an emulsifier to increase encapsulation efficiency and functionality.\cite{9,10}

The emulsifier in SLN plays an important role in determining dispersion stability and final particle size.\cite{11} Emulsifiers rich in MAG and DAG have advantages such as being anti-toxic and potentially increasing the bioavailability and shelf life of the product.\cite{12-15} Duan et al.\cite{16} reported that the SLN stability pattern was superior among all types of nanoparticles. Peres et al.\cite{17} and Mazur et al.\cite{18} have developed a solution for the encapsulation of phenolic compounds in SLN by applying the principle of melt dispersion and double emulsion (W1/O/W2) without solvent to interact with hydrophilic compounds.

SLN with the double emulsion method can be a solution to increase the availability of phenolic compounds that have low bioavailability.\cite{19} Fortification of gallic acid SLN in chocolate bars can assist in providing nutritious food as well as providing solutions for functional foods. Several studies reported that the fortification of phenolic compounds in chocolate bars had a positive effect on the availability of phenolic compounds and antioxidant activity. The concentration of fortificants can affect the characteristics of the resulting chocolate bar. The addition of fortificants that are too low gives the effect of antioxidant activity that is less than optimal. Meanwhile, too high a concentration can reduce glossy characteristics, increase hardness texture, and affect the taste and flavor of chocolate bars.\cite{20-22}

The purpose of this study was to obtain the best concentration of SLN-gallic acid to be applied in the production of chocolate bars so as to produce good physicochemical characteristics, antioxidant activity, and sensorial properties favored by panelists. SLN-gallic acid was prepared by double emulsion method with an emulsifier in the form of fat enriched with MAG and DAG from coconut stearin which also acts as a solid matrix in SLN.

**Materials and methods**

**Materials**

Cocoa powder and cocoa butter were obtained from PT. Barry Callebaut, gallic acid, glycerol, NaOH, hexane, and tert-butanol were obtained from Merck KgA (Darmstadt, Germany), and other supporting materials were obtained from supermarkets.

**Preparation of emulsifier fat rich in MAG and DAG**

Fat Rich in MAG and DAG was prepared by chemical glycerolysis in a solvent system, according to Subroto et al.\cite{23} 50 grams of coconut stearin and 20.85 grams of glycerol (molar ratio 1:5) were mixed at 60°C, then added tert-butanol at a ratio of 1:1 (w/v). Glycerolysis was carried out in a reflux reactor at a temperature of 90–100°C for 4 hours using NaOH 3% as a catalyst. The reaction mixture from reflux was then neutralized with 20% of citric acid, and then hexane was added at a ratio of 1:3 (v/v) to extract the fat rich in MAG and DAG. Hexane was evaporated using a rotary evaporator, while the fat rich in MAG and DAG was taken and stored in the refrigerator until used.

**Preparation of SLN-gallic acid**

SLN-gallic acid was prepared by double emulsion method (W1/O/W2) and melt dispersion technique without solvent.\cite{17} 12 g of stearic acid and fat rich in MAG and DAG (20% of total lipid) were melted at a temperature of 60–75°C. The mixture was added with 2 mL of 1% (w/v) gallic acid solution and homogenized with a high-speed homogenizer; then, the first ultrasonication was carried out on the mixture for 3 minutes (setting 40 seconds on, 1 second off) for the formation of the first emulsion
Preparation

Preparation of chocolate bars fortified with SLN-gallic acid

Chocolate bars were made following the method of Ibrahim et al.\(^{[24]}\) with some modifications. The ratio of cocoa powder, cocoa butter, and sugar used was 25:45:30. The ingredients were mixed, and conching was carried out for 3 hours at a temperature of 50°C. The chocolate paste was then conched again for 15 minutes to add gallic acid SLN for each treatment (0%, 2.5%, 5%, and 7.5%). The chocolate paste was then tempered by increasing the temperature to 50°C and then lowered to 32°C and 27°C and raised again to 32°C. The last stage was molding, cooling (T = 12–15°C; t = 24 hours), then demolding and packaging with aluminum foil.

Analysis of particle size and polydispersity index

The particle size was analyzed by HORIBA SZ-100 laser diffraction at a scattering angle of 90.\(^{[25]}\) The polydispersity index (PI) was determined from the Scattering Light Intensity representation. The SLN supernatant was diluted with distilled water to reach 1–100 ppm.

