

Original article

The stability of vitamin A in fortified palm olein during extended storage and thermal treatment

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Summary The stability of vitamin A in Refined Bleached Deodorised Palm Olein (RBDPOL) was studied for 24 months. Vitamin A decreased with time, temperature and thermal treatment (frying/cooking). RBDPOL fortification was observed over several temperature ranges, using PET, nylon and HDPE commercial packaging materials. After 24 months, the following vitamin A contents of 39–43 IU g⁻¹ (39–45%) at 16–20 °C; 35–40 IU g⁻¹ (43–49%) at 24–29 °C; and 28–39 IU g⁻¹ (45–73%) at 24–45 °C were detected at the respective temperature ranges. Results showed stability of vitamin A fortified RBDPOL vegetable oil was not stable over typical shelf life (12 months). Depletion of vitamin A accelerated when the RBDPOL vegetable oil was subjected to high temperature thermal treatment.

Keywords Fortification, oxidation, retinyl palmitate, stability, storage, temperature, vitamin A.

Background

Vitamin A (retinol) is considered essential for normal growth (FAO/WHO 2004). Since 2012, several nations have adopted fortification strategies, including vegetable oil as a fortified delivery mechanism; one example is Indonesia (Riskesdas (Riset kesehatan dasar), 2010). Vegetable oil is widely used as a food staple preparation in the world (Dary & Mora, 2002). It has been suggested that Indonesia is the leading global supplier of palm-based vegetable oil (Soekirman *et al.*, 2012). Studies have shown the fortification of vegetable oil is a suitable food staple to deliver fat-soluble vitamins such as vitamin A (Simonne & Eitenmiller, 1998). Recent studies have confirmed this for the Indonesia diet (Laillou *et al.*, 2013; Sandjaja *et al.*, 2015). Additionally, fortification of palm olein vegetable oil with vitamin A (retinyl palmitate), has been found to satisfy 94% of required dietary fortification for Indonesian families; where women receive 54% and children 51% to 57% of their estimated average requirement (Soekirman *et al.*, 2012; Laillou *et al.*, 2013). Currently, vitamin A deficiency remains a public health concern within Indonesia, especially in women and children. Periodic vitamin A supplements and a diet-based approach, has not resolved this

problem (Global alliance for Improved Nutrition (GAIN), 2010). A five-year study, has shown fortification of vegetable oil with vitamin A, improved intake amongst vulnerable populations without any increase in vegetable oil consumption (Global Alliance for Improved Nutrition (GAIN), 2010). Fortification of vegetable oil is considered to be cost effective and simple to implement (West & Darnton-Hill, 2008).

It is known that fortification of the vegetable oil with vitamin A, is largely determined by the initial oil quality (West & Darnton-Hill, 2008; Laillou *et al.*, 2012) The peroxide value (PV) is an important indicator of oil quality (FAO/WHO, 2011; Andarwulan *et al.*, 2014). PV has been shown to have a significant effect on the stability of vitamin A, regardless of storage conditions. A PV level of <2 meq kg⁻¹ has been proposed as a key parameter at the time of production (Laillou *et al.*, 2012). Concomitantly, the quality of vegetable oil (assessed by PV) prior to fortification has been shown to have a dramatic effect on the stability of retinyl palmitate (Andarwulan *et al.*, 2014); the same authors have shown that where a vegetable oil is oxidised through elevated levels of peroxides, it will significantly impact the loss of vitamin A (Viana *et al.*, 2007; Pignitter *et al.*, 2014a). Hydroperoxides are very unstable and decompose into a series of aldehydes, ketones, hydrocarbons, alcohols and many more reaction products as the oil oxidation process continues. In

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reality, these reactions can continue during storage of the packaged product, as the oil in the product continues to break down via autoxidation and develops oxidised or rancid flavour in the product (Gupta, 2005). Additionally, there is known to be a negative effect on vitamin A, when exposed to oxygen and ultraviolet light (Hariyadi, 2009; Andarwulan *et al.*, 2014; Hemery *et al.*, 2015). Other factors affecting stability of vitamin A during storage are acidity, temperature, moisture and impurities (Atwood *et al.*, 1995; Hariyadi, 2009; Pignitter *et al.*, 2014a,b).

Although the effect of storage on the stability of vitamin A in edible oil has previously been studied (Hemery *et al.*, 2015), there is insufficient information on the behaviour of vitamin A, when stored for extended duration (Soekirman *et al.*, 2012; Andarwulan *et al.*, 2014). These factors have implication for typical commercial food grade edible oil, fortified with vitamin A. Specifically, this information is not known to be available from a commercial context and subsequently, it requires investigation.

