Effect of iron fortification on gummy candies properties: basic nutrient, microstructure, and texture during the storage period

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Abstract. Iron fortification program relates to iron delivery through daily foods, which have a potential impact on many people. In this study, chitosan microparticle loaded with ferrous gluconate was used as fortificant, while gummy candies were selected as food vehicles. This work aimed to assess the effect of iron microparticles addition on basic nutrient composition, textural, and microstructural characteristics. The properties of iron microparticles obtained from the spray drying method were also discussed in this article. The iron fortification was carried out by adding the iron microparticles into the candies solution. This solution was then molded and cooled at room temperature before the analysis. The yield, iron loading, efficiency encapsulation, mean diameter, and SPAN factor of iron microparticles were 49%, 3.5 mg Fe/100 mg dried microparticles, 48%, 17 µm, and 1.7. The iron microparticle had a spherical shape with the wrinkles structure on the surface. Therefore, the fortified gummy candies had wavy structures with the prominent spheres. The protein and fat content of our gummy candies were higher than the commercial gummy candies. However, gummy candies' hardness and gumminess value had changed significantly, while the cohesiveness and springiness properties were not significantly different during the storage.

1. Introduction
Iron fortification is considered a productive, effective, and realistic way to prevent and reduce iron deficiency [1,2]. Iron deficiency is the most common cause of anemia, and its negative impacts are more perceived by pregnant women and children [3]. The fortification program relates to iron delivery through daily foods, thus increasing the iron status of many people [3].

A proper understanding of iron properties, including the possible chemical reaction between iron and food, could also enhance this program [3,4]. Some previous works investigated iron fortification of foodstuff, which contained calcium and zinc; however, the presence of this other divalent metal could inhibit the iron bioavailability [5,6]. Other previous research also reported that the existence of phytate and tannin could interrupt the iron absorption [7]. Therefore, in the case of fortification, food vehicle selection needs to be considered. Thus the nutritional content in these ingredients may not interfere with the iron. Gummy candies can be suitable food carriers since they are visually interesting.
for consumers, from children to elderlies [8,9]. The ingredients used in gummy candies production could also be chosen thereby reducing iron interaction with inhibitor and competitor.

However, the direct addition of iron mineral on food could be adverse to the sensory change [1,10]. Nowadays, the microencapsulation method is considered an appropriate way to protect and mask iron from inhibitor, competitor, and unacceptable flavor and color [10]. In this study, chitosan microparticle loaded with iron was used as fortificant, while gummy candies were selected as food vehicles. This work aimed to assess the effect of iron microparticles addition on basic nutrient composition, textural, and microstructural characteristics of gummy candies. The properties of iron microparticles obtained from the spray drying method were also discussed in this article.

2. Materials and methods

2.1. Materials
Iron (II) gluconate (FeG) was purchased from Sigma Aldrich. Chemicals for microparticles preparation, simulated gastrointestinal fluid, and iron determination were supplied from Merck. Chitosan was provided from CV. Chimultiguna Cirebon-Indonesia. Furthermore, gummy candy’s ingredients, such as corn sugar (glycemic index 22) and halal-gelatin (from cow bone, bloom 150), were provided from PT. Padihta-Jakarta and Global Capsule Ltd Bangladesh, respectively.

2.2. Microparticles Preparation
The feed solution was prepared by dissolving FeG into 2.5% v/v of Ac. Furthermore, Chitosan (0.5% w/v) and TPP (1% w/v) were added as encapsulating agent and cross-linking agent, respectively. The feed solution was then processed on Mini Buchi (B-290) spray dryer set at 160ºC of inlet temperature, 100% of the vacuum system, and 10% of pump flow. The yield of the spray dryer product was calculated by Eq. 1.

\[
\text{Yield} = \frac{\text{weight of dried microparticles}}{\text{weight of solid in feed solution}} \times 100\% \tag{1}
\]

