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Key properties of iodine-, iron- and zinc- fortified fish cracker: effects of ambient shelf storage on iodine retention and quality indicators

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Summary Multi-micronutrient (MN)-fortified fish cracker could be used as a means to improve iodine, iron (Fe) and zinc (Zn) status. We evaluated iodine stability as well as the quality and sensory properties of MN-fortified fish cracker, at the fortification levels of 5.3 mg per 100 g for Fe and Zn and 260 μ g per 100 g for iodine in the dough. On average, the overall retention of iodine after processing and storage for 4 months was ~65%. Fortification with KIO₃ + ZnO, or KIO₃ + ZnO + ferrous fumarate, significantly increased the loss of iodine during processing. The Fe compounds tended to promote iodine instability during storage and contributed to thiobarbituric reactive substances. Fortification did not affect linear expansion of the cracker. Triple-fortified fish cracker with KIO₃, ferric pyrophosphate and ZnO exhibited both high iodine retention (92.7% for steaming + drying and 72.7% for storage) in the tropical conditions ($30 \pm 1 \,^{\circ}$ C, $80 \pm 5\%$ R.H.) and achieved overall desirable sensory scores. Thus, such fortification of fish cracker might successfully supplement iodine, zinc and iron, while at the same time contributing to the palatability of cracker.

Keywords Fish cracker, fortified cracker, iodine stability, iron, linear expansion, zinc.

Introduction

Iron (Fe), zinc (Zn) and iodine deficiencies are global health problems, prevalent not only in developing countries but also in the developed world. Iodine deficiency disorder (IDD) (de Benoist et al., 2008) and Zn deficiency (IZiNCG, 2004) currently affect more than 2000 million people, and globally over 30% of the population is anaemic, mainly with Fe deficiency (WHO, 2008). These micronutrient deficiencies impair physical and cognitive development and lower immune competence, as well as increasing mortality and morbidity rates in children. In addition, the individual often suffers from multiple deficiencies (Winichagoon, 2008). In the less-developed countries, these deficiencies have serious health consequences, especially in women of reproductive age and in young children, with large impacts on economy and national development. The latest Thai national nutrition survey (Ministry of Public Health, 2006) reported that around 20-50% of pregnant women and young children suffered from anaemia. National surveillance data for control-

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ling IDD showed that 59% of Thai pregnant women had urinary iodine levels below the 150 μ g L⁻¹ threshold level that defines deficiency (Bureau of Nutrition, 2009). Prior studies have found Zn deficiency in 60% (Thurlow *et al.*, 2006) and in 50% (Pinkaew *et al.*, 2013) of school-aged children, in the north-eastern and southern parts of Thailand, respectively.

Food fortification is recognised as the most economical approach to combat micronutrient deficiencies because of its relatively low cost and long-term sustainability (WHO/FAO, 2006). Multifortification has become more attractive since the recognition that coexisting nutrition deficiencies can reduce the beneficial impact of a single nutrient supplement or fortification in improving nutritional status and reducing morbidity (Winichagoon, 2008).

Salt iodisation is a well-known efficient strategy for combating IDD (Chanthilath *et al.*, 2009), and there have been attempts to fortify salt with other micronutrients such as vitamin A, Fe, Zn, calcium and folic acid (Lotfi *et al.*, 1996). However, it is technically challenging to fortify salt or other foods with highly bioavailable Fe compounds and iodine, as iodine is unstable and has undesirable organoleptic properties (Diosady *et al.*, 2002a; Chavasit *et al.*, 2003; Wegmüller *et al.*, 2003). In both developing and developed countries, the major source of energy is cereal-based products, so it seems difficult to satisfy the iodine requirements without supplementation (Thomson, 2009; Longvah *et al.*, 2012), especially when low salt consumption is pursued due to health concerns. These factors contribute to the high prevalence of IDD (Winger *et al.*, 2008). Therefore, finding an appropriate food vehicle for iodine fortification is desirable.

Fish cracker is a popular snack commonly consumed in South-East Asia (SEA), and its consumption is growing annually (Karrila, 2011). In Thailand, fish cracker is the most popular in the southernmost provinces. Large amounts of fish cracker produced are distributed within Thailand, as well as exported to neighbouring Malaysia and other countries (Chaimongkol, 2012). Fish cracker is lightweight and has a relatively long shelf life as a half-finished intermediate product (IP). Its affordable price is another advantage as a potential food vehicle. Therefore, the fortification of Fe, Zn and iodine in fish cracker could help improve the nutritional status of people at risk, especially in SEA. However, one of the main problems in fortifying Fe and iodine is that they are both very reactive, and iodine is volatile, so it can easily be lost during processing and storage. Iodate is a strong oxidising agent, while iodide is a strong reducing agent, so iodine compounds can participate in redox reactions in general. They can react in food and cause colour changes, increase oxidative reactions, reduce shelf life and decrease the bioavailability of iodine itself as well as other nutrients (Winger et al., 2008).