The first aim of this study was to assess the long-term (24 months) stability of vitamin A added to RBDPOL cooking oil in several commercial grade packaging materials. One main use of vegetable oil is for deep and shallow frying (stir-frying), where food materials, such as vegetables, meat, seafood etc., are brought into direct contact with the vegetable oil (Orthofer & List, 2007; Farmer, 2014). The thermal temperature and contact time vary considerably depending on the specific food product to be cooked (Choe & Min, 2007). Thermal temperatures are usually within the range of 175–195 °C (Erickson, 2006; Banks, 2007). The thermal treatment of the food material is often accompanied with the presence of hydrolysing conditions namely water and steam, causing hydrolysis and triacylglycerol breakdown (free fatty acids) (Gupta, 2005). Hence, a second aim of this study, was to observe the effect of thermal treatment by simulation of shallow frying (sautéing) and deep frying fortified RBDPOL vegetable oil, then noting its impact to vitamin A (retinyl) retention. A third aim was the consideration of packaging material for food distribution, because it needs to be versatile enough to withstand, physical and chemical integrity (Galotto *et al.*, 2008).

Materials and method

Material

RBDPOL, was supplied from PT. SMART, Tbk, Indonesia. Table 1, describes the specification of the RBDPOL for testing an initial first sample series containing 70 IU g⁻¹ vitamin A. This concentration was selected based on recommendations from previous studies, to add Vitamin A (retinyl palmitate) from 60

to 75 IU g⁻¹ (Bagriansky & Ranum, 1998) and was sourced and supplied from DSM Nutritional Products Ltd., Singapore and BASF SE, Germany, with the parameters as described and shown in Table 2.

Iodine value (IV), Peroxide value (PV), Free fatty acid (FFA), Moisture & Impurities (M&I), colour measurement determination

The iodine value was determined by iodometric titration (AOCS Ca 1b-87). Peroxide value was determined by AOCS Cd 8-53, 2009. Free fatty acids were determined by AOCS Ca 5a-40. Moisture was determined by AOCS Official Method Ca 2b-38. Oil colour was determined by Lovibond Tintometer colour scale (AOCS Cc13e-92).

Storage and sampling of fortified oil

At the first series, oil samples were prepared in normal laboratory conditions and stored at the following three temperature ranges: 16–20 °C (cool storage), 24–29 °C (ambient nontemperature control storage) and 24–45 °C to represent typical seasonal outdoor, day and night equatorial temperatures. These temperatures were chosen from the most extreme meteorological temperatures (Data not shown). Samples were continuously stored within dark enclosed conditions for 24 months. Preparations of RBDPOL and vitamin A 1.0 mio IU were prepared at room temperature (approx. 25–28 °C).

Table 1 Refined Bleached Deoderised (RBD) POL specification

Analysis parameter	Method	Result	
		1st study	2nd study
Iodine value	AOCS Cd 1b-87	59.8 ± 3.0	61.5 ± 1.0
Peroxide Value	AOCS Cd 8-53	0.37 ± 0.2	0.45 ± 0.1
Free Fatty Acid	AOCS Ca 5a-40	0.038 ± 0.02	0.026 ± 0.02
Moisture & Impurities	AOCS Ca 2b-38	0.02 ± 0.01	0.01 ± 0.01
Colour 5.25" (R)	AOCS Cc 13e-92	1.8 ± 0.01	1.8 ± 0.01

Table 2 Description of essential parameters of vitamin A 1.0 mio IU specification

Parameter	BASF	DSM
Peroxide Value, meq kg ⁻¹	Max. 10.0	Max. 10.0
Acid value, mg KOH g ⁻¹	Max. 2.0	Max. 2.0
Assay	1.0 mio IU g ⁻¹ (vitamin A = 300 000 RE) per g	1.0–1.1 mio IU g ⁻¹

Vitamin A 1.0 mio IU was supplied from DSM Nutritional Products Ltd., Singapore, using a concentration of 70 IU g⁻¹ for RBDPOL within the following packaging types:

- 1 PET (Polyethylene terephthalate) Bottle (transparent) 250 mL, supplied from Indopet, Indonesia (100% PET).
- 2 Nylon LLDPE (linear low density polyethylene) Pouch 225 mL (transparent), supplied from Panverta, Indonesia, with the following specification nylon 15 µ/adhesive/LLDPE 120 µ.
- 3 HDPE (High density polyethylene) jerrycan (JC) 2 L, natural white, supplied from CV. Asia, Indonesia (100% HDPE).

Vitamin A (retinyl palmitate concentration) was quantified at intervals of one time cycle, every third month and analysed by BBIA (Center for Agro-Based Industry) Bogor, Indonesia using UPLC (Ultra performance liquid chromatography) system (supplied from Waters Acquity, Singapore) using AOAC official method 2001.13.

