Food Chemistry 44 (1992) 119–122

Antioxidant properties of extracts from tanshen (*Salvia miltiorrhiza Bunge*)

M. H. Gordon & X. C. Weng

Department of Food Science and Technology, Reading University, Whiteknights, PO Box 226, Reading RG6 2AP, UK

(Received 11 October 1990; revised version received and accepted 23 April 1991)

Powdered tanshen (*Salvia miltiorrhiza Bunge*) has strong antioxidant properties in lard. Powdered tanshen is less effective than rosemary but more effective than other plant materials with antioxidant properties including licorice, tea and ginseng. A hexane extract from tanshen is a very effective antioxidant both at 100°C and at 180°C. The tanshen extract acts as a primary antioxidant showing a synergistic effect with citric acid, and also stabilising lard containing tocopherols.

INTRODUCTION

The roots of the herb tanshen (*Salvia miltiorrhiza Bunge*) have been traditionally used in Chinese medicine for many ailments. The herb is a sedative, with antimicrobial, antispasmodic, anti-inflammatory properties and it also reduces aggregation of blood platelets and contributes to the prevention of myocardial infarction (Lee *et al.*, 1987). The composition of tanshen extracts has been studied in detail by several groups (Wang, 1984; Luo *et al.*, 1988*a*, 1988*b*; Zhang *et al.*, 1990). Extracts from tanshen with organic solvents have been found to possess antioxidant properties, and purified quinones including dihydrotanshinone I, tanshinone I, methylenetanshinquinone, cryptotanshinone, tanshinone IIA, tanshinone IIB and danshenxinkun B from these extracts have been found to contribute to this effect (Zhang *et al.*, 1990).

This study is concerned with an investigation of the activity of extracts from tanshen with the aims of comparing their activity with that of other plant extracts and understanding the mechanism by which they are active.

MATERIALS AND METHODS

Tanshen was purchased from a Chinese medicine company (The Herbal Medicine Company) in Zhengzhou, People's Republic of China. The roots were washed with water and freeze-dried. The dry tanshen was ground in a pestle and mortar, and sieved through a 20-mesh sieve.

The tanshen powder was extracted in a Soxhlet extractor with hexane, chloroform and methanol in sequence for 6 h each. The three solvent extracts were filtered and evaporated to dryness. The yields of the Soxhlet extracts were 1·5% with hexane, 0·8% with chloroform and 1·1% with methanol.

Lard was rendered from fresh pig fat tissue.

Ferric palmitate was prepared by a method based on that of Heaton and Uri (1961). Sodium hydroxide (4·2 g) was dissolved in distilled water (100 ml) and palmitic acid (25·6 g) was added and stirred for 5 min. The mixture was diluted to 400 ml with water and heated with stirring to 90°C. The sodium palmitate solution was poured into a beaker containing ferric chloride hexahydrate (20·0 g). The mixture was heated to 80°C with stirring. The brown precipitate was removed by filtration after 20 min and washed three times with hot distilled water at 80°C. The solid was dried in a vacuum oven at 80°C for 8 h to yield a chloroform-soluble product melting at 88–89°C. Analysis by atomic absorption spectrophotometry using a Pye Unicam SP9 spectrophotometer indicated that the product contained 6·41% iron compared with the theoretical value for ferric palmitate of 6·80% iron.

The oxidative stability of lard was assessed by means of the induction period determined with the Metrohm 617 Rancimat at 100°C, with the air flow rate at 16 litre/h. The glass tubes were washed with detergent solution twice, boiled in sodium hydroxide solution (2%) for 1 h, then soaked overnight in concentrated hydro-

Food Chemistry 0308–8146/92/$05.00

chloric acid. The tubes were then washed with water and dried before use. Six determinations of the induction period of a lard sample containing tocopherols showed the coefficient of variation was only 2·9% after this washing procedure. Iodine values were determined by the IUPAC Method (Paquot, 1979).

Tocopherols were analysed by high performance liquid chromatography using a 250 × 4·6 mm column packed with Apex silica (5 μm diameter), isocratic elution with isopropanol (1·5%) in hexane, and a Perkin Elmer 3000 Fluorescence detector with excitation wavelength 290 nm and emission wavelength 330 nm.

DL-α-Tocopherol was purchased from Sigma Chemical Company Ltd (Poole, UK).

RESULTS AND DISCUSSION

Tanshen powder had a marked antioxidant effect at 100°C with the induction period for lard increasing from 4·3 h to 41·5 h in the presence of 5·1% tanshen powder (Fig. 1). Comparison of the induction periods of lard containing various powdered plant materials at 0·5% showed that the order of antioxidant effectiveness was rosemary > tanshen > licorice > tea > ginseng with induction periods of 23·2 h, 17·5 h, 10·1 h, 5·2 h, 4·3 h compared with 2·3 h for the control.

