Kasetsart J. (Nat. Sci.) 45 : 510 - 520 (2011)

Production and Nutrition of Khi Lek *(Siamese cassia)* Curry from Central Thailand

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ABSTRACT

Khi Lek (locally called *Siamese cassia*) curry is an ancient food throughout Thailand that uses the tips, young leaves and young flowers of the plant. However, as they have a bitter taste, the Thai people must boil them in water 2–3 times before use. Boiling may impact on the characteristics of Khi Lek young leaves (KL). Thus, this project studied the effect of sodium chloride and eggplant, the optimum time (minutes) and number of times boiled of KL for curry making and its nutrition. The results showed that the anhydrobarakol content in KL boiled in water alone, in KL boiled in water containing sodium chloride and in KL boiled in water containing eggplant were not significantly different ($P > 0.05$). The anhydrobarakol content in boiled KL decreased with increasing time of boiling and/or the number times boiled. KL for curry should be boiled twice in a volume of water six times the KL weight, for 3 min/time. KL curry had a very low anhydrobarakol content (0.17 mg by fresh weight) and a low tendency to cause liver toxicity.

Keywords: Khi Lek young leaves, *Siamese cassia*, anhydrobarakol

INTRODUCTION

Senna siamea is a plant widely grown in Southeast Asia, including Thailand, where it is called 'Khi Lek'. Many parts of it have been used for a long time as medicine: the roots as an antipyretic and the leaves for treatment of constipation, hypertension, insomnia and asthma (Ahn *et al*., 1978), the stem bark for constipation, malaria and associated diseases such as fevers and jaundice (Ahn *et al*., 1978; Nsonde-Ntandou *et al*., 2005; Kaur *et al*., 2006). The main active constituent (barakol) in the flowers and young leaves of Khi Lek was first extracted in 1969 (Hassanali-Walji *et al.,* 1969) and identified as 3,4-dihydro-3,8-dihydroxy-2,5-dimethyl-1,4-dioxaphenalene, also known as 2,5-dimethyl-3 H-pyrano-[2,3,4-de]-1-benzopyran-3a,8-diol and was produced using a synthetic procedure by Bycroft *et al*. (1970). Barakol contains a tricyclic ring structure that is unstable and is converted from barakol ($C_{13}H_{12}O_4$) to anhydrobarakol ($C_{13}H_{10}O_3$) through the loss of a molecule of water. Anhydrobarakol has a melting point of 163 °C and is dark green in color (Thongsaard *et al*., 2001). Anhydrobarakol hydrochloride ($C_{13}H_{12}ClO_3$), a relatively stable salt of barakol, can be prepared as shown in Figure 1 by the addition of

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Received date : 15/06/10 Accepted date : 21/12/10

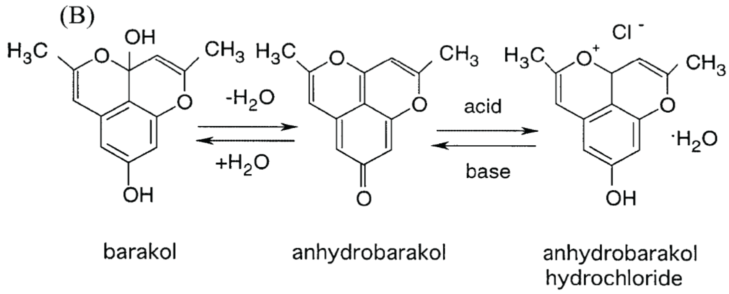

Figure 1 Structure of barakol, anhydrobarakol and anhydrobarakol hydrochloride (Thongsaard, 1998).

concentrated hydrochloric acid to a solution of barakol (Thongsaard, 1998). The physical characteristics of barakol are characterized by pale yellow needle crystals and it has melting point of 166–170 °C (Hassanali-Walji *et al.*, 1969; Thongsaard *et al.*, 2001). Barakol is soluble in methly alcohol and ethyl alcohol, and is moderately soluble in water (Chaichantipyuth, 1979). Barakol has anxiolytic properties similar to diazepam (Thongsaard *et al.*, 1996) but in 2009, Deachapunya and Thongsaard (2009) reported that acute and chronic oral administration in rats had no anxiolytic and locomotive effects and it exerted a sedative effect by a reduction in the directed exploratory behaviors.

