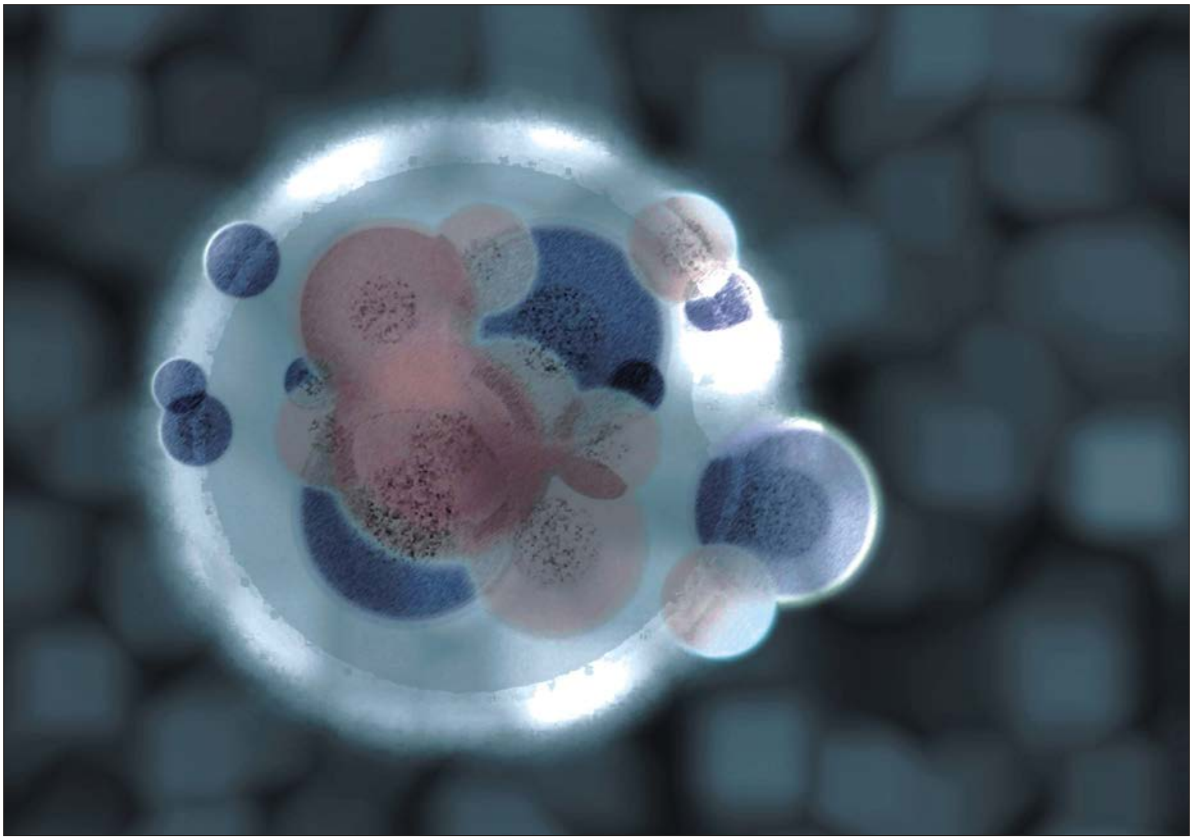

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Screening of Antioxidant Activity of Some *Samia ricini* (Eri) Silks: Comparison with *Bombyx mori*

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Abstract: Antioxidant is an interesting molecule which has been studied world wide for human health. The aims of this study are to screen the Total Phenolic Content (TPC) and antioxidant activity of sericin extracts from 5 varieties of Eri silk. In addition, domesticated Thai silk; Nanglai and Dokbua were also investigated in comparison. The sericin was firstly extracted by warm distilled water. The TPC of each silk variety was determined using Folin-ciocalteu assay. The results indicated that domesticated silk have higher TPC than all of Eri. DPPH radical scavenging and Ferric Reducing/Antioxidant Power (FRAP) were used for measurement the antioxidant activity of the sericin extracts. The results found that domesticated Thail silk had higher antioxidant capacity than that of Eri but with different values. The results suggested that TPC and antioxidant of the sericin extracts exhibited good correlation. This means the TPC is main factor for antioxidant capacity. However, the variation of both TPC and antioxidant of sericin extracts were influenced by silk varieties. Moreover, domesticated Thai silk composed of some pigments which are antioxidant compounds. However, Eri silk should be considered as alternative sources for antioxidant.