The objective of this study was to examine the effects of iodine, Fe and Zn fortification in fish cracker on iodine stability during processing and up to 4 months of storage, as well as determine the physical and chemical properties, and on sensory acceptability of fortified fish cracker stored for 3 months in tropical ambient conditions.

Materials and methods

Materials

Fresh rainbow sardines (*Dussumieria elopsoides*) were purchased from Pattani fish market, Thailand, and transported in an ice box to the Department of Food Science and Nutrition, Prince of Songkla University, Pattani campus, for sample preparation. The other fish cracker ingredients, namely tapioca flour (Lion brand, Thailand), salt, pepper and garlic, were obtained from a local market. The Fe, Zn and iodine (analytical grade) compounds used to fortify fish cracker were as follows: ferrous fumarate (FF), ferric pyrophosphate (FPP) (Sigma, Schnelldorf, Germany), zinc oxide (ZnO) and potassium iodate (KIO₃) (Univar[®], Ingleburn, NSW, Australia).

Sample preparation

The formulation for the nonfortified control samples contained 41.4% rainbow sardine, 41.4% of tapioca flour, 1.2% of salt, 1.4% of pepper, 2.1% of garlic and 12.5% of water, by fresh or 'as is' weight. The fortification levels per 100 g were approximately 260 μ g for iodine and 5.3 mg for Fe and Zn. Three batches of fish cracker product (2 kg per batch) were produced with each of six fortification treatments, namely KIO₃, KIO₃ + ZnO, KIO₃ + FF, KIO₃ + FPP , KIO₃ + ZnO + FF and KIO₃ + ZnO + FPP. In addition, three batches of control were produced. The steps of preparation are described below.

The spices, pepper and garlic, were ground with salt in a mortar to produce a paste. The fish were beheaded, gutted and washed with cold water. Then, they were chopped and homogenised for 30 min in a high-speed cutter (Cuttex M11N, Bayern, Germany), along with the obtained paste, to obtain fish mince. In the meantime, tapioca flour was mixed with the Fe and Zn compounds in a cube mixer for 20 min. An iodine solution was prepared by dissolving KIO_3 in deionised (DI) water. Then, the tapioca flour was kneaded with the fish mince in a mixer (Sinmax mixer, Bangkok, Thailand) for 5 min, before adding the iodine solution. The mixing was continued for another 25 min, producing dough with a smooth appearance. The dough (0.5 kg) was then rolled by hand to a cylindrical shape ~5 cm diameter and steam-cooked for 40 min. After cooling overnight in a refrigerator, the cooked rolls were sliced to 1.5 mm thickness with a mechanical slicer (Thai wasino, Bangkok, Thailand). To produce the IP, the sample chips were then dried with a tray dryer (Kluaynamthai, Bangkok, Thailand) at 60 °C until the moisture content of the samples decreased to $10 \pm 1\%$. The IP chips were collected and packed in high-density polyethylene bags with heat sealing (airtight) before storage (at 30 ± 1 °C, $80 \pm 5\%$ R.H.) for later use. The fish cracker-finished product (FP), that is ready for human consumption, was prepared by deep frying the IP in commercial palm oil, fresh out of the bottle, at 180-190 °C for 1 min, using an electric fryer (JAZE DF-81, Guangdong, China). The above laboratory steps in the preparation of IP and FP are closely similar to those used by the cottage industry in southern Thailand in the commercial production of fish cracker.

Iodine content

Retention of iodine was determined during production of the IP, and monthly during storage in tropical conditions for up to 4 months. The iodine content was determined by inductive coupled plasma spectrometer mass spectrometer (ICP-MS), according to Julshamn *et al.* (2001). The IP was ground to 40–80 mesh before weighing 1.0 g of a sample in a screw cap tube, to which 25 mL of DI water was added and mixed using a vortex mixer. The sample suspension was centrifuged at 867 g for 15 min, at room temperature. The supernatant was collected and filtered through a micromembrane of 0.45 micron pore size. The iodine content in the filtrate was determined with ICP-MS, using a standard curve generated with iodine standard samples.