Khi Lek herbal drug capsules have been produced in Thailand to reduce sleeplessness, with 400 mg of leaf powder per capsule (10 mg of anhydrobarakol, a product of barakol). The dosage of this herbal drug was 2–4 capsules before bed time but it was withdrawn from the market in 2003 due to hepatotoxicity (Hongsirinirachorn *et al.*, 2003). The hepatotoxicity of Khi Lek was explained by Chivapat *et al.* (2001) who found that Khi Lek capsules caused degeneration and necrosis of hepatocytes in Wistar rats and the severity of the lesion was dose dependent. However, curry food from Khi Lek leaves remains popular without any reports of hepatotoxicity after hundreds of years.

Because of the toxicity reports, it is still necessary to study the barakol content in Khi Lek curry compared with processed Khi Lek capsules. There were some reports on the barakol content, as Padumanonda *et al.* (2007) found that the barakol content in young leaves, mature leaves and young flowers was 1.67, 0.78 and 1.43% dry weight, respectively. Padumanonda and Gritsanapan (2006) found that fresh young leaves contained 0.40% w/w and the first and second boiled leaves that were boiled 1 h/time had 0.14 and 0.04% fresh weight, respectively. Boiling reduced the barakol content in boiled Khi Lek young leaves (KL). However, boiling may also impact the physical, chemical, sensory and nutrient characteristics of KL curry that have not been reported. Thus, this project studied the effect of boiling on KL and the optimum boiling process for KL curry and its nutrients, especially the anhydrobarakol content.

MATERIALS AND METHODS

Materials

Fresh KL were collected from Samut Prakan *province* Thailand in 2008, packed in plastic bags and kept overnight night at 10 °C. Eggplant (*Solanum torvum*) and sodium chloride were purchased from a local market. The anhydrobarakol standard was obtained from the Government Pharmaceutical Organization, Thailand.

Methods

Effect of sodium chloride and eggplants on the KL boiling process

A completely randomized design experiment with two replications was conducted in three treatments of: 1) KL boiled in a volume of water six times the weight of the KL; 2) KL boiled in water containing sodium chloride, 4% of the KL weight; and 3) KL boiled in water containing eggplant, 20% of the KL weight. All treatments were boiled twice at 97 °C for 4 min. The sodium chloride and eggplant were added in the first boiling time of the second and third treatments, respectively. After boiling, the KL samples were blended and determined for color using a spectrophotometer (Spectraflash 600 plus, Data-color International, USA), and CIE color values were recorded as L* = lightness (0 = black, 100 = white), a* (-a* = greenness, +a* = redness), b* (b* = blueness, +b* = yellowness). In addition, the samples were determined for anhydrobarakol content, modified from Pumpaisalchai *et al.* (2005) and Padumanonda *et al.* (2004), using high performance liquid chromatography (HPLC). Sensory evaluation of KL curry using a central Thailand recipe was undertaken using 30 untrained panelists (eight males and 22 females, aged from 24 to 57 y) and a 7-point hedonic scale (1 = dislike very much to 7 = like very much) for color, aroma, flavor, texture and overall liking. The first and second boiled filtrates were determined for total soluble solids (TSS) using a hand refractometer and for pH using a pH meter (Orion, Model 410). The data were analyzed using analysis of variance and Duncan's new multiple range test for mean comparisons at the 0.05 significance level in the SPSS statistical software, version 12.

Anhydrobarakol determination

Anhydrobarakol extracted from samples with methanol was subjected (1mL) to HPLC using the following parameters: acetronitrile: MeOH (80:20) mobile phase, flow rate 1 mL/min, using a UV spectrophotometer detector at 241 and 366 nm and calculation compared with standard anhydrobarakol. Equation 1 was used to determine the anhydrobarakol content:

$$\text{Anhydrobarakol, mg/1000g} = C * V1*1000 / Wt * V2 \quad (1)$$

where: C = anhydrobarakol content (mg)
V1 = sample volume (mL)
Wt = sample weight (g)
V2 = injection volume (mL) to HPLC (mL)

The 3 × 2 factorial experiment, in a completely randomized design with two replications, was conducted as six treatment combinations (Table 1). The boiled KL was kept at -13 to 15 °C. After that, the boiled KL was thawed for sensory evaluation in a KL curry using a central Thailand recipe using 30 untrained panelists (eight males and 22 females, aged from 24 to 57 y) and a 7-point hedonic scale. Some parts were blended to determine the color and anhydrobarakol content. The second and third boiled KL filtrates were determined for color, TSS and pH. The data were analyzed using the techniques described above.