Analysis of entrapment efficiency

Entrapment efficiency (EE) of gallic acid was determined by calculating the difference between total phenolic content (TPC) and surface phenolic content (SPC).\(^{[17]}\) SPC was obtained from the supernatant separated from the SLN. Gallic acid content was tested using a spectrophotometer at a wavelength of 725 nm. The value of entrapment efficiency can be calculated by the formula:

\[
\% EE = \frac{\text{EPC}}{\text{TPC}} = \frac{\text{TPC} - \text{SPC}}{\text{TPC}} \times 100\%
\]

where, EE is entrapment efficiency (%), TPC is the total phenolic content, SPC is surface phenolic content.

Analysis of color

The color of the chocolate bar was tested using a Conica Minolta CM-5 chromameter.\(^{[26]}\) The chromameter was calibrated, and then the samples were measured for the values of L*, a*, and b* for three repetitions. The degree of HUE (°H) of each sample was calculated by the following formula:

\[
H = \tan^{-1} \frac{b*}{a*}
\]

Analysis of texture profile

The texture profile of chocolate bars was tested using the Stable Micro System Texture Analyzer TAX-T2.\(^{[27]}\) The chocolate bars were stored at 32°C for 2 hours, and then the hardness and stickiness was measured using a 2 mm cylindrical probe 25 kg load cell. The probe speed was carried out at a pretest of 1.0 mm/s, a test of 5.0 mm/s, and a posttest of 10.0 mm/s, and the probe on the instrument was

(W1/O). Tween 80 concentration of 20% was added to the first emulsion and homogenized using a high-speed homogenizer at a temperature of 60°C for 10 minutes, and then a second ultrasonication was carried out on the mixture (A = 45% 500 W) for 5 minutes (10 seconds on, 2 seconds off) to the formation of a second emulsion (W1/O/W2). The SLN was dispersed in distilled water at a temperature of 5–10°C, and then a third ultrasonication was performed using the same time and set as the second ultrasonication. Samples were frozen at −50°C and then lyophilized for 72 hours. SLN-gallic acid was then stored in the refrigerator until used.
lowered at 50 mm above the sample surface and penetrated 30 mm down. The measurement parameters observed included hardness, springiness, cohesiveness, adhesiveness, chewiness, gumminess, and resilience.

**Analysis of microstructure**

The microstructure of chocolate bars was observed using a Polarized Light Microscope (PLM).[28] The chocolate bar was melted at a temperature of 60°C and cooled to 30°C. The chocolate was spread on a glass slide set on a temperature control unit that was kept at 30°C under circulating refrigeration.

**Analysis of total phenolic compound**

The total phenolic compound assay was carried out according to the modification of Lee et al.[29] The defatted chocolate bar was weighed as much as 0.5 g and then diluted with 24.5 mL of distilled water. 0.5 mL of the sample mother liquor filtrate was added with 0.5 mL of Folin-Ciocalteu 50%, 2.5 mL of 7% Na₂CO₃ and aquadest were added to a 25 mL volumetric flask. The solution was incubated for 40 minutes in a dark room and then measured with a spectrophotometer at a wavelength of 725 nm to obtain the phenolic content in mg GAE/gram.

**Analysis of antioxidant activity**

Testing of antioxidant activity was carried out following the modification of Molyneux et al.[30] 2.5 mL of the sample mother liquor (700 ppm) was added with 0.5 mL of DPPH. The solution was incubated for 30 minutes in a dark room and then measured with a spectrophotometer at a wavelength of 517 nm.

**Sensory evaluation**

Parameters for sensory evaluation included taste, color, texture, aroma, and after taste.[31] The test was conducted on 20 semi-trained panelists (aged 16–50 years). The panelist’s level of preference for the product was assessed with a score of 1–5, namely 1: very dislike, 2: dislike, 3: normal, 4: like, and 5: like very much). Data processing was continued with the Kruskal-Wallis test and the Mann-Whitney test.

**Analysis of crystal morphology**

Crystal morphology on the surface of chocolate bars was tested by Scanning Electron Microscope (SEM).[32] The sample was placed in a sample holder and then coated with gold. The sample was then transferred to the instrument to obtain different micrographs at various magnifications, namely 200x, 500x, and 1000x.