A second sample series was prepared using vitamin A 1.0 mio IU, supplied from BASF SE, Germany. Samples were prepared at concentrations of 45 IU g⁻¹ (to comply with SNI 7709/2012 regulation, requiring 45 IU g⁻¹ concentration at point of manufacture) and 180 IU g⁻¹ (to comply with Permenperind No. 87/2013 regulation, requiring minimum content of 40 IU g⁻¹ at point of procurement). Since preparing this work, a new regulation was adopted and updated to Permenperind No.35/M-IND/PER/3/2015, requiring a minimum content of 20 IU g⁻¹ at point of procurement. Only two packaging types: transparent PET (as in the first series) and opaque nylon LLDPE were used.

This second series was maintained at two conditions; firstly, storage to mimic typical ambient conditions and maintained between temperatures of 24–29 °C. In parallel, samples were maintained externally and treated to natural light, between temperatures of 24 and 45 °C for a duration of 24 months. Samples were analysed for vitamin A (retinyl palmitate) concentration at one measurement interval per month. As before, analysis of vitamin A was conducted by BBIA.

Head space measurement

Nominal capacity was determined by filling the empty packing with water, at 20 °C to the net volume. The brim capacity was determined by filling the empty pack to the maximum capacity of the pack filled to the brim. Headspace volume (mL) is the difference between full capacity brim volume and volume at nominal net volume.

Quantification of vitamin by high-performance liquid chromatography (HPLC)

The vitamin A was determined by liquid chromatography method (Association of Analytical Chemists, 2011). Standardised vitamin A (Retinyl palmitate) was supplied from Supelco, Sigma-Aldrich (purity 93.8%).

HPLC determination was started with baseline check for a minimum of 30 min with mobile phase: methanol:ultrapure water (95:5) at flow rate 0.5 mL min⁻¹; instrument: Shimadzu Prominence-i LC-2030 C; column: Shim-Pack GIST C18 4.6 × 150 mm. mobile phase: methanol:ultrapure water (95:5); flow rate: 0.5 mL min⁻¹; vol. Inject: 10 µL; detector: UV; wavelength: 328 nm; calculation: determination of vitamin A in sample using calibration curve.

$$y = bx + a; x = \frac{(y-a)}{b} \text{ Weight of sample (g)}$$

where, y = peak area; b = slope; a = intercept; x = Vitamin A IU g⁻¹ in sample.

The calculation of retained vitamin A in RBDPOL in n month was determined by the following:

$$\frac{C_n}{C_0} \times 100\%$$

C_0 = vitamin A content: 0 month (IU g⁻¹); C_n = vitamin A content: n month (IU g⁻¹).

The stability of vitamin A content in RBDPOL by heat stability test

The stability of vitamin A content in RBDPOL vegetable oil was determined using a heat stability test. After addition of vitamin A 1.0 mio IU, the vegetable oil was heated to a temperature of 180 °C, and sustained for 5 min using a standard laboratory hot plate stirrer (Thermo scientific: type SP131630 – 33Q). The vitamin A content, PV and FFA were analysed before and after the heating process. Samples were prepared in the second sample series as follows:

- 1 Sample A, RBDPOL vegetable oil with 45 ppm minimum vitamin A 1.0 mio IU.
- 2 Sample B, RBDPOL vegetable oil with 225 ppm (five times concentration of sample A) of vitamin A 1.0 mio IU.

Vitamin A, PV and FFA levels were analysed in duplicate (AOAC 2001.13 method for vitamin A (retinol) in foods) Analysis was conducted using ISO 17025 accredited laboratory (PT. SMART, Indonesia).

Statistical analysis

Statistical analysis was first examined using a combination of Student t -test to determine uncertainty

(Birch, 2003). Final analysis was carried out using analysis of variance ($\alpha = 0.05$) with MiniTab v.16.1.1 software.

Results

Calibration of vitamin A

A calibration of vitamin A level concentration (IU g^{-1}) is shown in Fig. 1. Using the same regression we calculated the vitamin A peak area for 20, 45, 70, 180 and 225 IU g^{-1} . Calculation of vitamin A based on fortification 20, 45, 70, 180 and 225 IU g^{-1} is shown below here:

$$Y = bx + A$$

where, x = found concentration of vitamin A; Y = peak area; b = slope; A = intercept.

Calculations show correlation between peak area obtained from analysis using HPLC and regression of linearity. Table 3, shows the initial PV and vitamin A analysis results, after prolonged temperature exposure and differing vitamin A concentrations at the end of storage time, for a duration between 16 and 24 months and in all packaging materials. Table 3 seems to indicate that higher temperature exposure, and longer storage leads to a decrease of vitamin A.