Table 1 Treatments to determine the effect of time (min) of KL boiling and number of times boiled.

Treatment (min)	Time	Number of times boiled	Treatment	Time (min)	Number of times boiled
1	3	2	4	5	3
2	3	3	5	7	2
3	5	2	6	7	3

Nutrition of KL curry

The KL curry was analyzed in triplicate for the content of moisture, fat, protein (N × 6.25), ash, crude fiber, carbohydrate, calcium, phosphorus and iron using an in-house method (AOAC, 2005 and 2008). The anhydrobarakol content of the curry was calculated from the boiled KL.

KL curry recipe from central Thailand

The curry was made using the following procedure:

a) Fresh KL (87 g) was boiled twice in water six times its weight for 3 min/time.

b) The ingredients of the curry (15 g dried red chili, 7 g lemon grass, 15 g shallot, 15 g garlic, 4 g galangal, 1.5 g skin of kaffir lime, 4 g sodium chloride, 60 g galingal and 8 g shrimp paste sauce) were blended and 15 g dried Spanish mackerel added and the ingredients mixed together.

c) Coconut milk (350 g) was added into the pot and heated until it boiled after which the ingredients from b) above were added and heated until boiling and then the boiled KL from a) above and 15 g roasted fish were added. The total ingredients were boiled for 10 min.

d) The curry was flavored with 35 g fish sauce and 35 g palm sugar and heated until boiling.

RESULTS AND DISCUSSION

To make KL curry, the Thais must boil the KL in water and they usually add sodium chloride or eggplant into the water to reduce the bitterness. Thus the study tried to determine the effect of sodium chloride and eggplant on the KL boiling process. With the first filtrate (Figure 2), it was found that only the TSS values were significantly different. The TSS of the first filtrate of KL boiled in water had the lowest value while the TSS of other treatments were higher and were not significantly different because the added sodium chloride and eggplant provided TSS also. The acidity of the KL (Figure 2) was shown by the low pH values of 3.61–3.69 which produced the sour taste. The color of the first filtrate in the three treatments was red-yellow with turbidity (Figure 2).

For the second filtrate (Figure 3), it found that only the TSS was significantly different, as was the case with the first filtrate. However, the TSS of all three treatments was lower than the TSS

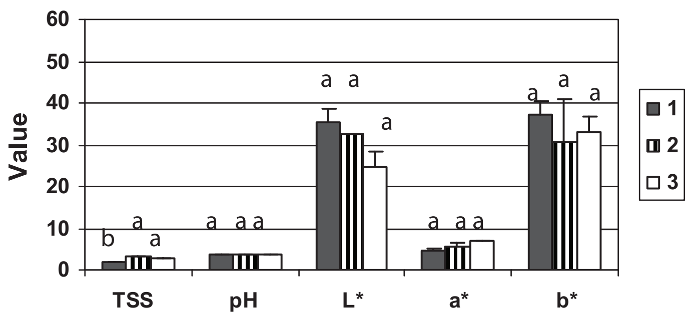

Figure 2 Effect of sodium chloride and eggplants on Khi Lek young leaves (KL) in the boiling process on TSS (°B), pH and color (L* a* b*) in the first filtrate.
1 = KL boiling in water; 2 = KL boiling in water containing sodium chloride;
3 = KL boiling in water containing eggplant.
Mean values of each characteristic with the same letter were not significantly different ($P > 0.05$).
(⊤ indicates the upper range of the mean plus the standard deviation.)

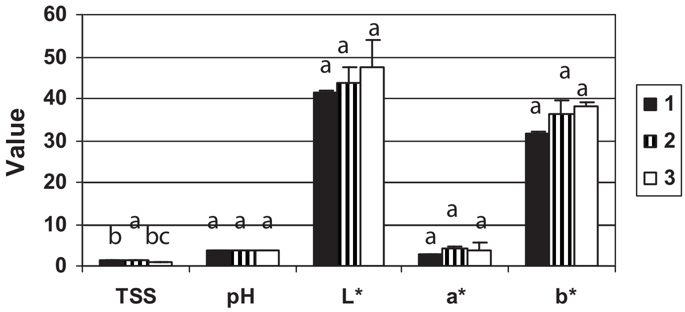

Figure 3 Effect of sodium chloride and eggplant on Khi Lek young leaves (KL) in the boiling process on TSS (°B), pH and color (L* a* b*) in the second filtrate.
1 = KL boiling in water; 2 = KL boiling in water containing sodium chloride;
3 = KL boiling in water containing eggplant.
Mean values of each characteristic with the same letter were not significantly different ($P > 0.05$).
(⊤ indicates the upper range of the mean plus the standard deviation.)

