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Organoleptic, hypoglycaemic, and *in vitro* starch digestion effects of formulated Melon Manis Terengganu peel powder

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Abstract

Melon Manis Terengganu (MMT) is comprised of 28 - 30% peel which is a by-product of food processing. The peel is a source of dietary fibre which has a potential role in glycaemic response. The present work thus aimed to develop formulated MMT peel powder, and examine its organoleptic properties, *in vitro* hypoglycaemic effect, and starch digestibility. The MMT peel powder was formulated as Formulations 0, 1, 2, and 3 with different sweetener ratios (0, 40, 50, and 60%), and subjected to sensory evaluations. Tukey's *post-hoc* test was used to evaluate significant differences between mean values following one-way analysis of variance (ANOVA). Meanwhile, the Friedman test followed by Wilcoxon signed ranks test were performed for sensory evaluation analysis. Results demonstrated that the most acceptable formulation for consumption assessed using sensory evaluation was Formulation 3; its total, digestible, and resistant starch content were the lowest among all the formulations. The same went to the hydrolysis index and estimated glycaemic index. However, Formulation 3 was the least effective in reducing glycaemic response due to the weakest *in vitro* hypoglycaemic activity. On the other hand, the mentioned attributes previously were observed in Formulation 0 in an opposite manner. In summary, these findings suggested that formulated MMT peel powder had the potential to be used in blood glucose control.

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Introduction

The incidence of type 2 diabetes mellitus (T2DM) keeps on increasing each year, and it is estimated that 10.2% of the world's population (578 million) will have T2DM by 2030 (Saeedi *et al.*, 2019). T2DM and its complications are a serious global issue. Therefore, it is imperative to adopt therapeutic approaches that can improve or prevent this epidemiological situation (Pino *et al.*, 2021). Dietary modifications play a vital role in T2DM management. A study claimed that low glycaemic index (GI) food plays a crucial role in reducing the glycaemic response as blood glucose levels do not rise following its consumption (Ruijgrok *et al.*, 2021). The pace of starch digestion in the intestine affects the glycaemic response, and the GI values have been found (*in vitro*) to be positively associated to rapidly available glucose, and negatively associated to slowly available glucose (Rojas-Bonzi *et al.*, 2020). This indicates that not only the amount, but also the quality of carbohydrates in the meal, can influence the glycaemic response (Stephenson *et al.*, 2014). Besides, dietary fibre consumption has also been proven to exert a beneficial effect in lowering T2DM risk (McRae, 2018). This is because dietary fibre can decrease a meal's glycaemic load (Raninen *et al.*, 2011) which reduces the glycaemic response. Hence, it is essential to explore innovative and practical strategies to increase daily dietary fibre consumption to achieve better glycaemic control (Tongyu and Chong-Do, 2021).

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Generally, the food industry generates large volumes of wastes due to the production, preparation, and consumption of food. Consequently, this results in the loss of valuable biomass and nutrients (Hao *et al.*, 2021), including dietary fibres. Based on the statistics from the Food and Agriculture Organization (FAO), 1.3 billion tons of food is discarded globally, in which 30% are derived from cereals; 40 - 50% are derived from root crops, fruits, and vegetables; 20% are derived from oilseeds; and 30% are derived from fish (UNEP, 2019). Therefore, the conversion of value-added products from food-processing waste and by-products is warranted to reduce pollution created by landfills (Soni and Saxena, 2020).

Recent research reported that 28 - 30% of peel derived from Melon Manis Terengganu (MMT; *Cucumis melo* L. var. Inodorus cv. Manis Terengganu 1) was discarded (Ong *et al*., 2021a). Several studies on MMT peel reported that it contains bioactive compounds that are worthy for further investigation (Ong *et al.*, 2020; 2021b). MMT peel has high protein, total dietary fibre, vitamin A, vitamin D, calcium, and magnesium, as well as a source of zinc and iron with low fat content.