Thiobarbituric reactive substances (TBARS)

The TBARS values of the control and the fortified fish crackers were determined twice during storage, at 0 and 3 months, both for the IP and after frying to FP. The TBARS was determined according to Egan et al. (1981). Ten grams of sample was homogenised with DI water and then transferred to a 250 mL round-bottom flask. Another 47.5 mL of DI water was used for rinsing the homogeniser and pooled into the same round-bottom flask before adding 2.5 mL of 4 N HCl. After adding 0.5 mL of antifoaming agent, the flask was attached to a distillation column and 50 mL of distillate was collected. A 5 mL aliquot of the distillate was mixed with 5 mL of TBA reagent, and the mixture was heated in a boiling water bath for 30 min. After cooling, the absorbance was determined at 538 nm. DI water was used as the blank. The result was calculated as follows:

TBARS value (mg malonaldehyde per kg sample) = $7.8 \times A_{538}$.

 A_{538} is the absorbance at 538 nm.

Linear expansion

The linear expansion (LE) occurring during puffing of the IP to the FP by frying in hot oil was determined for the control and fortified fish crackers after 1, 2 and 3 months storage. Each IP sample chip was marked with three lines across its planar face (ten replicates). Each line length was measured before and after frying that causes the puffing. The percentage of LE was calculated according to the method recommended by Yu (1991).

$$LE(\%) = \frac{\text{length after puffing} - \text{length before puffing}}{\text{length before puffing}} \times 100.$$

Colour

The colour of the control and the fortified samples during storage was measured both for the IP and the FP samples at 0 and 3 months using a colorimeter (Hunter Lab; Cokor Quest XE, Reston, VI, USA). The colour was expressed as lightness (L^*) , redness

 (a^*) and yellowness (b^*) . The equipment was standardised with black and white colour standards. The colour differences are expressed as ΔE_{ab} defined by:

$$\Delta E_{ab} = \sqrt{\left(\Delta L\right)^2 + \left(\Delta a\right)^2 + \left(\Delta b\right)^2},$$

where ΔL , Δa and Δb are the colour coordinate differences between the fortified fish cracker and the unfortified control sample, compared at the same storage time. Fifteen samples were used for each treatment.

Moisture

The moisture content was analysed according to an AOAC method (2000) by drying a sample in a hot-air oven at 105 °C until it showed a constant weight. The same samples that were used for determining iodine contents were used in these determinations.

Preliminary consumer study

Twenty-five trained panellists were asked to evaluate the FP products, both control and fortified fish crackers, after 3 months of storage as IP and preparation of FP immediately before consumption. The quality profile included colour, porosity, fish smell, crispness, taste and overall liking, with 9-point hedonic scale varying from 1 'dislike extremely', through 5 'neither like nor dislike', to 9 'like extremely'.

Statistical analysis

Data from the three batches of each treatment were pooled and presented as mean \pm SD for % iodine retention, TBARS, LE, ΔE , L^* and sensory scores. The retention of iodine during processing and at storage times of 1, 2, 3 and 4 months as well as the other physical and chemical properties was compared using one-way ANOVA with Duncan's test as the *post hoc* test. *P* values < 0.05 were considered significant.

Results and discussion

Stability of iodine during production and storage

Retention of iodine after production and storage for up to 4 months (based on the uncooked dough)

Figure 1 presents graphically the retention of iodine in the IP samples of variously fortified fish cracker, immediately after drying of the chips. Among six treatments, retention was lowest when fortified with $KIO_3 + FF + ZnO$ (86.0 ± 4.9%) and highest when fortified with KIO_3 alone (97.8 ± 1.3%). The average retention due to production across all fortifications was 91.5 ± 5.4%. While the iodine retentions with dual and triple fortifications were lower than with KIO_3 alone, statistically significant differences were found only with $KIO_3 + ZnO$ and $KIO_3 + FF + ZnO$.

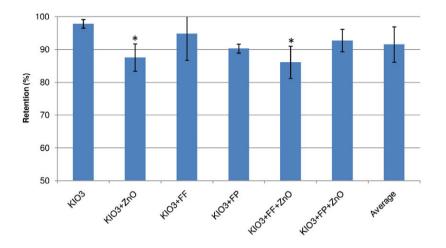


Figure 1 Retention of iodine in dough through steaming and drying to an intermediate product (IP). Fish cracker with various fortifications that are indicated by the labels. *Significantly different from the other treatments (P < 0.05).