of all three treatments from the first filtrate by 40, 59.38 and 59.26% for the filtrate from boiling KL in water, the filtrate from boiling KL in water containing sodium chloride and the filtrate from boiling KL in water containing eggplant, respectively. The TSS in the filtrate from boiling KL in water containing sodium chloride was the highest because it had sodium chloride residue from the first boiling. The pH of the second filtrate was higher than the pH of the first filtrate by 4, 2.49 and 3.82% for the filtrate from boiling KL in water, the filtrate from boiling KL in water containing sodium chloride and the filtrate from boiling KL in water containing eggplant, respectively. However the second filtrate was still acidic with a pH of 3.7–3.86. The color of the second filtrate for all three treatments was yellow and was less turbid than the first filtrate, especially for the L* and b* values. The L* value of the second filtrate from boiling KL in water containing eggplant increased by 72.85%, while the filtrate from boiling KL in water containing sodium chloride and the filtrate from boiling KL in water increased slightly by 35 and 18.18%, respectively. The b* value increased slightly by 15.79, 19.13 and 14.60% for the filtrate from boiling KL in water, the filtrate from boiling KL in water containing sodium chloride and the filtrate from boiling KL in water containing eggplant, respectively. The a* value decreased slightly also by 2.62, 4.04 and 3.88% for the filtrate from boiling KL in water, the filtrate from boiling KL in water containing sodium chloride and the filtrate from boiling KL in water containing eggplant, respectively.

The color and anhydrobarakol content (Figure 4) of the three treatments were not significantly different. In general, the color of the three treatments was brown. The sensory evaluation of the KL curry (Figure 5) by the 30 panelists showed there was no significant difference between any of the attributes among treatments and their average scores were moderate because the curry had a high concentration of coconut milk and spices so that these ingredients may have concealed its flavor, especially the bitterness.

Effect of time (min) of boiling KL and the number of times boiled

From the above data, it was found that there was no significant difference in the quality

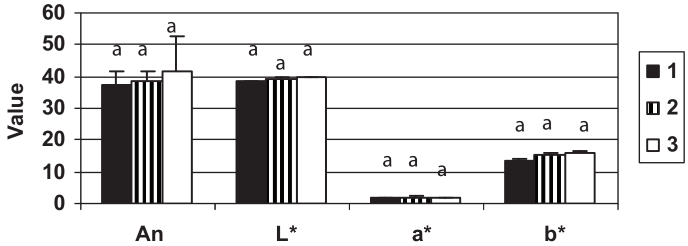

Figure 4 Anhydrobarakol content (An; mg/kg) and color (L* a* b*) of boiled KL.
1 = KL boiling in water; 2 = KL boiling in water containing sodium chloride;
3 = KL boiling in water containing eggplant.
Mean values of each characteristic with the same letter were not significantly different ($P > 0.05$).
(⊤ indicates the upper range of the mean plus the standard deviation.)

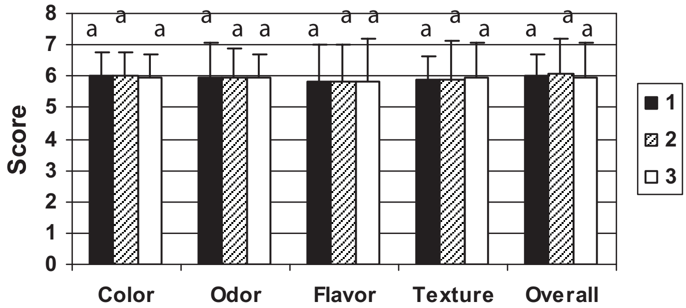

Figure 5 Sensory evaluation of the quality of the KL curry.
1 = KL boiling in water; 2 = KL boiling in water containing sodium chloride;
3 = KL boiling in water containing eggplant.
Mean values of each characteristic with the same letter were not significantly different ($P > 0.05$).
(⊤ indicates the upper range of the mean plus the standard deviation.)