Result of first series

Figure 2, shows the effect of packaging type, time and temperature, with initial vitamin A concentration of 70 IU g^{-1} . Results are vivid. Figure 2a, shows the concentration of vitamin A retained in the RBDPOL vegetable oil using PET bottles at each temperature condition to 24 months. At 12 months these are approx.

61% ($42.7 \text{ IU g}^{-1} \pm 0.1$) at 16–20 °C; 57% ($39.5 \text{ IU g}^{-1} \pm 1.2$) at 24–29 °C; 55% ($38.5 \text{ IU g}^{-1} \pm 1.6$) at 24–45 °C. After 18 months approx. 51% ($39.5 \text{ IU g}^{-1} \pm 3.2$) at 16–20 °C; 39% ($27.4 \text{ IU g}^{-1} \pm 0.3$) at 24–29 °C; 33% ($23.2 \text{ IU g}^{-1} \pm 2.5$) at 24–45 °C; and after 24 months approx. 49% ($43.5 \text{ IU g}^{-1} \pm 0.0$) at 16–20 °C and 0% ($0.0 \text{ IU g}^{-1} \pm 0.0$) at 24–29 °C and 24–45 °C.

Retention of vitamin A content in RBDPOL vegetable oil using the nylon LLDPE (linear low density polyethylene) pouch at temperatures 16–20 °C, 24–29 °C and 24–45 °C after 12 months was found to be approx. 59% ($41.4 \text{ IU g}^{-1} \pm 2.3$) at 16–20 °C; 51.6% ($36.1 \text{ IU g}^{-1} \pm 0.8$) at 24–29 °C; 27% ($19.2 \text{ IU g}^{-1} \pm 5.3$) at 24–45 °C. After 18 months approx. 44% ($30.8 \text{ IU g}^{-1} \pm 2.7$) at 16–20 °C; 33.4% ($23.4 \text{ IU g}^{-1} \pm 0.5$) at 24–29 °C; 28.5% ($1.9 \text{ IU g}^{-1} \pm 0.0$) at 24–45 °C; and after 24 months approx. 32% ($22.8 \text{ IU g}^{-1} \pm 0.0$) at 16–20 °C; 33% ($23.1 \text{ IU g}^{-1} \pm 0.0$) at 24–29 °C; ($0.0 \text{ IU g}^{-1} \pm 0.0$) at 24–45 °C, respectively. After a 24 month period, vitamin A retained in nylon LLDPE is shown in Fig. 2b.

The effect of vitamin A content in HDPE jerrycan packing is shown in Fig. 2c. Retention of vitamin A content in RBDPOL vegetable oil using the HDPE Jerrycan at temperatures 16–20 °C, 24–29 °C and 24–45 °C, and after 12 months was found to be approx. 55.7% ($39.0 \text{ IU g}^{-1} \pm 0.4$) at 16–20 °C; 51.2% ($35.8 \text{ IU g}^{-1} \pm 0.2$) at 24–29 °C; 40.3% ($28.2 \text{ IU g}^{-1} \pm 6.9$) at 24–45 °C. After 18 months approx. 26.1% ($18.3 \text{ IU g}^{-1} \pm 1.6$) at 16–20 °C; 36.7% ($25.7 \text{ IU g}^{-1} \pm 1.4$) at 24–29 °C; 22.5% ($15.8 \text{ IU g}^{-1} \pm 0.9$) at 24–45 °C; and after 24 months, 100% ($0.0 \text{ IU g}^{-1} \pm 0.0$) loss for all conditions.

Figure 2a, b and c show the vitamin A content declines rapidly within the first 3 months of storage. Reduction of vitamin A in all packaging materials (PET, nylon LLDPE, HDPE JC) at 12, 18 and 24 months and all temperature ranges, was significant

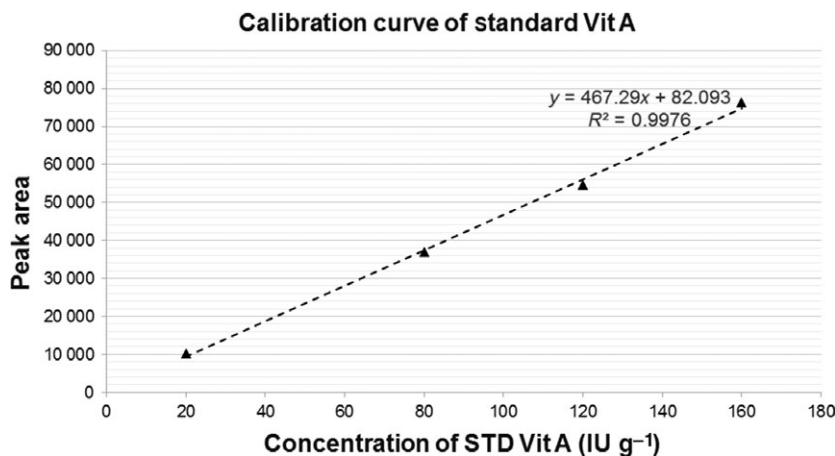


Figure 1 Standard curve of vitamin A.