Most studies on the stability of iodine have focused either on iodised salt itself or on the replacement of noniodised salt with iodised salt in various foods (Chanthilath et al., 2009), while in the present study, we added various fortificants directly to food vehicles. Therefore, prior data on iodine retention after production of fortified food are relatively limited. A recent study about the effects of various Indian cooking processes, using Indian recipes, showed an average $60 \pm 21\%$ retention of iodine from added iodised salt. The poorest $52 \pm 23\%$ retention was obtained with shallow frying in oil, and the best $82.2 \pm 6.2\%$ retention with pressure cooking (Longvah et al., 2012). The ICP method is widely used to determine iodine content; however, it might cause high variation (as seen in above-mentioned study) due to interfering with others elements. Another study with Indian recipes reported the mean losses of iodine from iodised salt: 6% in roasting; 20% in steaming, pressure cooking or deep frying; 27% in shallow frying; and 37% in boiling (Goindi et al., 1995). Panpipat & Yongsawatdigul (2008) reported that the cooking losses of iodine in freshwater fish sausage fortified with potassium iodide (KI) and omega-3 fatty acid were about 14–16%. The sausages were pre-incubated at 55 °C for 40 min before cooking at 80 °C for 15 min. In the present study, the loss of iodine during processing, namely steaming for 40 min + tray drying at 60 °C for 3-4 h, was $\sim 2-14\%$, which was relatively low compared to the above-mentioned studies. In particular, when fish cracker was fortified with KIO₃ alone, the losses were only ~2%, despite steaming losses of about 20% in prior studies. Several factors may contribute to the good iodine retention we observed in this particular case, and these are now discussed.

The fortificant KIO_3 which was used in the present study is more stable than KI and is recommended by the WHO for using in iodised salts in tropical countries (WHO/FAO, 2006).

Iodine retention may be affected by the food matrix. For example, in the Indian recipes (Longvah et al., 2012), the iodine retention within the same cooking method varied widely depending on the food variety. Chavasit et al. (2002) clearly state that pH, metal ions, ascorbic acid, sugar, various kind of spices and metal utensils affect iodine losses. The content of metal ions (Fe, Zn) in fortified fish cracker could contribute to our observed low retention of iodine in the dual- and the triple-fortified fish cracker. These metal ions can be oxidised and forming iodate by reduction (Chavasit et al., 2002), and this can convert to elemental iodine (I_2) which is readily evaporated at room temperatures, especially above 40 °C (Diosady et al., 2002b). More than seventy reducing agents are known and these can contribute to iodine losses from KIO₃, which is a strong oxidising agent (Winger et al., 2008). In the present study, the loss of iodine during processing to IP was increased when fish cracker was fortified with ZnO and even more when ZnO was combined with FF, which is more soluble than FPP. Results from our preliminary study showed that iodine loss increased by a further ~18% when sun drying was used instead of tray drying (data not shown).

Figure 2 shows that the average overall retention of iodine relative to uncooked dough, after processing and storage of the IP for 4 months was $\sim 64.5 \pm 8.6\%$ and the differences between the fortification treatments were not statistically significant. These results also implied that the iodine content of IP mainly lost during 4 months of storage, so the differences in losses during processing (cooking the dough and drying the IP, Fig. 1) were effectively erased.

For consumption, the IP needs to be fried to form puffed cracker or FP. The loss of iodine during deep frying could be as high as ~ 9% according to our preliminary experiments (data not shown). Using this as an estimate, the iodine retention might be ~ 55% in the FP, when puffed after 4 months of storage as IP.

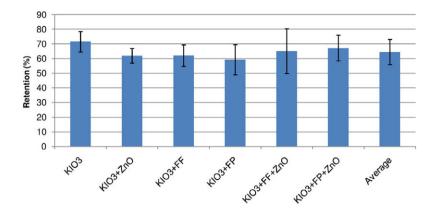


Figure 2 The overall retention of iodine after processing and storage as intermediate product for 4 months relative to uncooked dough in ambient tropical conditions. The labels indicate various fortifications of fish cracker.

When the overall losses of iodine from dough to FP are reliably estimated, an appropriate fortification level for the dough can be determined.