of the boiled KL from boiling the KL in water, boiling the KL in water containing sodium chloride or boiling the KL in water containing eggplant, so to save cost the treatment of boiling KL in water was chosen to study the optimum boiling time (min) and the optimum number of times to boil (times). From Table 2, the boiled KL from each treatment had significantly different amounts of anhydrobarakol and there was an interaction also. The anhydrobarakol content from the treatment boiling KL twice for 3 min was the highest and it was not significantly different from boiling KL twice for 5 min. The anhydrobarakol content from the treatment boiling KL three times for 5 min was lowest and was not significantly different from boiling KL three times for 7 min. In general, the anhydrobarakol content in boiled KL decreased with an increase in the boiling time and the number of times boiled, which agreed with an earlier report indicating a decrease in the anhydrobarakol content of about 90% after boiling fresh young leaves twice and 71.42% from boiling fresh young leaves once (Padumanonda and Gritsanapan, 2006).

Table 2 Anhydrobarakol content of boiled KL from the interaction of the main effects of time boiled (min) and number of times boiled (times) (average ± standard deviation).

Time (min)	Number of boiling (times)	Anhydrobarakol content (mg/kg)
3	2	12.76±0.19 [a]
3	3	2.08±0.52 [c]
5	2	11.30±1.87 [a]
5	3	1.09±0.08 [cd]
7	2	7.58±1.05 [b]
7	3	1.97±0.16 [c]

Mean values in a column followed by the same superscript were not significantly different at $P > 0.05$

There was no interaction between TSS, pH and color so those results are shown for the two main factors of the treatments only. The first factor of time boiled (Figure 6) showed that different boiling times provided TSS (0.51–0.72), pH (3.87–4.0) and color values that were not significantly different as was the color of the boiled KL (Figure 7) but the anhydrobarakol content (Figure 7) was significantly different as 7 min of boiling had an anhydrobarakol content (4.78) that was lower than that from 3 min of boiling (7.42) by 35.58%.

The second factor of the number of times boiled (Figure 8) showed that the TSS and pH of the filtrate were significantly different. The TSS decreased by 62.07% when the number of times boiled increased but the pH increased slightly by 8.8%. The color of the filtrate was not significantly different but the L* value tended to increase with an increase in the number of times boiled. The a* value of the color of boiled KL (Figure 9) only decreased significantly (10.79%) when the number of times boiled increased but the anhydrobarakol content of boiled KL decreased substantially by 84.63% when the number of times boiled increased.

The sensory evaluation of KL curry by the 30 panelists (Figure 10) found that there was no significant different between attributes among treatments and the average scores of all attributes were moderate so to save cost and maintain a high anhydrobarakol content in the curry, the treatment

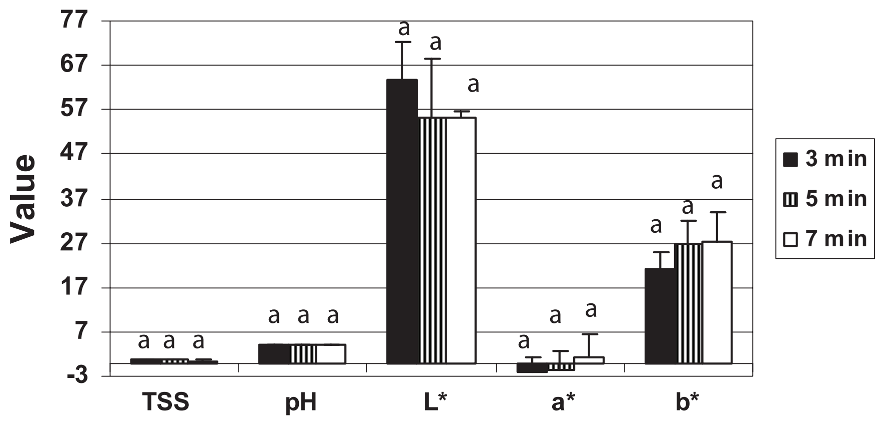

Figure 6 TSS (°B), pH and color (L* a* b*) of KL filtrate based on time boiled (min). Mean values with the same letters were not significantly different ($P > 0.05$). (⊤ indicates the upper range of the mean plus the standard deviation.)

of boiling twice for 3 min was selected. This boiling process differed from Padumanonda and Gritsanapan (2006) where the KL was boiled twice in three times the volume of water (w/w) but with 1 hour between successive boiling events. In Thailand, KL curry is made using many recipes in different families or in different parts of the kingdom, so the KL boiling process may differ in time, quantity of added water, number of times boiled and also the quality of selected young leaves.