**Analysis of polymorphism**

The pattern of chocolate bar polymorphism was tested by X-Ray Diffraction (XRD) according to Le Révérend et al.[33] The sample was placed on the slide, then the X-ray diffraction pattern was observed at room temperature (25–30°C) at 2θ 3–30 deg.
Results and discussion

Characteristics of fat rich in MAG and DAG from glycerolysis

Characteristics of fat rich in MAG and DAG from glycerolysis of coconut stearin can be seen in Table 1. Glycerolysis produced MAG with the highest value of 84.53 ± 0.04%. These results were in accordance with the research of Zhong et al.,\textsuperscript{[34]} who carried out chemical glycerolysis and obtained products that have more than 80% MAG content with a TAG conversion ratio of 97%. Chemical glycerolysis using a NaOH catalyst is one of the effective MAG and DAG production methods.\textsuperscript{[35]} The results of the high MAG composition were in line with the high value of the resulting emulsion capacity. MAG and DAG-rich fat had an emulsion capacity of 97.78 ± 3.85%, which indicates this emulsifier was effective and stable to be applied to emulsion products. According to Li et al.,\textsuperscript{[36]} the emulsion capacity depends on the acylglycerol composition and the formulation used. The higher the emulsification ability along with the high content of MAG. This fat rich in MAG and DAG was then used as an emulsifier and a solid matrix mixed with stearic acid for the synthesis of SLN-gallic acid.

Characteristics of SLN-gallic acid

Characteristics of SLN-Gallic Acid can be seen in Table 2. The particle size of SLN-gallic acid is in the range of 224.40–3596.30 nm, with a polydispersity index of 0.85. This indicates that the particle size of SLN-gallic acid produced varies, some less than 1000 nm, but some are greater than 1000 nm. The variation in particle size and the high polydispersity index of SLN-gallic acid may be caused by agglomeration between SLN-gallic acid particles to form large particles. This could be due to the fact that lipids are easy to associate with each other through hydrophobic interactions because they have the same affinity, and lipids have the property of being able to crystallize and transform into three polymorphic forms, namely α, β', dan β.\textsuperscript{[37]} According to Danaei et al.,\textsuperscript{[38]} the greater the polydispersity index value and closer to 1, it means that the particle distribution is not uniform. The particle size of the resulting SLN can affect the EE value of the SLN-gallic acid produced. In this study, the EE value obtained was 93.75%. Lal et al.\textsuperscript{[39]} explained that relatively large nanoparticles tend to encourage the faster release of drug/bioactive compounds and easily for polymer degradation.

Characteristics of chocolate bars fortified with SLN-gallic acid

Color of chocolate bar

Color characteristics are one of the important factors that influence people’s attitudes and perceptions when choosing products. The effect of SLN-gallic acid fortification on the color characteristics of chocolate bars showed an increase in the values of L, a*, b*, and HUE, as shown in Table 3. These

| Table 1. Characteristic of fat rich in MAG and DAG. |
|-----------------------------------------------|
| Characteristics                  | Value            |
| Composition of acylglycerols:          |                  |
| ● Monoacylglycerol (MAG)              | 84.53 ± 0.04%    |
| ● Diacylglycerol (DAG)                | 12.53 ± 0.03%    |
| ● Triacylglycerol (TAG)               | 5.98 ± 0.03%     |
| Emulsion Capacity                    | 97.78 ± 3.85%    |

| Table 2. Characteristics of SLN-Gallic Acid. |
|-----------------------------------------------|
| Characteristics                  | Value            |
| Particle Size                     | 224.40–3596.30 nm|
| Polydispersity index              | 0.85             |
| Entrapment Efficiency (EE)        | 93.75%           |
results are in accordance with the study of Afoakwa et al., who reported that the high-fat content of SLN-gallic acid in chocolate increased the value of $L^*$ (brightness), $C^*$ (chroma), and HUE, so that the resulting chocolate bar will be lighter in color. Based on the HUE value, all chocolate bars were still in the category of reddish-brown intensity.