Retention of iodine in IP during 4 months of storage (based on IP at baseline, time 0)

During storage in tropical conditions $(30 \pm 1 \text{ °C},$ $80 \pm 5\%$ R.H.) for 4 months, the retention of iodine relative to IP at baseline decreased gradually in all treatments, as shown in Table 1. The only significant difference within a treatment was detected between 1 and 4 months of IP fortified with KIO₃. At 4 months of storage, the highest $(75.2 \pm 14.0\%)$ retention of iodine was found in triple fortification with $KIO_3 + FF + ZnO$. Dual-fortified fish crackers with FF or FPP had the lowest iodine retentions of $\sim 65\%$ after storage. However, there were no statistically significant differences between treatments at any of the time points. The loss of iodine during storage is partly due to the reaction of KIO₃ with moisture, and the moisture content of fish cracker increased during storage from 9.5% at base line to 10.5% at 4 months. Diosady et al. (1998) suggest that moisture plays a critical role in the stability of iodine. Moreover, the interactions between iodine and Fe also contributed to the loss of iodine in dual-fortified fish crackers, as mentioned before. Several prior studies have reported on dual-fortified salt, or on fish salted with iodine and Fe (Chavasit et al., 2003; Wegmüller et al., 2003; Andersson et al., 2008; Li et al., 2010). Various types of encapsulated and nonencapsulated FF, FPP and ferrous sulphate (FS) have been tested for stability in salt dual fortified with Fe and iodine (Wegmüller *et al.*, 2003). Dual-fortified salt with FS showed rapid losses of iodine and turned unacceptably yellow while salt dual fortified with FPP retained 56–90% of its initial iodine content after 4 months of storage. A new formulation of encapsulated agglomerated FF was tested for the stability and efficacy in India where 20% of iodine was lost over 6 months of storage (Andersson *et al.*, 2008). Recently, 80% iodine retention was reported in a dual-fortified salt (over a year of storage in tropical conditions) when an extrusion process was used to agglomerate the FF powder before coating it with a hydrophilic edible powder (Li *et al.*, 2010).

The comparatively high retention of iodine in triplefortified fish cracker relative to those dual fortified with FF or FPP as observed in the present study be contributed to the dilution effect with ZnO. This may have lessened or prevented the interactions between Fe and iodine in the food matrix, as described in a prior study (Pinkaew *et al.*, 2012).

TBARS values

The TBARS values indicate the secondary products of lipid oxidation, which tend to degrade the aroma and the flavour of a food product (Min & Boff, 2002). The TBARS of IP and FP forms of the fortified fish crackers, at 0 and 3 months of storage, are shown in Table 2. The

Table 1 Retention of iodine (%) during
storage of intermediate product (IP) rela-
tive to the initial iodine content in IP at
time 0

Premix	1 month	2 month	3 month	4 month
KIO ₃	97.5 ± 2.4	95.4 ± 11.9	75.7 ± 11.2	73.1 ± 6.2*
KIO ₃ + ZnO	91.1 ± 6.4	95.3 ± 18.0	$\textbf{67.9} \pm \textbf{8.3}$	71.0 ± 8.1
$KIO_3 + FF$	$\textbf{85.0}\pm\textbf{10.2}$	103.0 \pm 16.3	75.5 ± 7.5	65.4 ± 3.5
$KIO_3 + FPP$	88.5 ± 7.6	89.9 ± 10.9	$\textbf{72.6} \pm \textbf{14.7}$	65.6 ± 11.0
$KIO_3 + ZnO + FF$	94.5 ± 5.6	84.0 ± 10.1	$\textbf{74.2} \pm \textbf{7.0}$	$\textbf{75.2} \pm \textbf{14.0}$
$KIO_3 + ZnO + FPP$	91.0 ± 8.7	94.8 ± 13.2	$\textbf{75.3} \pm \textbf{12.9}$	$\textbf{72.7}\pm\textbf{11.2}$

*Significant difference from 1 month of storage (P < 0.05).

TBARS values increased with storage time for all treatments, for both IP and FP. The control IP at storage times 0 and 3 months had TBARS values comparable to the samples fortified with KIO₃ alone or with $KIO_3 + ZnO$, but significantly lower than the samples with Fe. This suggests that Fe promoted lipid oxidation but Zn did not. This is corroborated by prior results on iodine and omega-3-fortified freshwater fish emulsion sausage, where KI used at 150 µg per 100 g sample had no effect (P > 0.05) on TBARS at any level of omega-3 addition (Panpipat & Yongsawatdigul, 2008). Iron is a pro-oxidant, so that fish cracker fortified with Fe (with or without Zn) tends to have higher TBARS values than the control or the fortified samples without Fe. Although high TBARS values were found in our fish cracker fortified with Fe, differences could not be detected by human testing and frying decreased the values as shown by the results for FP. In a study of fish cracker made from minced fish and kept in vacuum packaging for 90 days, the TBARS value found was $1.38 \pm 0.31 \text{ mg kg}^{-1}$ (Neiva et al., 2011) for the FP, which is similar to our control FP at 3 months of storage.