In view of its good nutritional compositions, it is expected that MMT peel could be useful in the product formulation for T2DM individuals. A recent scoping review also reported that *C. melo* exhibited antidiabetic properties (Ong *et al.*, 2019). Therefore, it is imperative to further confirm the therapeutic effect of MMT peel towards glycaemic control, so that it can be used as functional ingredients in food products. However, MMT peel is still underutilised in the functional food industry, in which, very little is known about MMT peel in the product formulation at this moment. Besides, to date, there is scarcity with reference to the investigation on the MMT peel, whereby no published work on its hypoglycaemic effect and starch digestibility has been reported. As such, it remains unclear of the role of MMT peel on glycaemic response. The present work thus sought to obtain data which will help to address these knowledge gaps, with an aim to develop formulated MMT peel powder besides examining its organoleptic properties, hypoglycaemic effect, and *in vitro* starch digestibility.

Materials and methods

Development of formulated MMT peel powder

The MMT peel powder was produced following Ong *et al*. (2021a). Various trials were carried out to formulate the MMT peel powder. Finally, the MMT peel powder was formulated with different monk fruit [*Siraitia grosvenorii* (Swingle) C. Jeffrey ex A.M. Lu & Zhi Y. Zhang; also known as *luo han guo* in Chinese] sweetener ratios as shown in Table 1 (Djarot and Badar, 2017). Monk fruit sweetener is a combination of monk fruit extract (30 - 40%) with erythritol (60 - 70%) that offers a sweet taste like that of sugar with significantly lower calorie (0.02 kcal/g) as compared to other sugar-based sweeteners (Ellis, 2019). Carboxymethyl cellulose (CMC), citric acid, and orange flavouring were added to the formulations, and thoroughly mixed for uniform distribution of ingredients. CMC was used to improve the viscosity of a liquid without considerably altering the other properties, and also to enhance the suspension of other ingredients or emulsions, which subsequently increases the stability of the formulations (McClements, 2015), whereas citric acid acts as a flavour enhancer, preservatives, and antioxidants (Mirza *et al*., 2017). Next, flavouring agent was used to enhance the flavour of the formulations. Formulations 0, 1, 2, and 3 represented 100, 60, 50, and 40% MMT peel powder, respectively. The formulated MMT peel powder was then kept in an airtight container in a freezer (-21°C) for further analyses.

Table 1. Formulation of MMT peel powder.

Ingredient (%w/w)	Formulation 0	Formulation 1	Formulation 2	Formulation 3
MMT peel powder	95	57	47.5	38
Monk fruit sweetener	0	38	47.5	57
CMC	1	1	1	1
Citric acid	2	2	2	2
Orange flavouring	2	2	2	2

Organoleptic evaluation

The sensory evaluation of the samples was conducted by 65 untrained panellists recruited from the Faculty of Bioresources and Food Industry, Universiti Sultan Zainal Abidin, Malaysia using the seven-point hedonic scale. The samples were prepared in identical containers coded with a 3-digit random numbers, and each sample was presented with a different number. The randomised order of the sample was presented once at a time to each panellist. The samples were served fresh on the test day by dissolving in water, in a ratio of 1 g:6 mL. Panellists were asked to evaluate the samples for the sensory parameters such as colour, flavour/aroma, taste, mouth feel/texture, and overall acceptability based on their degree of liking; 1 (dislike very much), 2 (dislike moderately), 3 (dislike slightly), 4 (neither like nor dislike), 5 (like slightly), 6 (like moderately), and 7 (like very much). In between each sample, the panellists were allowed to cleanse their palate (Ho and Dahri, 2016).

In vitro hypoglycaemic effect

Glucose adsorption capacity

The experiment was conducted following Ahmed *et al.* (2011). The samples (250 mg) were mixed with 25 mL of glucose solution with increasing concentrations (5, 10, 50, and 100 mM), and stirred well, followed by incubation in a water bath shaker (Memmert, Schwabach, Germany) with continuous shaking (108 rpm) at 37°C for 6 h. After that, the mixture was centrifuged at 3,000 *g* for 10 min, and the glucose content in the supernatant was determined using a glucose oxidase-peroxidase (GOPOD) assay kit (Randox Laboratories Ltd., UK). Bound glucose (mM/g) was calculated using Eq. 1:

$$\text{Bound glucose (mM/g)} = (G1 - G6) / \text{sample weight (g)} \times \text{solution volume (L)} \quad \text{(Eq. 1)}$$

where, G1 = glucose concentration of original solution, and G6 = glucose concentration after 6 h of incubation.