Table 3 The effect of vitamin A retention in RBDPOL vegetable oil as a result of temperature, initial vitamin A (IU g⁻¹) and packaging material after 16 and 24 months

Pack Type	Temp. Condition	2nd series (end of 16 months)					
		1st series (end of 24 months)		(Vitamin A 45 IU g ⁻¹ - When packed)		(Vitamin A 180 IU g ⁻¹ - When packed)	
		Parameter	Analysis result	Parameter	Analysis result	Parameter	Analysis result
PET Bottle	16-20	PV (meq O ₂ kg ⁻¹)	5.2	PV (meq O ₂ kg ⁻¹)	Not measured	PV (meq O ₂ kg ⁻¹)	Not measured
		Vit A (IU g ⁻¹)	34.5	Vit A (IU g ⁻¹)	Not measured	Vit A (IU g ⁻¹)	Not measured
	24-29	PV (meq O ₂ kg ⁻¹)	6.8	PV (meq O ₂ kg ⁻¹)	10.2	PV (meq O ₂ kg ⁻¹)	11.1
		Vit A (IU g ⁻¹)	0	Vit A (IU g ⁻¹)	32.5	Vit A (IU g ⁻¹)	148.2
	24-45	PV (meq O ₂ kg ⁻¹)	8.6	PV (meq O ₂ kg ⁻¹)	9.7	PV (meq O ₂ kg ⁻¹)	12.7
		Vit A (IU g ⁻¹)	0	Vit A (IU g ⁻¹)	0	Vit A (IU g ⁻¹)	0
Nylon LLDPE Pouch	16-20	PV (meq O ₂ kg ⁻¹)	7.4	PV(meq O ₂ kg ⁻¹)	Not measured	PV (meq O ₂ kg ⁻¹)	Not measured
		Vit A (IU g ⁻¹)	22.8	Vit A (IU g ⁻¹)	Not measured	Vit A (IU g ⁻¹)	Not measured
	24-29	PV (meq O ₂ kg ⁻¹)	25.8	PV (meq O ₂ kg ⁻¹)	44.7	PV (meq O ₂ kg ⁻¹)	46.4
		Vit A(IU g ⁻¹)	23.1	Vit A (IU g ⁻¹)	34.4	Vit A (IU g ⁻¹)	126.8
	24-45	PV (meq O ₂ kg ⁻¹)	35.4	PV(meq O ₂ kg ⁻¹)	89.8	PV (meq O ₂ kg ⁻¹)	89.8
		Vit A(IU g ⁻¹)	0	Vit A (IU g ⁻¹)	0	Vit A(IU g ⁻¹)	60.3
HDPE JC	16-20	PV (meq O ₂ kg ⁻¹)	11.4	PV (meq O ₂ kg ⁻¹)	Not measured	PV (meq O ₂ kg ⁻¹)	Not measured
		Vit A (IU g ⁻¹)	0	Vit A (IU g ⁻¹)	Not measured	Vit A (IU g ⁻¹)	Not measured
	24-29	PV (meq O ₂ kg ⁻¹)	20.1	PV (meq O ₂ kg ⁻¹)	20.1	PV (meq O ₂ kg ⁻¹)	126.8
		Vit A (IU g ⁻¹)	0	Vit A (IU g ⁻¹)	0	Vit A (IU g ⁻¹)	89.8
	24-45	PV (meq O ₂ kg ⁻¹)	26	PV (meq O ₂ kg ⁻¹)	26	PV (meq O ₂ kg ⁻¹)	89.8
		Vit A (IU g ⁻¹)	0	Vit A (IU g ⁻¹)	0	Vit A (IU g ⁻¹)	60.3

Cooking Oil standard refer to SNI 7709 - 2012. FFA = max. 0.3% and PV = max. 10.