The nutrition of KL curry

On average, 100 g fresh weight of KL curry had moisture, fat, protein, ash, crude fiber, carbohydrate (100 - moisture, fat, protein, ash, crude fiber), calcium, phosphorus and iron contents of 84.21 g, 6.27 g, 3.40 g, 2.12 g, 2.51 g, 1.49 g, 29.50 mg, 57.30 mg and 0.37 mg, respectively, and had 0.17 mg of anhydrobarakol by calculation because the KL curry had a high level of coconut milk and spices that made it difficult to filtrate and analyze. The calculation of

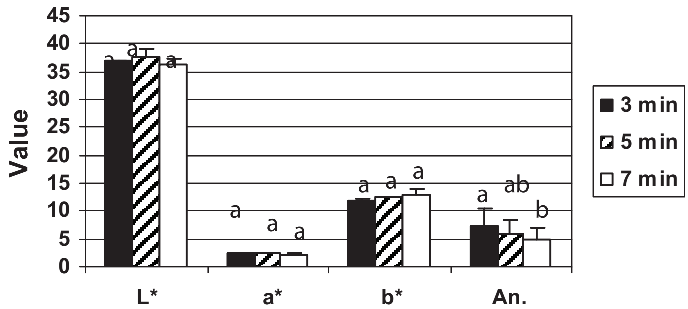

Figure 7 Anhydrobarakol (An; mg/kg) and color (L* a* b*) of boiled KL based on time boiled (min). Mean values with the same letters were not significantly different ($P > 0.05$). (⊤ indicates the upper range of the mean plus the standard deviation.)

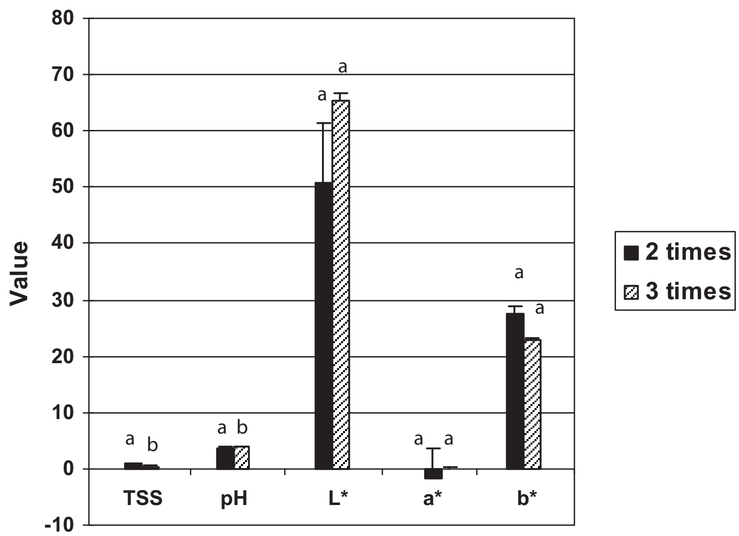

Figure 8 TSS (°B), pH and color (L* a* b*) of the KL filtrate based on the number of times boiled. Mean values with the same letters were not significantly different ($P > 0.05$). (⊤ indicates the upper range of the mean plus the standard deviation.)

the anhydrobarakol content involved starting from 100 g KL curry that used 8.7 g fresh KL which produced 13.40 g boiled KL that in turn contained 1.28 mg/100 g anhydrobarakol. The anhydrobarakol content of this boiled KL contained about 0.00128% (w/w) with less than 0.04% lost in the second boiled leaves samples from the research of Padumanonda and Gritsanapan (2006) because there may have been variation in the sourcing of the KL, material preparation and also in the method used for analysis. Padumanonda and Gritsanapan (2006) used TLC-densitometry instrumentation but the present research used HPLC.