In vitro residual α-amylase activity

The residual α-amylase activity was determined following Cañas *et al.* (2020) by mixing 250 mg of the sample with 250 mg of α-amylase and 25 mL of potato starch solution (4%, w/v). The mixture was incubated (37°C, 30 min), then 20 mL of 0.1 N sodium hydroxide (NaOH) was added to terminate the α-amylase activity, and later centrifuged (3,000 *g*, 10 min). Finally, the glucose content was evaluated using the GOPOD assay kit as earlier described.

In vitro starch hydrolysis

Measurement of total starch content

The determination of total starch (TS) content was performed following Sęczyk *et al*. (2016). Approximately, 100 mg of sample was added to 6 mL of 2 M potassium hydroxide (KOH) solution with continuous magnetic stirring for 30 min at room temperature. Then, 3 mL of 0.4 M sodium acetate buffer (pH 4.75) was added to the mixture. The samples were hydrolysed to glucose by the addition of 100 µL of amyloglucosidase (20 U/mL), and incubated at 60°C for 45 min in a constant shaking water bath. Samples were then centrifuged at 3,000 *g* for 10 min. Next, 20 µL from each sample was transferred into a new test tube, and 2 mL of GOPOD was added. Samples, blank, and glucose standards were incubated at 37°C for 30 min. Absorbance was read at 500 nm to determine the glucose content in each sample. Then the glucose content was measured using the GOPOD assay kit as earlier described. Finally, a factor of 0.9 was used to convert the reducing sugar content to TS content.

Measurement of free glucose content

The procedure was carried out following Ng *et al.* (2017). Briefly, the samples (3 g) and 25 mL of acetate buffer (0.1 M, pH 5.2) were added into a 50-mL centrifuge tube, and incubated in a boiling water bath (Memmert, Schwabach, Germany) for 30 min with continuous shaking. The tubes were then cooled to 37°C for 30 min and 0.2 mL of invertase was added. The mixture was then incubated in a shaking water bath at 37°C for 30 min. Following incubation, each sample (1 mL) was pipetted into a test tube comprising 2 mL of 96% ethanol. Each sample tube was centrifuged at 3,000 *g* for 10 min, and the free glucose (FG) content was determined using the GOPOD assay kit as earlier described.

In vitro starch digestibility and estimated glycaemic index (eGI)

In vitro starch digestibility was assayed following Zeng *et al.* (2019). Approximately 100 mg of samples were put into a test tube, and mixed with 15 mL of sodium acetate buffer (0.2 M, pH 5.2). The

mixture was incubated at 37°C with continuous shaking for 5 min. Then, the hydrolysis of carbohydrates was initiated by the addition of 150 µL of mixed enzyme solution containing 75 µL of α-amylase (35 U/mL) and 75 µL of amyloglucosidase (23 U/mL) at 37°C. An aliquot sample (100 µL) was taken and transferred into a new tube, and 2 mL of absolute ethanol was added to stop the enzymatic hydrolysis. This step was repeated at 10, 20, 30, 60, 90, 120, 150, and 180 min. Each tube was centrifuged at 3,000 *g* for 10 min, and the % starch hydrolysis was determined using the GOPOD assay kit as earlier described. Finally, a factor of 0.9 was used to convert the reducing sugar content to starch content.

The percentage of starch hydrolysis of each sample over time was plotted in a graph, and the area under the curve (AUC) was calculated using the trapezoidal rule. The hydrolysis index (HI) was determined from the percentage of the AUC of the sample to the AUC of the reference glucose. The estimated glycaemic index (eGI) was then calculated using the formula 39.71 + 0.549HI (Chusak *et al.*, 2018).