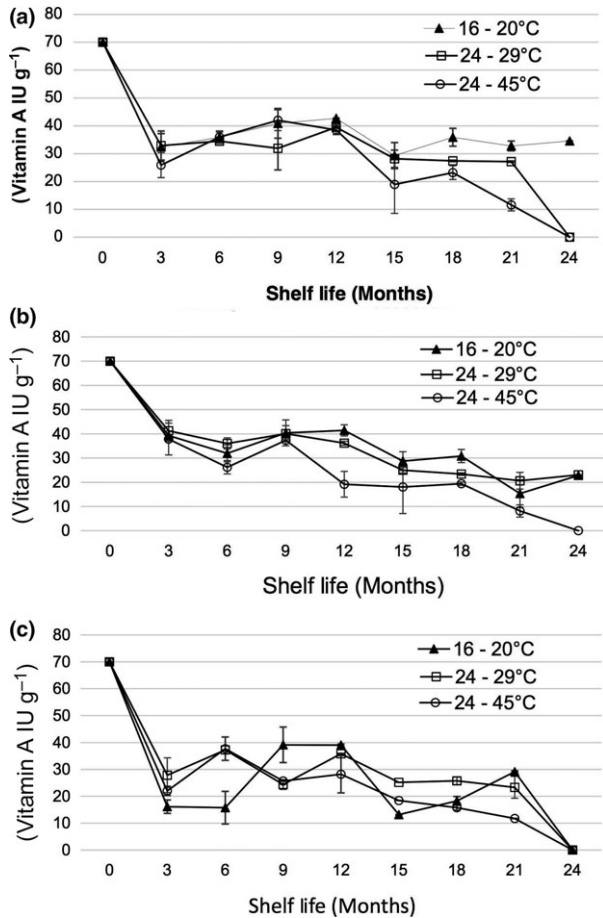


Figure 2 Effect of time and temperature on vitamin A (70 IU g^{-1}) retention for 24 month duration in (a) transparent PET packaging; (b) transparent nylon LLDPE packaging and (c) HDPE packaging.

($P < 0.01$). Overall, based on the differentiation of the three packaging materials, it is found that vitamin A loss in RBDPOL vegetable oil using HDPE jerrycan tended to be higher. The most extreme temperature condition ranged from 24 to 45 °C. Although these sample did not have direct exposure to UV or sunlight, the reduction of vitamin A during storage is likely influenced by effect of oxidation process caused through OTR (Oxygen transmission rate) which is possibly higher in HDPE than PET bottle or nylon LLDPE; the dynamic head space for the HDPE jerrycan is higher. Respective head space volumes (two determinations) were PET 250 mL bottle, 32.5 ± 5 mL; nylon LLDPE 225 mL pouch, 7.5 ± 2.5 mL; HDPE 2.0 L JC, 156.11 ± 50 mL.

Although not the main focus of our results, the PV concentration cannot be ignored and could have accelerated the decrease of vitamin A concentration (Andarwulan *et al.*, 2014). Results in Table 3, show high PV levels as result of oxidation, at 16 and

24 months. We believe the presence of these high peroxide compounds are a concomitant factor, initially onset through extended exposure to high environmental temperature for extended periods. These also may have contributed and influenced the decomposition of vitamin A (Laillou *et al.*, 2012; Pignitter *et al.*, 2014a, b). In the event of fulfilling vitamin A fortification requirements for intervention programmes, the PV level may need to be monitored if exceeding beyond acceptable level of 10 meq kg^{-1} (Pignitter *et al.*, 2016).

Result of second series

The second series (vitamin A concentrations 45 and 180 IU g^{-1}) of results reveal interesting differences. Starting with the PET packaging, Fig. 3a shows that with a concentration of 45 IU g^{-1} vitamin A, the results show a similar trend compared to 180 IU g^{-1} at 24–45 °C (Fig. 4a). However, at 45 IU g^{-1} (24–45 °C), the decline is rapid to 100% loss ($0.0 \text{ IU g}^{-1} \pm 0.0$) during only one month duration. Whereas with 180 IU g^{-1} , the vitamin A content is not detectable after 3 months, at 24–45 °C. At the slightly cooler temperature spectrum, Fig. 3a shows retention of initial 45 IU g^{-1} vitamin A, in PET bottle at 24–29 °C was $32.5 \text{ IU g}^{-1} \pm 0.2$ after 16 months. When compared with increased dosage over the same time period, Fig. 4a shows the retention of initial 180 IU g^{-1} of vitamin A in PET to be approx. 82.3% ($148.2 \text{ IU g}^{-1} \pm 0.0$) at 24–29 °C.

Figures 3b and 4b, shows the effect of opaque nylon packaging, time and temperature on reduced vitamin A (45 and 180 IU g^{-1}) retention for 16 month duration. The vitamin A content in the opaque nylon pouch at 24–29 °C after 16 months was approx. 70% ($126.8 \text{ IU g}^{-1} \pm 0.2$) to 77% ($34.4 \text{ IU g}^{-1} \pm 0.2$), while at 24–45 °C, retention was approx. 33% ($60.3 \text{ IU g}^{-1} \pm 0.1$) to 35% ($15.5 \text{ IU g}^{-1} \pm 0.0$).