The anhydrobarakol content in the KL curry in the present study was very low and Thai people do not consume KL curry every day. Compared to the recommended dose of 2–4 Khi Lek capsules containing 10 mg or more of anhydrobarakol (product of barakol) per capsule

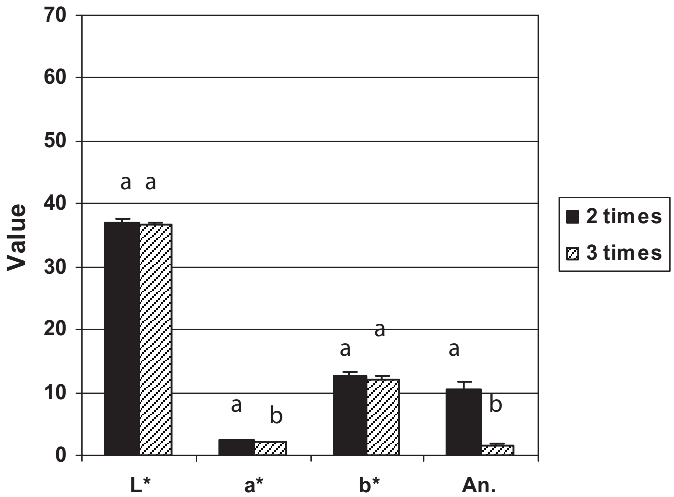

Figure 9 Anhydrobarakol (An; mg/kg) and color (L*a*b*) of boiled KL based on the number of times boiled.
Mean values with the same letters were not significantly different ($P > 0.05$).
(⊤ indicates the upper range of the mean plus the standard deviation.)

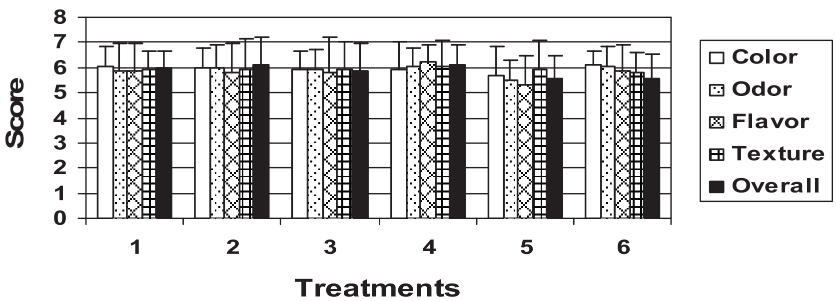

Figure 10 Sensory evaluation quality of KL curry based on the effect of time boiled and number of times boiled.
(⊤ indicates the upper range of the mean plus the standard deviation.)
Each attribute of mean between treatments was not significantly different at $P > 0.05$.

(Hongsirinirachorn, *et al*. 2003), this would require the consumption of 1.56–3.12 kg Khi Lek curry/day which would be impossible under normal conditions. Thus, the anhydrobarakol consumed from KL curry was unlikely to cause hepatotoxicity. The consumption of unboiled Khi Lek leaves gives a much higher anhydrobarakol content. The toxicity data of barakol in mice (intraperitoneal injection), showed that a lethal dose of barakol was high (LD50 = 324.09 mg/kg; Jantarayota, 1987). Barakol in doses of 60–240 mg/kg also decreased triglycerides and the effect persisted for at least two weeks in the recovery group. Barakol may disrupt liver functions, especially lipid metabolism and bilirubin, in a dose-dependent manner. These effects were reversible. The literature indicated that barakol may be clinically used in short-term treatments (Pumpaisalchai *et al*., 2005). Despite the toxicity reports for Khi Lek capsules, KL curry is still a popular food in Thailand. Other ingredients in KL curry may have an influence on the low level of toxicity.

CONCLUSION

The anhydrobarakol content in KL boiled in water, in KL boiled in water containing sodium chloride and in KL boiled in water containing eggplant was not significantly different ($P > 0.05$). The anhydrobarakol content, TSS and acidity in boiled KL decreased with an increase in the boiling time (min) and the number of times boiled. KL used to make curry should be boiled twice in a water volume only six times the weight of the KL for 3 min/time. KL curry had a very low anhydrobarakol content (0.17 mg by fresh weight) and so had a low tendency to cause liver toxicity.

ACKNOWLEDGEMENTS

The researchers wish to thank the National Research Council of Thailand for supporting the funding of this project and Food Research and Product Development, Kasetsart University for providing research laboratory facilities.

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