Measurement of nutritionally important starch fractions

Nutritionally important starch fractions based on the hydrolysis rate are identified as rapidly digestible starch (digested within 20 min; RDS), slowly digestible starch (digested between 20 and 120 min; SDS), and resistant starch (undigested after 120 min; RS), which were calculated using Eq. 2 – 4 (Ng *et al.*, 2017):

$$RDS\ (g/100\ g) = S20 - FG \quad \text{(Eq. 2)}$$

$$SDS\ (g/100\ g) = S120 - S20 \quad \text{(Eq. 3)}$$

$$RS\ (g/100\ g) = TS - (RDS + SDS) \quad \text{(Eq. 4)}$$

where, S20 = the starch content after conducting starch hydrolysis for 20 min, S120 = the starch content after conducting starch hydrolysis for 120 min, FG = the free glucose content in starch before conducting enzymatic hydrolysis, and TS = the total starch content in the sample.

Statistical analysis

All data were stored and analysed using the IBM SPSS Version 21.0 at a significance level of $p \leq 0.05$. Results were expressed as mean ± SD, or median (interquartile range). One-way analysis of variance (ANOVA) was performed, and significant differences between the mean values were determined using the Tukey's *post-hoc* test. Meanwhile, the Friedman test followed by Wilcoxon signed ranks test were performed for sensory evaluation analysis.

Results and discussion

Organoleptic evaluation

The results of the sensory evaluation of different formulated MMT peel powder are shown in Table 2. Overall, the different sensory attributes rated by the panellists ranged from '3' (dislike moderately) to '5' (like slightly). Statistically significant differences ($p < 0.001$) existed for all sensory attributes, except colour. All the panellists rated the colour of all samples as 'neither like nor dislike' from 4.0 (2.0) to 5.0 (2.0). In the formulation, no colouring agent was used, hence the colour of the samples was solely derived from the yellow colour of the MMT peel. Colour is one of the more critical quality parameters of dried fruits which can be influenced by

Table 2. Sensory evaluation of formulated MMT peel powder.

Attribute	Formulated MMT peel powder[1]			
	Formulation 0	Formulation 1	Formulation 2	Formulation 3
Colour	4.0 (2.0)^{a}	5.0 (2.0)^{a}	5.0 (2.0)^{a}	5.0 (2.0)^{a}
Flavour/aroma	4.0 (2.0)^{a}	4.0 (2.0)^{ab}	4.0 (2.0)^{b}	5.0 (2.0)^{c}
Taste	3.0 (3.0)^{a}	4.0 (2.0)^{b}	4.0 (2.5)^{b}	5.0 (2.0)^{c}
Mouth feel/texture	3.0 (2.0)^{a}	4.0 (2.0)^{b}	4.0 (2.0)^{c}	5.0 (2.0)^{d}
Overall acceptability	3.0 (2.0)^{a}	4.0 (1.0)^{b}	4.0 (3.0)^{c}	5.0 (2.0)^{d}

Values are median (interquartile range) (n = 65). Values followed by different lowercase superscripts within a row indicate statistically significant among the group at level $p \leq 0.0083$, tested using the Friedman test followed by Wilcoxon signed ranks test. [1]Formulation 0/1/2/3: MMT peel powder with 0/40/50/60% sweetener.

the Maillard reaction which may lead to the generation of browning reaction's components during peel drying. Also, it is also possible that the colour variation is caused by the destruction and degradation of carotenoid and flavonoid pigments responsible for the yellow MMT peel colour (Akubor and John Ike, 2012).

Formulation 3 had a significantly higher flavour score as compared to Formulations 0, 1, and 2. Formulation 2 also had a significantly higher flavour score than Formulation 0. There was no significant difference in flavour score between Formulations 1 and 2, as well as between Formulations 0 and 1. Flavour is a dominant determinant of human consumption behaviour (Forestell, 2017). This is because food odours and flavours are considered to exhibit taste-like characteristics (Ramaekers *et al.*, 2014) which can stimulate the brain triggering reward processes and manipulate subsequent food choices (Han *et al*., 2020). Similarly, it was also proved that food odours are potent appetitive signals that are intrinsic to the hedonic feature of foods, which are critical for food choices (Boesveldt and de Graaf, 2017).