**Texture of the chocolate bar**

The texture of chocolate bars fortified with SLN-gallic acid can be seen in Table 4. Fortification of SLN-gallic acid caused the characteristic parameter of chocolate bar texture to decrease, especially on hardness. Hardness characteristics were the main characteristics observed in chocolate bars. Afoakwa et al. stated that hardness characteristics showed an inverse relationship with fat content, particle size, and lecithin content in chocolate bars. The fat constituent of SLN-gallic acid composed of MAG and DAG-rich fat from coconut stearin has a lower hardness than cocoa butter, so the addition of SLN-gallic acid in chocolate bars increased the fat content and decreased the hardness of the chocolate bars. Cohesiveness also decreased, especially with the addition of more than 5% SLN-gallic acid. The decrease in cohesiveness can be affected by the presence of small size SLN-gallic acid particles which were distributed and filled the space between the larger particles, thereby reducing the compactness of chocolate bars and becoming. This was consistent with the decrease in adhesiveness as a parameter of stickiness. Fibrianto et al. reported that the smaller the particle size, the stickier the chocolate bar texture. The decrease in gumminess and chewiness was in line with the decrease in the resilience characteristics of the chocolate bar, which indicated that the product was easier to chew and more easily crushed. Gumminess value was directly proportional to hardness. Chewiness is the amount of energy required to chew the product.

The springiness characteristic increased with the addition of SLN-gallic acid. This was due to the presence of lipid additives from SLN. Mamat and Hill reported that the addition of palm-mild fraction could reduce springiness characteristics. Gallic acid SLN, which was a double emulsion in chocolate bars, also had the potential to change the texture. Leister and Karstein stated that multiple emulsions could experience some coalescence and diffusion phenomena after the production process, which consequently has an impact on product properties, such as texture.

**Microstructure of chocolate bar**

The microstructure of chocolate bars fortified with SLN-gallic acid can be seen in Figure 1. Fortification of SLN-gallic acid did not significantly affect the microstructure of the crystals visually in chocolate bars. The chocolate bar crystals were predominantly spherulite and did not show any needlelike crystals. If a needlelike crystal appears, it can indicate that the chocolate bar has experienced a fat bloom.

### Table 3. The color of chocolate bars fortified with SLN-gallic acid.

| Concentration of SLN-Gallic Acid | $L^*$     | $a^*$     | $b^*$     | HUE      |
|----------------------------------|-----------|-----------|-----------|----------|
| 0%                               | 22.38 ± 1.19 | 4.14 ± 0.45 | 3.15 ± 0.31 | 37.05 ± 1.01 |
| 2.5%                             | 23.97 ± 0.43 | 4.42 ± 0.46 | 3.26 ± 0.29 | 37.07 ± 0.84 |
| 5%                               | 24.37 ± 0.11 | 4.82 ± 0.88 | 3.91 ± 0.77 | 39.11 ± 0.89 |
| 7.5%                             | 25.55 ± 0.40 | 5.13 ± 0.18 | 4.59 ± 0.20 | 41.81 ± 0.35 |

### Table 4. The texture of chocolate bars fortified with SLN-gallic acid.

| Concentration of SLN-Gallic Acid | Hardness | Springiness | Cohesiveness | Adhesiveness | Gumminess | Chewiness | Resilience |
|----------------------------------|----------|-------------|--------------|--------------|-----------|-----------|------------|
| 0%                               | 2888 ± 123 | 0.70 ± 0.06 | 0.78 ± 0.04  | −61 ± 1      | 2251 ± 26 | 1758 ± 113 | 0.63 ± 0.03 |
| 2.5%                             | 2777 ± 106 | 0.79 ± 0.13 | 0.86 ± 0.07  | −16 ± 1      | 1953 ± 186 | 1684 ± 281 | 0.63 ± 0.06 |
| 5%                               | 2125 ± 173 | 0.78 ± 0.14 | 0.67 ± 0.06  | −16 ± 1      | 1409 ± 129 | 940 ± 159  | 0.51 ± 0.05 |
| 7.5%                             | 1290 ± 240 | 0.92 ± 0.03 | 0.58 ± 0.11  | −8 ± 2       | 748 ± 217  | 443 ± 186  | 0.39 ± 0.07 |
The microstructure is an indicator of the appearance of fat blooms. Fat bloom can be caused by an imperfect tempering process, interaction with other materials, and storage temperature. The addition of SLN-gallic acid could prevent fat bloom in chocolate bars because SLN contains emulsifiers rich in MAG and DAG, where the emulsifier plays a role in preventing the formation of fat blooms. This means that fortification of SLN-gallic acid contained stearic acid and an emulsifier rich in MAG and DAG can result in good and stable crystal formation in fat-based food products. The relationship between all ingredients present in chocolate can affect microstructural properties, rheological characteristics, and texture characteristics. However, the fortification of SLN-gallic acid did not affect the microstructure of chocolate bars.

