

Antioxidants from a Chinese medicinal herb – *Psoralea corylifolia* L.

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Abstract

The powder and extracts of *Psoralea corylifolia* L. were tested in lard at 100 °C by using the oxidative stability instrument (OSI) and were found to have strong antioxidant effects. Six compounds, bakuchiol, psoralen, isopsoralen, corylifolin, corylin and psoralidin were isolated from the herb and identified by UV, IR, Mass, ¹H and ¹³C NMR spectra and melting point. Their antioxidant activities were investigated individually and compared with butylated hydroxytoluene (BHT) and α -tocopherol by the OSI at 100 °C. The results showed that bakuchiol, corylifolin, corylin and psoralidin had strong antioxidant activities, and especially psoralidin (stronger antioxidant property than BHT), but psoralen and isopsoralen had no antioxidant activities at 0.02% and 0.04% levels. The antioxidant activities of the compounds decrease in the following order: Psoralidin > BHT > α -tocopherol > bakuchiol > corylifolin > corylin > isopsoralen ~ psoralen.

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1. Introduction

Lipid peroxidation not only produces rancid odours and flavours, but also decreases safety and nutritional quality by destruction of essential fatty acids and vitamins in foods during cooking, processing and storing. Lipid peroxidation causes aging, heart disease and carcinogenesis (Edwin, 1996). Oxidation of foods can be retarded in several ways, such as conditions of vacuum, or air replaced by nitrogen or low temperature. In industrial processing, addition of highly effective antioxidants has become a popular and highly effective means to lengthen the shelf life of foods and to reduce nutritional losses and harmful substances formed (Kanner, Harel, & Jeffe, 1991; Tsuda, Ohshima, Kawakishi, & Osawa, 1994).

Synthetic antioxidants, such as butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT) and *tert*-butylhydroquinone (TBHQ), are widely used in the food industry. However, animal test have demonstrated that BHA and BHT accumulate in the body and result in liver damage and carcinogenesis (Ames, 1983; Baardseth, 1989; Grice, 1986; Ito, Fukushima, Hasegawa, Shibata, & Ogiso, 1983; Ito et al., 1986; Wichi, 1988). Therefore, development and utilization of more effective and non-toxic antioxidants of natural origin are desired (Namiki, 1990).

Psoralea corylifolia L. has been used traditionally as medicine in China and recommended for the treatment of stomachic, deobstruent, anthelmintic, diuretic, vitiligo and also certain skin diseases, e.g., leucoderma, psoriasis and leprosy (Kotiyal & Sharma, 1992; Zhu, 1998). Few reports, however, have addressed the antioxidant activity of *P. corylifolia* L.

In this paper, the antioxidant activity effects of the components isolated from this herb were investigated and their antioxidant effects were compared with those of the most commonly used antioxidants, BHT and α -tocopherol, by OSI, in lard at 100 °C.

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2. Materials and methods

2.1. Materials

P. corylifolia L. was purchased from Shanghai Drug Company, PR China, dried with ventilation at ambient temperature, and stored at 4 °C until use.

2.2. Chemicals

BHT and α -tocopherol were purchased from the Chemical Company, Shanghai, China. Silica gel was bought from Qingdao Ocean Chemical factory, PR China. Lard was rendered in the laboratory from fresh pig fat tissue purchased from Shanghai Slaughter House, PR China. Other chemicals used in this experiment were all AR grade and from Shanghai Chemical Reagent Co.

2.3. Extraction

One kilogramme of air-dried and pulverized *P. corylifolia* L. was exhaustively extracted with petroleum ether and chloroform at room temperature, successively.