Next, by looking at the sensory attributes namely taste, only Formulations 1 and 2 showed no significant difference, whereas significant difference was observed among all formulations for mouth feel and overall acceptability. Median mouth feel, taste, and overall acceptability rating for Formulation 3 was significantly higher as compared to Formulations 2, 1, and 0. Meanwhile, Formulations 1 and 2 had a significantly higher score than Formulation 0 for mouth feel, taste, and overall acceptability. Formulation 3 received the highest median scores for all the sensory attributes of 5.0 (2.0). Conversely, the lowest median score for colour, flavour, mouth feel, taste, and overall acceptability were observed in Formulation 0 [4.0 (2.0), 4.0 (2.0), 3.0 (3.0), 3.0 (2.0), and 3.0 (2.0), respectively]. Comparative analysis of sensory attributes of the different formulations revealed that Formulation 3 scored high in the hedonic scale for most attributes, thus indicating its overall acceptability which was rated as 'like slightly'.

A product is considered acceptable if its overall acceptability receives an average rating of greater than 4 ('neither like nor dislike') (Ho *et al*., 2013). Therefore, Formulations 1, 2, and 3 were considered acceptable with rating ranging from 4.0 (1.0) to 5.0 (2.0), whereas Formulations 0 was considered not acceptable with a rating of 3.0 (2.0). However, all formulations were found to be superior to Formulation 0 in all sensory attributes, in which Formulation 3 > Formulation 2 > Formulation 1 > Formulation 0. This could have been due to the fact that usually fibre-rich foods or fibre addition to meals are less palatable (Ruhee, 2018). In the present work, Formulation 0 had the highest MMT peel content which was the least palatable. The ratings of the different samples by the panellists could have been influenced by different backgrounds, and possible prior exposure to similar samples (Obilana *et al*., 2018).

All the sensory attributes score increased with a decrease in the MMT peel powder substitution with sweetener. These results agreed with other research which reported that formulation with 5 g of mango peel powder demonstrated the best sensory attributes in terms of flavour, taste, and mouth feel as compared to 10 and 15 g. This could have been attributed to the increase of acidity among the different formulations (Ahmed *et al*., 2020). Interestingly, the most obvious difference between the present work and Ahmed *et al.* (2020) was that the present work utilised a greater amount of MMT peel (16.7 to 25 g/250 mL for formulations, while only 5 to 15 g/250 mL were observed in the other work). Therefore, the overall acceptance of formulated MMT peel among panellists was slightly lower as compared to the other work. In the present work, panellists preferred Formulation 3 which had a higher proportion of sweetener over other samples. This finding agreed with another work which claimed that a high sweet beverage was preferred over a low sweet beverage (Gous *et al*., 2019).

In vitro hypoglycaemic effect

The present work investigated the formulated MMT peel powder on glucose-lowering effect *in vitro*. Concerning glucose adsorption, the formulated MMT peel powder was able to retain glucose in its structure. Figure 1 demonstrates that all formulated MMT peel powder could adsorb glucose at all concentrations (5 to 100 mM), in which, a greater amount of glucose was bound with elevated glucose concentration. The results also showed that the samples could bind glucose even at a lower glucose concentration of 5 mM, hence minimising the amount of glucose available for transport across the intestinal

membrane and subsequent postprandial hyperglycaemia alleviation (Ahmed *et al.*, 2011). The findings agreed with previous studies performed on different samples such as coffee pulp (Cañas *et al.*, 2020), as well as wheat bran, guar gum, oats, barley, and psyllium husk (Ahmed *et al.*, 2011). Formulation 0 had a significantly greater glucose adsorption capacity as compared to the three formulations at a concentration of 5, 10, and 100 mM. At 50 mM, a significantly greater glucose adsorption capacity was observed in Formulation 0 as compared to Formulation 3. However, there was no significant difference in glucose adsorption capacity in all three formulations across the glucose concentration ranged from 5 to 100 mM.