2.3. Determination of total Fe, encapsulation efficiency, and iron loading
The destruction process of dried microparticles was carried out before the analysis. A 0.45 µm nylon filter syringe (Aijiren) was used to filter the destruction solution; hence it was ready for total Fe determination using the Phenanthroline method [4,11]. The absorbance was recorded using Thermo Scientific Genesys 10 S UV-Vis Spectrophotometer at 510 nm. Encapsulation efficiency (EE) and iron loading were calculated using Eq. 2 and 3, respectively.

\[
\text{Encapsulation efficiency (EE)} = \frac{\text{mg total Fe in microparticles}}{\text{mg total Fe used}} \times 100\% \tag{2}
\]

\[
\text{Iron loading} = \frac{\text{mg total Fe in microparticles}}{100 \text{ mg dried microparticles}} \tag{3}
\]

2.4. Iron fortification
Gummy candies were prepared by diluting gelatine 30% (w/v) with cold water. Corn sugar 25% (w/v) was then added into previously diluted gelatin and heated until the high viscosity was obtained. The iron fortification was carried out by adding the FeG microparticles (100 mg, 200 mg, 300 mg) into the already cool solution. The final mixture was poured into molds and cooled at room temperature; hence the gels were obtained and ready to analyze. The unfortified gummy candies were also prepared for the reference sample.
2.5. Scanning electron microscopy – SEM and Particle size analyzer – PSA
The morphology and size distribution of dried microparticles were analyzed using scanning electron microscopy – FEI Inspect F50 at 20.0 kV of excitation voltage and laser particle sizer LLPA-C10. SPAN factors were also determined to represent the particle size distribution.

2.6. Texture analysis
Textural properties and surface morphology of gummy candies were determined using a texture analyzer (CT3 Texture analyzer from Brookfield). The specific properties of the texture profile included hardness, cohesiveness, springiness, and gumminess.

2.7. Protein and fat analysis
The protein and fat content of gummy candies were determined by SNI 012354 42006 and SNI 2891 01 1992.

3. Results and Discussions
3.1. Properties of iron microparticles
Chitosan-TPP microparticles loaded with FeG were produced using a spray dryer method. The SEM and photo image of these microparticles are represented in Figures 1a and 1b. The microparticles had a spherical shape with a hollow in the middle and wrinkles on the surface. Chitosan microparticles loaded with FeG also had greenish-white color since the original color of FeG is green. These results were similar to previous research [12]. Figure 1c depicts that the mean diameter and SPAN factor of FeG microparticle were 17 µm and 1.7. This SPAN factor’s value indicates that the spray-dried microparticles have a narrow particle size distribution. The fortified particle size is important to be considered so as not to change the organoleptic properties, such as texture and taste of food [1,13]. Hence, these micro-sized particles might be a suitable carrier for the fortification program.

Table 1 depicts yield, water content, iron loading, and EE of FeG microparticles. The yield of spray-dried microparticles is relatively low due to the encapsulating agent's highly adhesive properties, thus increasing the likelihood of the particles sticking to the chamber wall [14]. In this study, chitosan had an adequate ability to encapsulate FeG showed by EE properties. Foregoing research used chitosan to entrap vitamin C and GSH, resulting in the efficiency of encapsulation properties, which was similar to this study [15,16]. The number of iron retained in dried microparticles was also higher than in our previous work.