Linear expansion

An important quality characteristic of cracker is crispness that tends to correlate with the LE, so that LE provides a quantitative laboratory measure closely related to human perception of quality (Yu, 1991; Nurul *et al.*, 2009; Neiva *et al.*, 2011). Consumers tend to prefer fish cracker with a high LE (Nurul *et al.*, 2009). The LEs of our samples are shown, for various storage times of the IP, in Fig. 3. There were no statistically significant differences in the LE between treatments or within a treatment due to storage time. However, it is noticeable that with KIO₃ + ZnO, KIO₃ + FPP and

Table 2 TBARS of the IP and the FP at 0 and 3 months ofstorage, for various fortifications in fish cracker

	TBARS (mg kg ⁻¹)					
	IP		FP			
Premix	0 month	3 month	0 month	3 month		
KIO ₃	$\textbf{2.3}\pm\textbf{0.2}^{a}$	$\textbf{3.1}\pm\textbf{0.8}^{a}$	1.2 ± 0.4^{a}	$2.1\pm0.4^{\text{ab}}$		
KIO ₃ + ZnO	$\textbf{3.1}\pm\textbf{0.4}^{a}$	4.7 ± 0.6^a	1.6 ± 0.5^{ab}	$\textbf{2.6} \pm \textbf{0.8}^{\text{ab}}$		
$KIO_3 + FF$	$8.7\pm0.4^{\rm c}$	9.0 ± 1.3^{b}	$\textbf{3.8}\pm\textbf{0.5}^{c}$	$4.8\pm0.6^{\text{bc}}$		
KIO ₃ + FPP	$6.8\pm0.6^{\rm b}$	$9.5\pm1.1^{ m b}$	3.0 ± 0.6^{bc}	5.7 ± 0.7^{c}		
$KIO_3 + ZnO + FF$	$7.2\pm0.9^{\rm bc}$	8.6 ± 0.8^{b}	$\textbf{2.3}\pm\textbf{0.4}^{ab}$	5.5 ± 1.1^{c}		
$KIO_3 + ZnO + FPP$	8.3 ± 0.2^{bc}	$9.4\pm0.2^{\rm b}$	$\rm 1.6\pm0.4^{ab}$	$4.7~\pm~1.3^{ m bc}$		
Control	1.9 ± 0.2^a	$\textbf{3.5}\pm\textbf{0.3}^{a}$	1.0 ± 0.3^a	1.6 ± 0.5^a		

IP, intermediate product; FP, finished product; TBARS, thiobarbituric reactive substances.

In a single column, different superscripts indicate significant differences (P < 0.05).

 $KIO_3 + ZnO + FPP$, the fish cracker tended to have low LE in the range 50-59%, while the control had LE 59-62%. Interestingly, fish cracker fortified with KIO_3 alone and dual fortified with $KIO_3 + FF$ may have improved expansion relative to control as they consistently provide the highest LE averages. Therefore, KIO₃ and FF may promote LE, while ZnO and FPP may decrease it. It is possible that KIO₃ and FF are more active compounds than ZnO and FPP, neither of which is water soluble, in terms of interacting with the starch gel and preventing its full gelatinisation. This matches prior results on fish cracker enriched with natural calcium sources, namely fish bone powder, small freshwater shrimp and anchovy powder. In that case, adding more calcium lowers LE (Chaimongkol, 2012). Salt (NaCl), on the other hand, improves LE. Adding $\sim 2\%$ salt facilitates the mixing of fish and starch and helps achieve full gelatinisation (Cheow et al., 1999), which improves the LE of cracker. This could explain the positive effects of KIO₃ and FF on LE of fish cracker. However, these findings need to be confirmed in the future.

Colour

Colour differences (ΔE) and lightness (L^*) of fortified fish cracker (both IP and FP forms) are presented in Table 3. Fish cracker can be sold in either FP or IP, but IP is the most popular form as it has a long shelf life and is easier to transport. The ΔE values of IP increased slightly over 3 months of storage as a general rule, with the exception of $KIO_3 + ZnO$. With six treatments and two time points, comparing within IP or within FP, the only statistically significant colour differences were at time 0 in IP. The values of ΔE in IP fortified with KIO₃ + FPP (0.8 \pm 0.03) and with $KIO_3 + ZnO + FF (0.8 \pm 0.1)$ were significantly lower than when fortified with KIO₃ alone (3.7 ± 1.5) . In other words, KIO3 alone affected the colour of IP more than $KIO_3 + FPP$ or $KIO_3 + ZnO + FF$ fortifications. Wegmüller et al. (2003) demonstrated a good correlation between the mean acceptability score for salt and its ΔE value, and the human panellists considered products with $\Delta E < 10$ acceptable in this case. Considering in our data a fixed treatment and time point, ΔE was increased by frying/puffing to FP in general, with the exception of KIO₃ fortification at 3 months of storage. The fortificants used in this study only had an effect on ΔE in the IP but not in the FP.