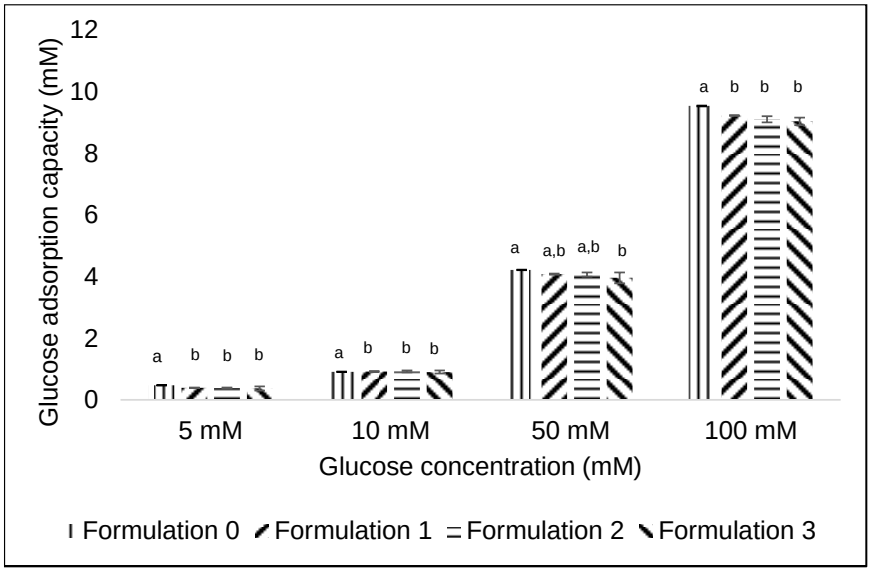

Figure 1. Glucose adsorption capacity of different samples at various glucose concentrations. Values with different lowercase letters within each concentration indicate statistically significant among the group at level $p \leq 0.05$.

The higher glucose adsorption capacity of the formulated MMT peel powder might have been attributed to their dietary fibre content as various studies reported that both insoluble and soluble fibre from diverse sources (wheat bran, oat, barley, psyllium husk, xanthan gum, and guar gum) can adsorb glucose (Ou *et al*., 2001; Ahmed *et al.*, 2011). Insoluble fibre can suppress glucose diffusion in the small intestine by adsorption or inclusion of the smaller sugar molecule within the structure of the fibre particles. In contrast, soluble fibre forms a viscous gel in an aqueous solution which can delay the delivery of glucose across the small intestine's absorptive epithelium, thus lowering postprandial glycaemic response (Ahmed *et al.*, 2011). The effectiveness of dietary fibre in manipulating hyperglycaemia is usually influenced by its composition, source, and preparation (Ahmed *et al*., 2011).

Table 3 demonstrates that the addition of MMT peel powder exerted an impact on glucose reduction following amylase digestion. Formulation 0 had a significantly lower glucose production as compared to the three formulations, which indirectly implied that Formulation 0 exhibited superior inhibition of the α-amylase activity. On the other hand, no significant difference was observed between the three formulations.

The presence of MMT peel reduced glucose production with subsequent restriction of glucose diffusion by inhibiting α-amylase activity. One of the mechanisms is that the dietary fibre can be adsorbed to starch in the food, which may interfere with the starch hydrolysis by α-amylase by reducing the starch accessibility to the enzyme, and act as a physical barrier between starch and α-amylase, by adsorption of the enzyme on fibre which inhibits the enzymatic reaction between starch and α-amylase (Ou *et al.*, 2001). Furthermore, dietary fibre (insoluble fibre such as cellulose) may disrupt the endogenous digestive enzymes by physical entrapment and binding, which suppress their activity and reduce their availability to digest starch (Bhattarai *et al.*, 2017). Next, dietary fibre (soluble fibre such as gum) can reduce the gut transit time which minimises starch digestion in the small intestine, and they are

moved to the large intestine quickly (Meng *et al.*, 2019). Also, soluble fibre can increase the viscosity of content in the small intestine, which is key to the glucose liberation rate and absorption, as well as hamper the accessibility of digestive enzymes to the starch (Repin *et al.*, 2018).

Table 3. Effect of different samples on the α-amylase activity.

	Formulated MMT peel powder[1]				
	Formulation 0	**Formulation 1**	**Formulation 2**	**Formulation 3**	**Blank[2]**
Glucose produced (mg/mL)	0.42 (0.01)^{a}	0.49 (0.02)^{b}	0.52 (0.03)^{b}	0.54 (0.01)^{b}	0.92 (0.04)^{c}

Values are mean (standard deviation) (n = 3). Means followed by different lowercase superscripts within a row indicate statistically significant among the group at level $p \leq 0.05$. [1]Formulation 0/1/2/3: MMT peel powder with 0/40/50/60% sweetener. [2]Blank: without the addition of MMT peel powder.