Key words: Antioxidant, Eri, radical scavenging, silk, sericin, total phenolic content

INTRODUCTION

Antioxidant is known as beneficial health substances and has recently been increased interest in exploration world wide (Uddin *et al*., 2008; Yang and Gadi, 2008). Most of natural antioxidants are phenolic compounds which universally distributed in the plant kingdom as secondary metabolic products (Tian *et al*., 2004; Vicente *et al*., 2011). Fruits and vegetables, especially red, blue and black colors, are known to be major dietary sources of the phenolic compounds (Heim *et al*., 2002; Atak *et al*., 2011). Wildly studied about antioxidants on free radical protection and effect to human health were reported by Seal (2011). Another one is synthetic antioxidants, among them Butylated Hydroxyanisole (BHA) and Butylated Hydroxytoluene (BHT) is the most commonly used (Iqbal *et al*., 2005). However, it has also been reported about adverse effect of synthetic antioxidants. Therefore, increasing in identifying naturally occurring antioxidants from some biomaterials, by-products and residual sources was gradually focused (Seal, 2011). Silk, a natural protein source produced generally by mulberry (domesticated) and non-mulberry (wild) silks (Altman *et al*., 2003; Acharya *et al*., 2009). In each silk fiber has fibroin and sericin protein which are the main components (Kim *et al*., 2005; Takasu *et al*., 2010). Silk has been used in textile for long history and recently applied for various applications (Meinel *et al*., 2005; Fang *et al*., 2009; Schneider *et al*., 2009; Nogueira *et al*., 2010). Moreover, silk has been used in various forms depending on their application (Srihanam, 2011; Baimark *et al*., 2009; Baimark and Srihanam, 2009; Simchua *et al*., 2011). Sericin contained of bulky polar amino acids and proposed to have potential activity on human health. Currently, sericin protein is used for various applications including cosmetics additive, functional biomaterials and pharmacological and medical uses (Prommuak *et al*., 2008).

In Thailand, a yellow color of domesticated Thai silk has been studied and found to have antioxidant activity (Prommuak *et al*., 2008). However, information about antioxidant activity of wild silk was rarely published. *Samia ricini* (Eri) silk is a kind of wild silk that has been cultured in Thailand. Among their properties, antioxidant activity was little information, especially the effect of different variety. Depending on locally name, Eri silk in Thailand can be classified into at least 5 varieties; China, Chiang Mai, Lamphun, Thai and SIC. Therefore, this study was aimed to measure the antioxidant activity of different kinds of Eri silk varieties. The results were then compared to those of domesticated Thai silk; Dokbua and Nanglai varieties.

MATERIALS AND METHODS

This study was performed for 3 months from March 10, 2011 to June 10, 2011. Most of the experiment was performed at Department of Chemistry, Faculty of Science, Mahasarakham University, Thailand.

Materials

Eri cocoons: The cocoons of 5 Eri silk varieties called China, Chiangmai, Lamphun, Thai and SIC and domesticated Thai silk names Nanglai and Dokbua were kindly supplied by Silk Innovation Center (SIC), Mahasarakham University, Maha Sarakham, Thailand. All chemical reagent of analytical grade were used.

Methods

Extraction of sericin solution: The cocoons of all silk varieties were boiled in distilled water with magnetic stirred at 70-75°C for 60 min to extract sericin. The weight ratio of 1 g silk per 10 mL of distilled water was used in this experiment. The sericin solution was then filtrated and collected at 4°C until investigation.

Determination of antioxidant activity and total phenolic content: This investigation of radical-scavenging activity was applied from Chew *et al.* (2008) by using DPPH (2,2'-diphenyl-1-picrylhydrazyl) method. Ferric Reducing/Antioxidant Power (FRAP) assay was also performed according to previously modified method of Kubola and Siriamornpun (2011) to confirm the activity. In addition, Total Phenolic Content (TPC) of the sericin extract was also investigated which applied from Bonoli *et al.* (2004).

Statistical analysis: All the results are shown as Mean±SD of triplicate measurement.

RESULTS

Antioxidant activity of the sericin extracts: In present study, the antioxidant activities of sericin extracts was determined by DPPH and FRAP assays as shown in Table 1 and 2, respectively.

DPPH radical scavenging activity: The amount of DPPH radical scavenging of sericin extracts was expressed by % inhibition (Table 1). From results, significant differences in scavenging activity were observed. The highest scavenging activity at concentration of 100 (%v/v) was found for Nanglai, then Dokbua which were 90.4866 and 83.7879% inhibition, respectively. The scavenging of Eri silk was lower than domesticated silk. When Eri silks are compared, SIC (62.6882%) and Chiangmai (62.1863%) demonstrated higher radical scavenging activities, followed by Lamphun (58.3242%), China (35.0862%) and Thai (34.4534%), respectively.

Ferric Reducing/Antioxidant Power (FRAP): The FRAP values of the sericin extracts are shown in Table 2. The results indicated that Dokbua had the greatest reducing power, followed by Nanglai silk. Eri silk had also lower reducing power than that of domesticated silk. Chiangmai, Lamphun and SIC varieties showed similar FRAP values followed by China and Thai varieties.

Total Phenolic Content (TPC): The Total Phenolic Content (TPC) of all silk extracts was investigated using a modified Folin-Ciocalteu method and results were expressed in terms of mg GAE/g as shown in Table 2. TPC of the extracts differed among the silk varieties and was in the range of about 0.2-1.2 mg GAE/g of sericin extracts. Both domesticated Thai silks; Dokbua and Nanglai had nearly equal with about 3 times of TPC higher than Eri silk. Comparison between Eri, the highest of TPC values was obtained from Lamphun, followed by SIC, Chiangmai, Thai while China had the lowest TPC.