The L^* of FP products decreased due to the fortificants and this significantly in FP fortified with $KIO_3 + FPP$ at both time points. However, this trend was not detected in IP. Millard browning reaction and caramelisation at high frying temperature may be the main contributors to darkening of the final products. Millard reaction depends on several factors, for

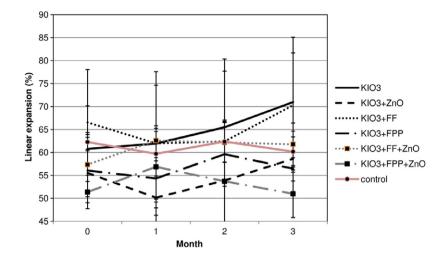


Figure 3 The linear expansion on frying of intermediate product to puffed final product. The fortifications are shown in the legend, while the timescale indicates storage time of the intermediate product before frying.

Table 3 The ΔE and L^* values of IP and FP fish cracker with various fortifications, at 0 and 3 months of storage

	ΔΕ				L*			
	IP		FP		IP		FP	
Premix	0 month	3 month ^{ns}	0 month ^{ns}	3 month ^{ns}	0 month	3 month ^{ns}	0 month	3 month
KIO ₃	$3.7~\pm~1.5^{ m b}$	4.0 ± 1.7	$\textbf{4.2}\pm\textbf{0.8}$	3.3 ± 1.4	37.4 ± 1.1^{b}	37.9 ± 1.5	60.4 ± 0.8^{ab}	$61.2 \pm 1.4^{\mathrm{ab}}$
KIO ₃ + ZnO	$\textbf{2.9}\pm\textbf{0.4}^{ab}$	2.3 ± 1.1	4.5 ± 0.1	4.3 ± 0.8	$\textbf{36.8} \pm \textbf{0.3}^{b}$	$\textbf{36.3} \pm \textbf{1.0}$	60.5 ± 0.3^{ab}	$61.6\pm0.3^{ m ab}$
$KIO_3 + FF$	1.7 ± 0.5^{ab}	$\textbf{2.3} \pm \textbf{0.6}$	4.8 ± 1.9	4.5 ± 0.4	$\textbf{35.7}\pm\textbf{0.5}^{ab}$	$\textbf{35.3} \pm \textbf{1.7}$	60.2 ± 2.2^{ab}	60.2 ± 0.4^{a}
KIO ₃ + FPP	0.8 ± 0.03^{a}	$1.7~\pm~0.1$	5.0 ± 0.7	5.5 ± 0.7	$\textbf{34.1} \pm \textbf{0.2}^{a}$	$\textbf{35.0} \pm \textbf{0.8}$	$58.4 \pm \mathbf{2.0^a}$	59.8 ± 0.3^{a}
$KIO_3 + ZnO + FF$	0.8 ± 0.1^{a}	$\textbf{3.5} \pm \textbf{0.3}$	4.3 ± 1.1	4.6 ± 1.9	$\textbf{34.4}\pm\textbf{0.4}^{a}$	$\textbf{37.6} \pm \textbf{0.3}$	$60.6\pm1.4^{\rm ab}$	60.0 ± 2.0^{a}
$KIO_3 + ZnO + FPP$	1.7 ± 0.9^{ab}	3.2 ± 1.3	$4.4~\pm~1.7$	5.4 ± 0.9	$\textbf{35.7}\pm\textbf{0.9}^{\text{ab}}$	$\textbf{37.3} \pm \textbf{1.3}$	$\textbf{62.1} \pm \textbf{2.4}^{\text{ab}}$	$61.0~\pm~1.3^{ m ab}$
Control	0 ± 0	0 ± 0	0 ± 0	0 ± 0	$\textbf{34.2} \pm \textbf{1.3}^{a}$	35.1 ± 1.5	$64.5\pm1.8^{\rm b}$	$64.9\pm1.6^{\rm b}$

IP, intermediate product; FP, finished product; ns, not significantly difference.

In a single column, different superscripts indicate significant differences (P < 0.05).

example chemical composition of food, Aw, pH and reaction temperature (Nurul *et al.*, 2009).