The reduction of vitamin A in RBDPOL vegetable oil using PET bottle and opaque nylon LLDPE during the shelf life is significant ($P < 0.05$). The nylon LLDPE with opaque translucence may have protected the vegetable oil from exposure to UV light, whereas the transparent PET bottle, could potentially have helped to initiate and accelerate oxidation of vitamin A in the vegetable oil. Irrespective of these factors, all samples were stored within dark, enclosed environment. It would seem that after 16 months, the effect of opaque nylon may have contributed to greater vitamin A retention, than PET at 24–45 °C. However, at the same temperature, with 180 IU g^{-1} vitamin A concentration, the opposite effect was observed.

Result of heat stability test

Results from examination of thermal treatment of RBDPOL fortified with vitamin A, are shown in

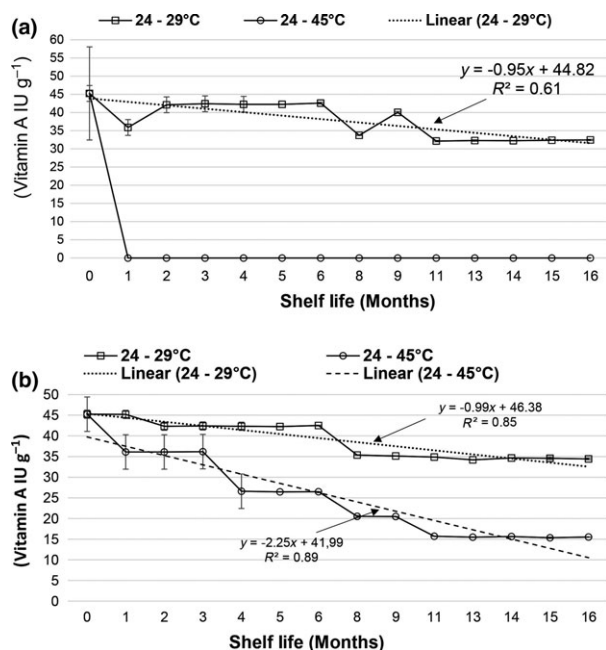


Figure 3 Effect of time and temperature on vitamin A concentration (45 IU g^{-1}) and retention for 16 months duration in (a) transparent PET packaging and (b) opaque nylon packaging.

Table 4, where applied heat treatment to the RBDPOL was designed to simulate a simple domestic thermal treatment procedure (Warner, 1999). Table 4 shows the PV, FFA and vitamin A content in the RBDPOL samples before and after single heat treatment (180°C for 5 min). The data show the vitamin A reduced from between 33% and 49% after sustained thermal treatment. Twin analysis and double determinations, consistently show an increase in the PV, ranging from 7.61 to $9.35 \text{ meq O}_2 \text{ kg}^{-1}$. This suggests that additional thermal treatment ($\sim 180^\circ\text{C}$) associated within a typical domestic application could further accelerate the onset of peroxides, evident from monitoring the PV value and the degradation of vitamin A in RBDPOL vegetable oil. Pignitter *et al.* (2014a,b) have previously shown a tendency for a compound decrease of retinol when compared to a mildly oxidised oil; this and the aforementioned factors may explain another mechanism by which means the vitamin A in RBDPOL vegetable oil will be rapidly depleted during any subsequent domestic heating application.

Conclusion & recommendation

Overall, the average vitamin A content in RBDPOL vegetable oil reduced more than 30% during 12 month storage. The implication of this is better understood in the context of one recently proposed fortification programme. While fortification at 70 IU g^{-1} of vitamin A

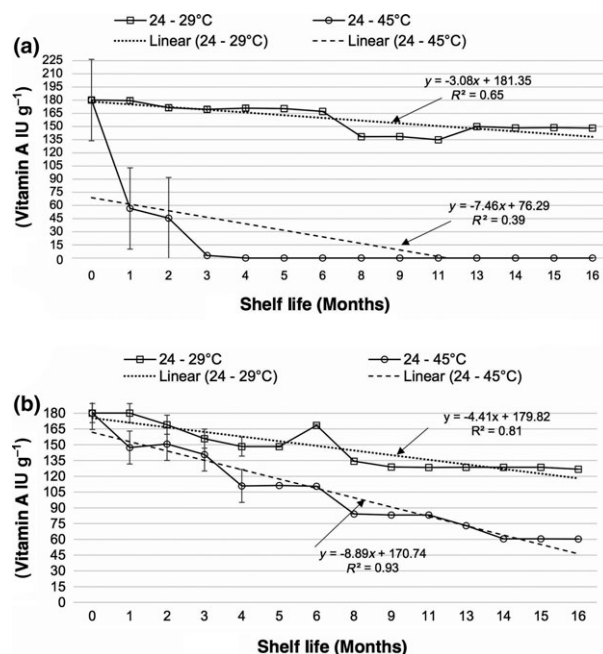


Figure 4 Effect of time and temperature on vitamin A concentration (180 IU g^{-1}) and retention for 16 months duration in (a) transparent PET packaging and (b) opaque nylon packaging.