The hexane extract from tanshen was also a highly effective antioxidant but at a much lower concentration than tanshen powder. A strong increase in antioxidant effectiveness up to 0·46% hexane extract was observed (Fig. 2). This contrasts with α-tocopherol which did not show a strong increase in antioxidant effectiveness above 0·02% with the induction period being 18·5 h for lard containing 0·021% α-tocopherol and 20·5 h for a sample containing 0·041% α-tocopherol. The lack of any strong increase in antioxidant effects of α-tocopherol at levels above 0·02% has been recognised by many workers (Loliger, 1983). Interpolation of the data from the tanshen extract indicated that approximately 0·015% of the hexane extract had a similar antioxidant effect to 0·021% α-tocopherol. Further extraction of the tanshen residue with chloroform and methanol produced extracts that were less effective than the hexane extract (Fig. 2). Thus the components with the greatest antioxidant properties were extracted by hexane.

The induction periods at 100°C of lard samples containing 2·2 ppm ferric ions (32·3 ppm ferric palmitate)

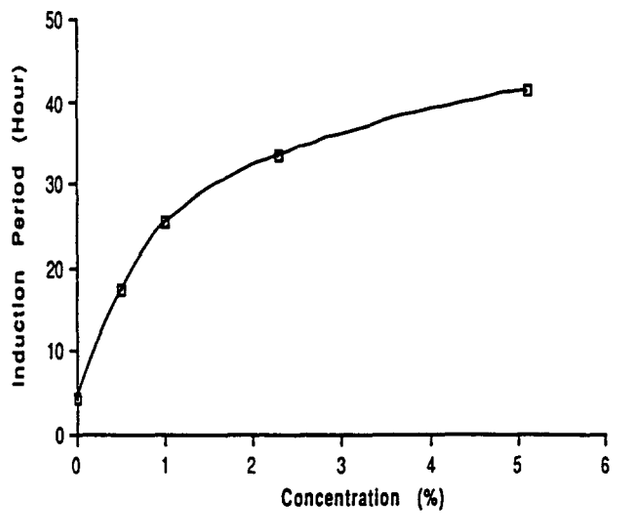

Fig. 1. The effect of tanshen powder on the induction period of lard at 100°C.

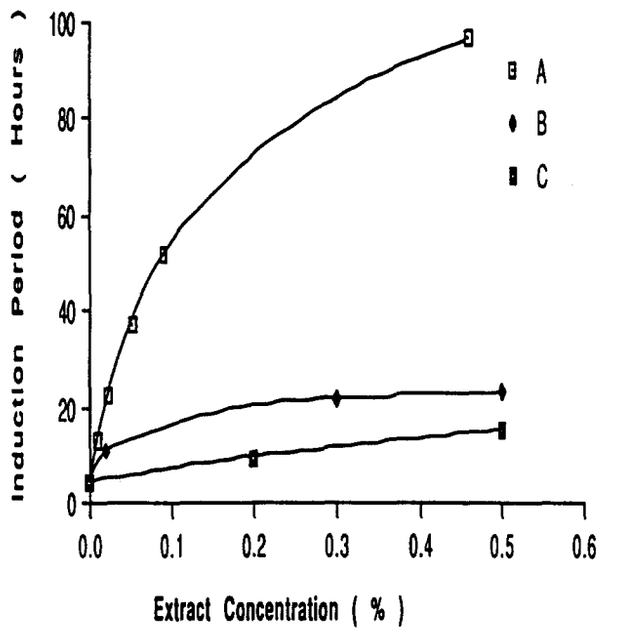

Fig. 2. The effect of hexane (A), chloroform (B) and methanol (C) extracts from tanshen on the induction periods of lard at 100°C.

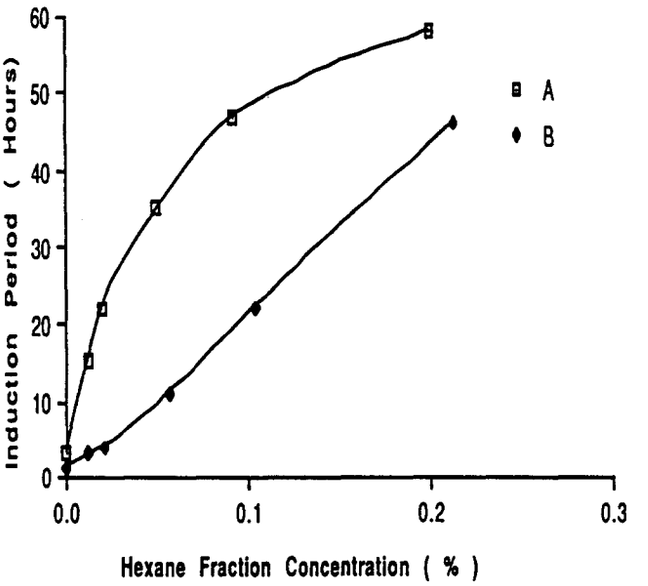

Fig. 3. The effect of hexane extract from tanshen on the induction periods of lard containing ferric ions (2·2 ppm): (A) with citric acid (0·01%); and (B) without citric acid, at 100°C.