2.4. Chromatographing on silica gel column

After removal of the solvents in vacuum, 110 g petroleum ether extract and 65 g chloroform extract, respectively, were obtained. Both of the extracts were tested for their antioxidant activities. Twenty-four grammes of petroleum ether extract were chromatographed on a silica gel column (200–300 mesh, 350 g, 6.0 i.d. \times 100 cm), eluting with petroleum ether and EtoAc mixtures of increasing polarity. Five fractions were obtained. From fraction 1 with petroleum, oil was obtained and fractions 2 and 3 were obtained with petroleum ether/EtoAc (9:1). Fractions 2 and 3 were purified by a further silica gel column (200–300, mesh) with petroleum ether/EtoAc (19:1), to give a light-yellow oil-like liquor, compound 1, (200 mg). The latter two fractions were obtained with petroleum ether/EtoAc (8:2) and were recrystallized from MeOH to give isopsoralen (compound 2, 10 mg) and psoralen (compound 3, 62mg).

Thirty-two grammes of chloroform extract were chromatographed on a silica gel column (200–300 mesh, 400 g, 6.0 i.d. \times 100 cm) each with 3.0 l of a developing solvent system of petroleum ether/EtoAc (9:1, 8:2, 7:3 and 6:4, v:v) and fractions 1 and fraction 2 were obtained with petroleum ether/EtoAc (8:2). The two fractions were crystallized and recrystallized from MeOH to give isopsoralen (compound 2, 218.3 mg) and psoralen (compound 3, 215.2 mg). Four fractions were obtained with petroleum ether/EtoAc (7:3) and merged into two fractions according to TLC analysis. Corylifolin (com-

pound 4, 10 mg) and corylin (compound 5, 25 mg) was obtained from fraction 2 after recrystallization. Psoralidin (compound 6, 23 mg) was obtained from fractions with petroleum ether/EtoAc (6:4).

2.5. Recording spectra

Mass spectra were recorded with an HP5989 mass spectroscopic instrument. Melting points (MP) were determined on a WRS-1B melting point apparatus, which was not calibrated. Ultraviolet (UV) spectra were recorded with a UV-260 spectroscopic instrument, and methanol was used as solvent and a quartz cuvette was used. IR-spectra were recorded on a Nicolet 5DX IR spectrometer. Samples were prepared for IR spectroscopy by incorporating the crystals into a KBr disc. Nuclear magnetic resonance (NMR) spectra were recorded with a Bruker AM-400 and tetramethylsilane (TMS) was used as an internal standard.

2.6. Antioxidant activity

The lard without any additives was used as the substrate to evaluate the antioxidant activity of the powder, extracts and components of *P. corylifolia* L. Antioxidant activities of the six compounds and the extracts from the herb were added to lard in OSI sample tubes. The OSI instrument was set at 100 °C and the air flow rate was fixed at 20 l/h; BHT and α -tocopherol were used as comparison samples.

The effect of the structures on activity, interpreted as the protection factor (Pf), was calculated according to the expression: Pf = the induction period (IP) of lard with antioxidant/the IP of lard without antioxidant. All IP results were means of four parallel experiments.

3. Results and discussion

The chemical structure confirmation of the components from the *P. corylifolia* L. was accomplished by comparing the melting point, UV, IR, Mass, ^1H and ^{13}C NMR data obtained to those published.

Compound 1 was isolated and rechromatographed on a silica gel column with petroleum ether/EtoAc (19:1) as yellow oil-like liquor. It had maximum absorption at 262 nm in its UV spectrum. Its molecular weight was 256, determined by mass spectrum. Its IR (KBr, cm^{-1}) spectrum had strong absorption at 3397 (OH), 1720 (C=O), 1611, 1591, 1513 (C=C). Its detailed spectral data of IR, MS, ^1H and ^{13}C NMR listed in Table 1 agree well with the reported compound, bakuchiol (Labbe, Faini, & Coll, 1996; Mehta, Nayak, & Sukh, 1973).

Compound 2 was isolated and recrystallized from MeOH as white powder, MP: 137–138, MS m/z (%): 186(100), 158(85.00), 102(49.00), 130(31.36), 51(20.90),

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