Total, resistant, and digestible starch contents

Figure 2 compares the TS, RS, and DS contents of different formulations of MMT peel powder. The TS content of the formulated MMT peel powder increased with increasing amount of MMT peel powder incorporated into the formulation. A significant difference was observed among all the formulations, in which Formulation 0 had the highest TS content as compared to the other three formulations. In terms of nutritional perspective, the starch in food is typically divided into three categories namely RDS, SDS, and RS (Englyst *et al.*, 2007). RDS is readily and fully degraded in the small intestine which is related to postprandial blood glucose elevation within the first 20 min. SDS is comprehensive starch digestion in the small intestine with a slow rate ranging from 20 to 180 min. RS is the starch that resists digestion in the small intestine, and may be fermented in the large intestine (Chusak *et al.*, 2018). *In vitro* means, RS can be defined as starch which is indigested after 180 min during *in vitro* digestion (Warren *et al.*, 2015). In the present work, significantly higher RS content was observed in Formulation 0 followed by Formulations 1, 2, and 3. A study claimed that a high dietary fibre intake content may aid starch re-association, and boost the RS content even further (Li *et al.*, 2020), as observed in the present work. On the other hand, Formulation 0 had the highest digestible starch (DS) content as compared to the rest of the formulations. Formulation 1 had a significantly higher DS content than Formulation 3, but was not significantly different from Formulation 2. Meanwhile, no significant difference was observed between Formulations 2 and 3.

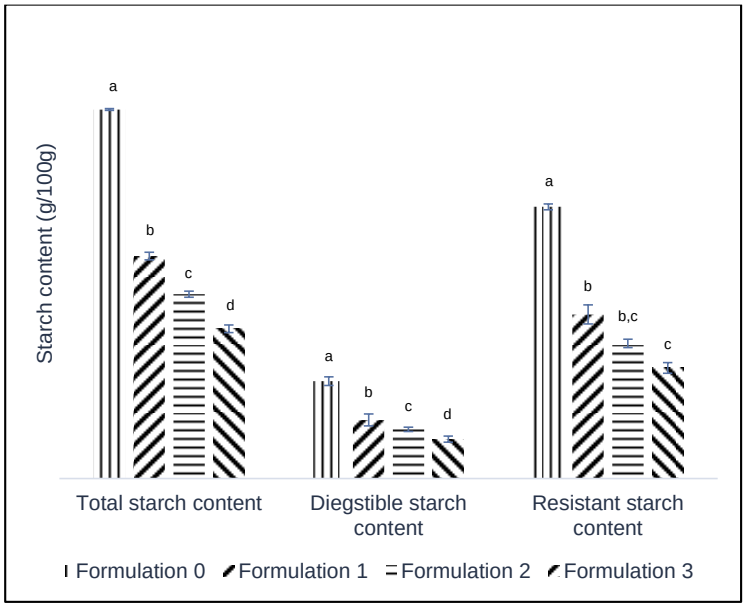

Figure 2. Total, digestible, and resistant starch contents of the formulated MMT peel powder. Values with different lowercase letters indicate statistically significant among the group at level $p \leq 0.05$.

In vitro starch digestibility, hydrolysis index, and estimated glycaemic index

Figure 3 displays the rate of *in vitro* starch digestion of formulated MMT peel powder in comparison with reference (glucose). All formulated MMT peel powder had a lower AUC as compared to the reference. This indicated that reference had a faster digestion rate than formulated MMT peel powder. The lower digestibility of formulated MMT peel powder with decreasing levels of MMT peel powder might have been due to lower DS content. Consequently, lower glucose was released, and lower eGI can be attained (Perry and Ying, 2016). This result agreed with previous studies which demonstrated flour with higher RDS content had considerably higher HI and eGI levels (Chusak *et al.*, 2018).