Table 4 The PV, FFA and vitamin A concentration data before and after thermal treatment at 180°C for 5 min

No	Sample	Vit. A content (ppm)	PV ($\text{meq O}_2 \text{ kg}^{-1}$)	FFA (%-as palmitic)
1	P01	51.8 ± 0.40	0.74 ± 0.01	0.041 ± 0.00
2	P01.1	33.7 ± 0.14	9.07 ± 0.04	0.046 ± 0.00
3	P01.2	34.3 ± 0.13	9.35 ± 0.00	0.047 ± 0.00
4	PO2	228.4 ± 1.73	0.75 ± 0.00	0.041 ± 0.00
5	PO2.1	120.0 ± 0.11	7.61 ± 0.02	0.050 ± 0.00
6	PO2.2	116.7 ± 0.78	8.13 ± 0.02	0.049 ± 0.00

Means \pm standard deviation of two determination.

P01 = RBDPOL + 45 ppm vit. A $1.0 \text{ mio IU g}^{-1}$ (before heating).

P01.1; 1.2 = RBDPOL + 45 ppm vit. A $1.0 \text{ mio IU g}^{-1}$ (after 5 min at 180°C).

PO2 = RBDPOL + 225 ppm vit. A $1.0 \text{ mio IU g}^{-1}$ (before heating).

PO2.1; 2.2 = RBDPOL + 225 ppm vit. A $1.0 \text{ mio IU g}^{-1}$ (after 5 min at 180°C).

is sufficient to meet Indonesian national fortification standard, (SNI 7709:2012), which requires 45 IU g^{-1} at period of manufacture; it is not sufficient to satisfy Indonesian Industrial ministry regulation Permenperind No.35/M-IND/PER/3/2015, requiring a minimum content of 20 IU g^{-1} at time of procurement. Consequently, compliance with mandatory regulatory requirements for vitamin A content of 45 IU g^{-1} in RBDPOL vegetable oil for the duration of typical shelf life conditions, is questionable.

Overall in the first series we found PET to be superior, when compared to transparent nylon. Several factors could be gas transmission rates and water vapour permeability of crystalline polymer are affected and in fact reduced the deterioration process in storage time. Mangaraj & Goswami (2009) have shown the O₂ permeability values of four plastic film types in cm³.µm.m⁻².h⁻¹.atm⁻¹ which are 105.83 for Polyamide (nylon 6); 50–100 for Polyethylene Terephthalate (PET); 2916.66–8333.34 for Linear Low Density Polyethylene; and 1640.41–3280.83 for High Density Polyethylene. Possibly, this is one explanation why the first study showed PET, was the best material, minimising vitamin A deterioration, because of lower OTR (Siracusa, 2012). However, in the second series, the opaque nylon enabled better vitamin A retention, compared to the transparent PET, possibly because vitamin A is known to be more sensitive to UV light than oxygen contact (Andarwulan *et al.*, 2016).

The results showed vitamin A content in the RBDPOL vegetable oil is reduced when subjected to thermal treatment and is further related to typical conditions associated with domestic cooking processes (Andarwulan *et al.*, 2014).

Although some losses occur during processing, distribution and storage, special attention to handling, such as addition of nitrogen before sealing, low light storage or low UV light adsorbent packaging can minimise excessive losses of vitamin A during distribution and storage (Andarwulan *et al.*, 2014). The technique of handling and storing fortified food before consumption can positively or negatively influence the content of vitamin A (Dwyer *et al.*, 2015). Another factor to consider is the choice and availability of the particular vegetable oil for fortification. Although a more unsaturated vegetable oil is more susceptible to oxidation, a low level oxidised oil may be more acceptable for vitamin A fortification programmes (Pignitter *et al.*, 2016). When comparing other vegetable oils, for example soybean oil to palm oil, it has been suggested that palm oil is a superior vehicle for successful delivery of vitamin A fortification programmes due to its good oxidative stability (Pignitter *et al.*, 2016). Additionally, it should also be recognised that effective fortification practices should be coupled with clear and measurable consumer communication best practices. The benefits of fortification to the public should be communicated effectively through consumer education programmes providing simple information about domestic storage, handling and consumption (Dwyer *et al.*, 2015; Nair *et al.*, 2015).

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Author contributions

Dewi K. N. Silalahi, Bayu Segara and Isti Christianti conceived and designed experiments; Dewi K. N. Silalahi, Bayu Segara and Isti Christianti performed experiments; Dewi Yulianti, Latifun Jayanti, Monica da Silva and Karyanto Mulyono analysed the data; Dewi K. N. Silalahi and Paul Wassell participated in transcription; All authors reviewed and endorsed the manuscript.

Conflict of interests

The authors declare no conflicts of interest.

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