DISCUSSION

Sericin, a glue-like protein, is a part of silk fiber that acts as coating substances of fibroin core proteins together. The sericin was excluded and became by-product in textile production process. Recently, it has been known for its potential activity such as antibacterial and antioxidant activities (Sarovart *et al.*, 2003; Wu *et al.*, 2007). Previously, almost antioxidant substances were phenolic compounds. They

Table 1: DPPH scavenging activities of different sericin extracts

Concentration (% v/v)	% inhibition ($mg\ mL^{-1}$) Nanglai	Dokbua	China	Chiang-mai	Lam-phun	SIC	Thai
1.5625	24.1420±0.0062	22.2800±0.0242	16.3403±0.0524	22.9377±0.0004	22.7576±0.0010	20.7915±0.0044	15.9003±0.1004
3.1250	28.4713±0.0149	25.0742±0.0153	18.4713±0.0044	23.4897±0.0026	23.7374±0.0064	22.6226±0.0062	20.0562±0.0172
6.250	33.1593±0.0045	32.0093±0.0802	23.7993±0.0122	24.3790±0.0005	25.2984±0.0115	23.0951±0.0114	20.4921±0.0226
12.50	44.1700±0.0137	44.5200±0.0089	24.4700±0.0037	25.5750±0.0125	27.8696±0.0009	24.1583±0.0025	22.0492±0.1087
25.00	55.1005±0.0066	51.3630±0.0106	31.2235±0.0096	28.6109±0.0045	34.2516±0.0062	26.9344±0.0145	25.1635±0.0126
50.00	62.1000±0.0108	60.0008±0.0040	35.2007±0.0140	36.0626±0.0154	36.5473±0.0231	27.1412±0.0020	29.2267±0.0152
100.00	90.4866±0.0033	83.7879±0.0047	35.0862±0.0012	62.1863±0.0249	58.3242±0.0116	62.6882±0.0033	34.4534±0.0046

Table 2: Comparison of Total Phenolic Content (TPC) and Ferric Reducing/Antioxidant Power (FRAP) of different sericin extracts

Silk varieties	TPC (mgGAE/g)	FRAP (mM Trolox/g)
Nanglai	1.2145±0.0012	5.3209±0.0034
Dokbua	1.2022±0.0005	5.5137±0.0122
China	0.2059±0.0005	1.2957±0.0038
Chiangmai	0.3870±0.0005	2.9266±0.0026
Lamphun	0.5928±0.0016	2.9400±0.0051
SIC	0.4117±0.0009	2.9057±0.0029
Thai	0.3335±0.0022	0.9708±0.0065

are widely distributed in plant and have been reported of their potential benefit for human health (Govindarajan *et al*., 2007). Total Phenolic Content (TPC) of the sericin extracts was investigated using the Folin-Ciocalteu assay. This method was suggested as a fast and reliability to quantify phenolic content (Kubola and Siriamornpun, 2011). The results found that significant differences were observed for TPC among the silk varieties. The TPC and antioxidant activity exhibited good correlation (Butsat *et al*., 2009). The DPPH radicals are widely used to investigate the radical scavenging activity of antioxidant compounds. Generally, antioxidant molecules acted as H-atom donor to stabilize the DPPH radicals. Scavenging activity of the sericin extract is indicated by % inhibition. The results found that % inhibition values varied from 34.45-90.50%. The highest of % inhibition was found by Nanglai. It may be suggested that Nanglai had higher sericin ratio per silk fiber than Dokbua and all of Eri. The varied radical scavenging activity of the sericin extracts depend on the TPC values in each silk varieties (Butsat and Siriamornpun, 2010). This result suggested that different types of silk composed of different antioxidant molecules and capacity. In addition, FRAP assay was used for investigation the ability of antioxidant to reduce Fe^{3+}-Fe^{2+} (Benzie and Strain, 1996). The FRAP values of the sericin extracts indicated that domesticated Thai silk. Dokbua had the highest reducing power equally to Nanglai. This may be due to their higher TPC. The obtained result was positively associated with other reports (Butsat and Siriamornpun, 2010; Govindarajan *et al*., 2007). The finding results clearly evidenced that domesticated Thai yellow silk (Dokbua and Nanglai) had higher both TPC and antioxidant capacity than that of Eri silk. This may be due to domesticated silk composed of some pigments in their fibers. The main components of silk pigment are carotenoids and flavonoids which corresponding for antioxidant activity (Prommuak *et al*., 2008). Differences of TPC and antioxidant activity in same types may depend on many factors such as host plant, environmental weather and habitat.

CONCLUSION

The different Total Phenolic Content (TPC) of the sericin extracts was found to be important factors on antioxidant activity. The different values of both TPC and antioxidant activity depend on silk varieties. However, the TPC and antioxidant activity of the sericin extracts showed good correlation. When types of silk were compared, domesticated Thai silk have higher TPC and antioxidant capacity than those of Eri. This may be due to the sericin content of domesticated silk was higher than Eri as well as the pigments composition on the silk fibers.

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