![Figure 1. (a) SEM micrograph, (b) photo image, and (c) particle size distribution of FeG microparticles](image-url)
Table 1. Physical properties of FeG microparticles.

|                      | Yield (% | Water content (%) | Iron loading (mg Fe/100 mg dried microparticles) | Efficiency encapsulation (%) |
|----------------------|----------|-------------------|--------------------------------------------------|-------------------------------|
| FeG microparticles   | 49       | 8.5               | 3.5                                              | 48                            |

3.2. Basic nutrient composition of gummy candies

Gummy candies were formulated using gelatine as a gelling agent and corn sugar as a source of sugar. The fortification was conducted by adding the FeG microparticles into the already cool candies solution. The nutrient analysis represents that gummy candies produced by our team had higher protein and fat levels compared to the commercial gummy candies (Figure 2a). The higher level of protein might be due to the used of gelatine in this study. Gelatine is a protein compound obtained from partial hydrolysis of collagen [17]. The protein level was analyzed using SNI 01 2354 42006 method, which determined the protein by calculating the products' total nitrogen (N). Thus, it might also be caused by the presence of N from chitosan. The molecular structures of gelatine and chitosan are shown in Figure 3.

![Figure 2. (a) Fat and protein of gummy candies and (b) Fe total of fortified gummy candies](image)

The fortified gummy candies also had a higher level of iron than the unfortified. It is caused by FeG microparticles which were intentionally added. Recently, SNI 3547.2-2008 does not regulate the minimum or maximum standard of iron levels contained in candies. However, previous work reported that the recommended daily intake (RDI) of iron is 18-20 mg/day, and only 10% of iron can be absorbed in the duodenum and jejunum [7]. Another research represented that RDI of iron for men, women, and pregnant women are 8, 18, and 27 mg/day, respectively [18]. Commonly, people have a regular, natural diet containing iron, such as spinach and meat. However, they could have an additional extra iron by consuming gummy candies contained 2.5-7.4 mg of iron for every 100 g of candies; thus it can help to meet the iron needs of their body.
3.3. SEM analysis of gummy candies

The morphological structures of the unfortified and fortified candies surface were depicted in Figure 4. The unfortified gummy candies had homogenous and fine-grained structures (Figure 4a). The fine-grained structures might be due to the low bloom strength (bloom 150) used in this formulation [9]. However, the gummy candies fortified with 100 mg of FeG microparticles had a wavy structure with faint spheres (Figure 4b). The higher number of FeG microparticles added would induce a more wavy structure with the apparent spheres (Figure 4c and 4d). These structures might be due to the addition of FeG microparticles, which had a spherical shape and wrinkle surface (Figure 1a).

3.4. Texture analysis of gummy candies

The texture profiles of unfortified and fortified gummy candies stored for eight days are shown in Figure 5. Hardness and gumminess values of gummy candies had similar trends during the storage period, increasing on day 3, decreased on day 5, and increased on day 8. The decrease in water content probably caused the higher value of hardness on day three. The decrease of hardness at day five might be due to the decrease of pores number, which was caused by the breakage and fusion of the protein
matrix so that the texture became soft. The further reaction might increase the binding of added microparticles and the fused protein, thus increasing gummy candies’ hardness on day 8 [19]. However, the cohesiveness and springiness values of gummy candies were no significant differences during the storage period. Figure 5b and 5c also represent that the addition of FeG microparticles had not influenced the cohesiveness and springiness values of gummy candies. This result was similar to previous work, which reported that the addition of betanin-NLPs did not significantly effect the cohesiveness and springiness values of gummy candies [8].

Figure 5. Texture parameters as (a) hardness, (b) cohesiveness, (c) springiness, and (d) gumminess of unfortified and fortified gummy candies

4. Conclusions
The effect of iron fortification on gummy candies properties was already investigated. The fortification was conducted using iron microparticles produced by spray drying. The yield, iron loading, efficiency encapsulation, mean diameter, and SPAN factor of iron microparticles were 49%, 3.5 mg Fe/100 mg dried microparticles, 48%, 17 µm and 1.7, respectively. The iron microparticle had a spherical shape with the wrinkles structure on the surface. Therefore, the fortified gummy candies had wavy structures with the prominent spheres. The protein and fat content of our gummy candies were higher than the commercial of gummy candies. However, the hardness and gumminess value of gummy candies were significantly changed, while the cohesiveness and springiness properties were not significantly different during the storage.

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