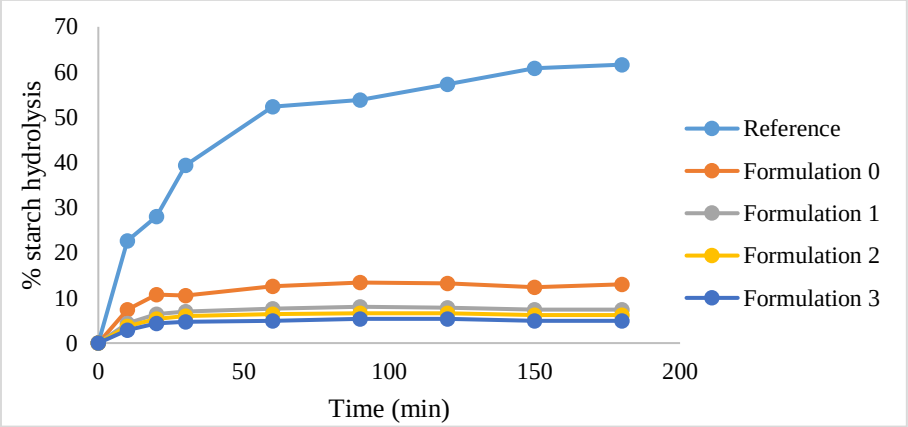

Figure 3. *In vitro* starch hydrolysis curve of formulated MMT peel powder.

It can be seen from the data in Table 4 that there was a significant difference of HI and eGI across all the formulations, in which Formulation 3 had the lowest HI and eGI. The addition of MMT peel powder into the formulation led to increased HI and eGI in a directly proportional manner. The eGI of all formulated MMT peel powder ranged from 45.19 (0.14) to 53.32 (0.27), which implied that they fell within the low GI food classification. The lower GI of formulated MMT peel powder in the present work might have been attributed to the low starch content, especially digestible starch and the presence of dietary fibre. Dietary fibre does not increase the glycaemia level (Krawęcka *et al.*, 2019) which allows for better control of postprandial glucose liberation (Scazzina *et al.*, 2016). Moreover, other ingredients used in the formulation might also play a crucial role in providing a special effect in lowering eGI of the formulated MMT peel powder via synergistic means. The ingredient of interest is monk fruit sweetener. It is a natural non-nutritive sweetener with low GI (Pandey and Chauhan, 2020).

The eGI is associated with the parameters of the digestible starch fractions such as RDS and SDS. Evidence showed that SDS is the main contributing factor to GI (Meynier *et al.*, 2015). On the other hand, consumption of diet with high RDS level can stimulate a rapid hyperglycaemic response followed by glucose-induced insulin secretion from pancreatic β-cells (Chusak *et al.*, 2018). Study stated that RS is a vital index for starch digestibility measurement which influences the GI (Saidu *et al.*, 2017).

Table 4. Hydrolysis index and estimated glycaemic index of the formulated MMT peel powder.

Formulated MMT peel powder[1]	Formulation 0	Formulation 1	Formulation 2	Formulation 3
Hydrolysis index (%)	24.79 (0.48)^{a}	14.90 (0.35)^{b}	12.45 (0.29)^{c}	9.98 (0.26)^{d}
Estimated glycaemic Index	53.32 (0.27)^{a}	47.89 (0.19)^{b}	46.55 (0.16)^{c}	45.19 (0.14)^{d}

Values are mean (standard deviation) (n = 3). Means followed by different lowercase superscripts within a row indicate statistically significant among the group at level $p \leq 0.05$. [1]Formulation 0/1/2/3: MMT peel powder with 0/40/50/60% sweetener.

Interestingly, RS exhibits functional characteristics which can delay postprandial glucose (Slavin, 2013), and enhance postprandial insulin (Ells *et al.*, 2005). In contrast, another study by Xiong *et al.* (2018) proposed that the digestive enzyme accessibility instead of structural characteristics of starch modulates the glycaemic response.

Conclusion

Formulation 3 was the most acceptable for consumption with the weakest *in vitro* hypoglycaemic effect. Besides, it had the lowest TS, DS, and RS contents, as well as HI and eGI. In contrast, Formulation 0 yielded the complete opposite for the mentioned parameters. These suggested that formulated MMT peel powder could have the potential to be used in blood glucose control.

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