**Total phenolics and antioxidant activity of chocolate bar**

Based on Table 5, fortification of SLN-gallic acid increased the total phenolic content of chocolate bars. These results showed that the fortification of gallic acid SLN at concentrations of 2.5%, 5%, and 7.5% in chocolate bars gave an increase in the total phenolic content. The total phenolic content produced ranged from 7.54–11.62 mgGAE/g. Total phenolic increased linearly as the concentration of fortified SLN-gallic acid increased. The results were in line with the research of Godočíková et al., who made
Table 5. Total phenolic content and antioxidant activity of chocolate bars fortified with SLN-gallic acid.

| Concentration of SLN-Gallic Acid | Total Phenolic Content (mg GAE/g) | IC50 (ppm) |
|----------------------------------|------------------------------------|------------|
| 0%                               | 7.54 ± 0.11                        | 200.67 ± 1.53 |
| 2.5%                             | 8.43 ± 0.10                        | 179.07 ± 4.55 |
| 5%                               | 9.48 ± 0.03                        | 174.24 ± 2.48 |
| 7.5%                             | 11.62 ± 0.09                       | 184.66 ± 3.75 |

dark chocolate bars fortified with mulberries. The control chocolate bar had a total phenolic content of 7.32 ± 1.02 mg GAE/g, while the chocolate bar fortified with mulberry fruit had a total phenolic content of 8.83 ± 1.29 mg GAE/g. SLN has been known as a solution to increase the stability and bioavailability of gallic acid encapsulated in lipid matrix while protecting the active compound during processing.  

Fortification of SLN-gallic acids 2.5%, 5%, and 7.5% also affected the antioxidant activity of chocolate bars (Table 5). The increase in the concentration of SLN-gallic acids increased the total phenolic content and was positively correlated with the increase in antioxidant activity, as indicated by the smaller IC50 value. The smaller IC50 value in a material indicates that the antioxidant activity is getting higher. However, the resulting IC50 value range was still in the weak criteria, namely in the range of 100–200 ppm.  

The addition of SLN-gallic acid 2.5% and 5% showed the IC50 value, which was not too much different, presumably caused by the polyphenol component in the inhibitory activity of gallic acid encapsulated by SLN against hydroperoxides which were equally weak. Based on Table 5, the total phenolic content gave a positive correlation with antioxidant activity, but the concentration of SLN-gallic acid 7.5% was not proportional to the antioxidant activity. The addition of antioxidant compounds in large quantities can allow the formation of pro-oxidant compounds, which can reduce the antioxidant activity of the product.  

The decrease in the SLN-gallic acid treatment of 7.5% can also be caused by the high-fat content in chocolate which can interfere with the free radical scavenging process because hydrogen atoms from fat can bind to hydroxyl radicals in DPPH and make DPPH radicals more active, so that the reduction process becomes less than optimal.

Gallic acid is a good source of antioxidants, is relatively safe, and has low acute oral toxicity. The use of gallic acid up to 0.2% or consumption of gallic acid as much as 119 mg/kg/day on male rats did not cause any side effects. Therefore, the use of SLN-gallic acid as much as 5% and 7.5% in this study was equivalent to the gallic acid content of 9.48 ppm and 11.62 ppm, respectively (Table 5), still very far from the safe limit for the use of gallic acid of 0.2%. However, excessive use of gallic acid can cause toxic effects, including a reduction of hematocrit concentration, hemoglobin, and an increase in reticulocytes.  