Preliminary consumer study

The sensory scores of FPs are listed in Table 4. The mean scores ranged from 5 (neither like nor dislike) to

7 (like moderately) in all six evaluated attributes, and the control samples scored in the range 6.2–6.7 for any of these attributes. The samples triple fortified with $KIO_3 + ZnO + FPP$ received significantly higher scores in all characteristics than samples with $KIO_3 + ZnO + FF$, especially in overall liking. This may have been an effect of the FPP on colour, as the

Table 4 Sensory evaluation scores of differently fortified fish cracker. The cracker was stored for 3 months in the form of an intermediate product, under ambient tropical conditions

Premix	Colour	Porosity	Fishy smell	Crispiness	Taste	Overall liking
Control	$6.3\pm2.0^{\rm bc}$	$6.4\pm1.6^{\rm bc}$	$6.3\pm1.9^{ m ab}$	$\textbf{6.2}\pm\textbf{2.2}^{a}$	6.6 ± 1.9^{bc}	$6.7\pm1.8^{ m bc}$
KIO ₃	6.9 ± 1.3^{c}	$6.4\pm1.4^{ ext{bc}}$	$7.0\pm1.3^{ m b}$	$6.4\pm1.8^{ m ab}$	6.2 ± 1.7^{abc}	$6.8\pm1.3^{ m bc}$
$KIO_3 + ZnO$	$5.4\pm1.7^{ m ab}$	5.1 ± 1.6^{a}	6.2 ± 1.5^{ab}	5.7 ± 2.0^{a}	5.9 ± 1.5^{ab}	$5.9\pm1.5^{\rm ab}$
$KIO_3 + FF$	$6.9\pm1.3^{\circ}$	$6.8\pm1.3^{\circ}$	$6.5\pm1.5^{ m b}$	$6.8\pm1.7^{ m ab}$	$6.4\pm1.7^{ m abc}$	$6.8\pm1.4^{ ext{bc}}$
$KIO_3 + FPP$	$6.8\pm1.3^{\circ}$	$6.9\pm1.2^{\circ}$	$6.8\pm1.5^{ m b}$	$7.6\pm1.3^{ m b}$	7.2 ± 1.3^{c}	7.4 ± 1.2^{c}
$KIO_3 + ZnO + FF$	$5.0\pm1.8^{\rm a}$	5.3 ± 2.0^{ab}	5.1 ± 2.2^{a}	$6.6\pm1.5^{ m ab}$	5.3 ± 2.0^{a}	$5.4\pm1.9^{ m a}$
$KIO_3 + ZnO + FPP$	7.4 ± 1.3^{c}	$7.1 \pm 1.3^{\circ}$	$6.6\pm1.7^{\rm b}$	7.5 ± 1.3^{b}	$\rm 6.9\pm1.5^{bc}$	$7.1\pm1.3^{\circ}$

In a single column, different superscripts indicate significant differences (P < 0.05).

latter samples had a beige tint, while the former were brownish. Consumers tend to prefer a brownish colour that could come from high fish content. In general, consumers also like fish smell in the cracker, perhaps for the same reason. Although the percentage of fish was fixed in our study, the triple-fortified cracker with FPP scored higher in fish smell than the triple-fortified case with FF. However, the crispiness scores were not aligned with the measured LE, while such positive correlation is commonly observed (Yu, 1991; Nurul et al., 2009; Neiva et al., 2011). Human sensory testing thus remains a necessity, and while a correlation may be widely accepted within the scope of baseline products. the effects of additives or fortificants may be unpredictable. However, only preliminary consumer evaluation could be conducted in the present study; a larger consumer test will be done in the near future.

Conclusions

Fish cracker is a popular snack in SEA and was considered as a potential food vehicle to improve iodine, Fe and Zn statuses especially for children. Triple-fortified fish cracker with $KIO_3 + ZnO + FPP$ retained iodine well during processing (92.7% of addition to dough) and further over 4 months of storage (72.7% remaining), and it received relatively high sensory scores. The retention percentages are essential to estimating appropriate fortification levels in the dough. We found that during processing to an IP, the loss of iodine was significantly accelerated by ZnO and ZnO + FF. The iodine loss during storage was not significantly affected by choice of fortification. Iron fortification significantly increased the TBARS value of the IP or the final product (FP), but effects on flavour remained undetected by human tasters. The ΔE values of FP did not depend on the type of fortification. The LE on frying/puffing to FP was unaffected by treatment. Sensory evaluation ranked triple fortification with FPP (not FF) the highest across multiple attributes.

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