Sensorial properties of chocolate bar: Sensorial properties of chocolate bar fortified with SLN-gallic acid can be seen in Figure 2. SLN-gallic acid fortification did not show significant differences in color, aroma, and texture parameters (P > .05). This indicated that the fortified chocolate bar with SLN-gallic acid was well received by the panelists. Meanwhile, the parameters of taste, after taste, and overall acceptance showed that they were continued with the Mann-Whitney test and had significant differences in the treatment of SLN-gallic acid fortification, especially at a concentration of 7.5%. The addition of SLN-gallic acid that was too high increases the bitter taste of the chocolate bar. Gallic acid in SLN caused a bitter taste which was not favored by the panelists. The increase in bitter taste also had an impact on the appearance of an after-taste that was less favored by the panelists. The taste parameter has a high correlation with the after-taste parameter.  

Figure 2. also shows a positive correlation that the addition of SLN-gallic acid 2.5–5% can be accepted or favored by the panelists. Meanwhile, the addition of SLN-gallic acid at a concentration of 7.5% showed the lowest average value for all hedonic test parameters. The 5% SLN-gallic acid fortification treatment was chosen as the best sample because it had the highest average overall...
acceptability parameter and antioxidant activity. The product in the best treatment was then analyzed on its morphology and polymorphism pattern to determine the crystallization characteristics.

**Morphology of chocolate bar**

Morphology of the chocolate bar fortified with SLN-Gallic Acid 5% and 0% (control) can be seen in Figure 3. The best surface morphology of chocolate bars (SLN-gallic acid 5%) and control (0%) showed that visualization by SEM was not significantly different in where the particles were evenly distributed, and the crystals were scattered in the form of small spherulites. Microstructure visualization by SEM also showed that the chocolate bar crystals did not show any needle like crystals. Kinta and Hatta\cite{Kinta and Hatta};
reported that a good chocolate morphology had a uniform composition; there were no needlelike crystals. The crystals in good chocolate bars moved to the most stable energy state even though the chocolate was thermodynamically unstable. These results indicated that fortification of SLN-gallic acid at 5% was compatible with being applied and had no effect on the formation of chocolate bar crystals.

**Polymorphism of chocolate bar:** The diffractogram of the chocolate bar fortified with SLN-Gallic Acid 5% and control (0%) can be seen in Figure 4. The best chocolate bar (SLN-gallic acid 5%) and the control chocolate bar showed a polymorphic pattern that was not significantly different. Chocolate bar fortified with SLN-gallic acid 5% had the highest peak at short spacing or the value of d = 4.57 Å indicated β crystal form and shows another peak at d = 3.5–4.2 Å indicates β’ form crystal.[52,53] These results showed that the SLN-gallic acid fortified chocolate bar had a combination of β (V) and β’ (IV) crystals. This also indicated that the constituent fat of SLN-gallic acid was still compatible up to 5% with cocoa butter in chocolate bars.

Stearic acid and fat rich MAG and DAG from coconut stearin as a constituent of SLN-gallic acid have a polymorphic pattern that is almost the same as cocoa butter. The results of this polymorphic pattern were also in line with the research of Le Révérend et al.[33] which showed the highest diffraction peak in dark chocolate was produced at d = 4.5–4.6 Å. The highest peak intensity of gallic acid SLN fortified chocolate bars was at d = 4.57 Å indicating that the chocolate bars were still dominated by β-crystals. The β-crystal form is recommended for chocolate bars because it is not easy to melt (stable), creates a suitable mouthfeel, and the texture is firm, shiny, and snaps when bitten.

**Conclusion**

Fortification of SLN-gallic acid in chocolate bars affected the characteristics of physicochemical, sensorial, and antioxidant activity. The higher the concentration of SLN-gallic acid fortified in chocolate bars caused a decrease in texture and sensorial properties. In contrast, total phenolic and antioxidant activity increased. The microstructure of chocolate bars did not show any significant difference. The best treatment was obtained from chocolate bar fortified with SLN-gallic acid of 5%, which resulted in an IC₅₀ of 174.24 ± 2.48 and was the most preferred by the panelists. The chocolate bar had a spherulite crystal morphology, small size, and was evenly distributed, and the polymorphic pattern was dominated by beta crystals. Thus, the fortification of SLN-gallic acid has compatibility of up to 5% on chocolate bars.
Disclosure statement

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