increase as increasing concentrations of hexane extract are included (Fig. 3). However, the addition of citric acid has a synergistic effect with the hexane extract. The induction period for lard containing 2·2 ppm ferric ions is increased from 1·1 h to 3·3 h by the addition of 0·012% hexane extract, but lard containing a combination of citric acid (0·01%) and hexane extract (0·011%) has an induction period of 13·3 h. This synergy is clear evidence that the hexane extract is acting as a primary antioxidant. Since it acts by chelation of pro-oxidant metal ions, citric acid does not have any antioxidant properties in the absence of metal ions. The induction period at 100°C for lard containing 0·05% hexane extract was 37·0 h compared with 33·5 h when 0·01% citric acid was present. The induction period of lard containing 0·02% α-tocopherol and 0·017% hexane extract was 27·2 h compared with 18·5 h for lard containing 0·02% α-tocopherol and 20 h for lard containing 0·017% hexane extract (interpolated from Fig. 2). The small degree of increased stabilisation of α-tocopherol by the tanshen extract is consistent with that expected from a mixture of primary antioxidants.

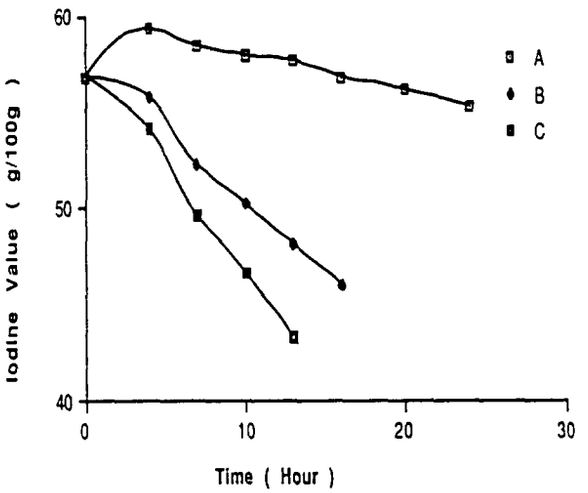

Fig. 4. The stability of lard containing: (A) the hexane extract from tanshen (0·2%), (B) α tocopherol (0·02%), compared with (C) pure lard at 180°C.

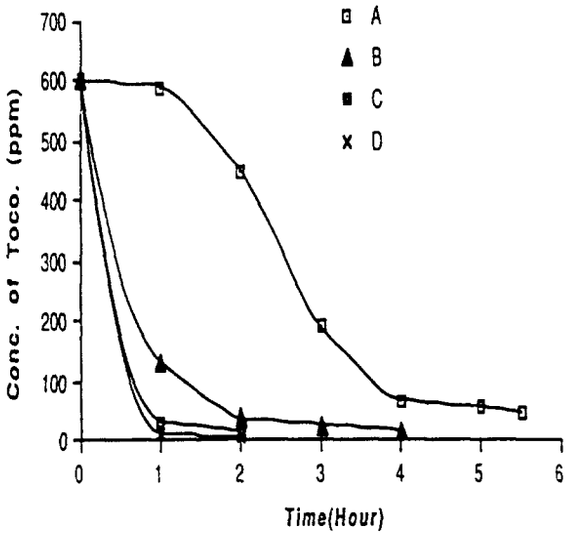

Fig. 5. Changes in the tocopherol contents of lard containing 600 ppm tocopherol in the presence of the hexane extract from tanshen at various concentrations: (A) 0·34%; (B) 0·15%; (C) 0·058%; (D) 0·00%, at 180°C.

The hexane extract from tanshen was also effective in stabilising oil heated on a hot-plate at 180°C which is a typical temperature used in deep-fat frying. The addition of 0·2% tanshen extract stabilised the lard compared with the control whereas 0·02% tocopherol was relatively ineffective (Fig. 4). Oil deterioration at elevated temperatures was monitored by iodine value determinations, since this parameter is a measure of the loss of unsaturated fatty acids under these conditions (Waltking & Zmachinski, 1970).

The tanshen extract also stabilised tocopherols which were present in lard heated at 180°C (Fig. 5). A concentration of 0·34% of the tanshen extract extended the tocopherol half-life from 0·3 h to 2·5 h at this temperature.

Hence, the above studies have shown that the hexane extract from tanshen is effective as a primary antioxidant. The extract may have important commercial applications as an antioxidant at temperatures up to frying oil temperatures.

Quinones such as dihydrotanshinone I have been isolated from tanshen after extraction with non-polar organic solvents, and shown to have strong antioxidant properties (Zhang *et al.*, 1990). These quinones may act as primary antioxidants by reacting relatively rapidly with lipid-free radicals to form radicals which are stabilised by electron delocalisation both around the aromatic ring and onto an oxygen atom. Hence, this reaction interrupts the autoxidation chain reaction.

REFERENCES

Heaton, F. W. & Uri, N. (1961). The aerobic oxidation of unsaturated fatty acids and their esters. *J. Lipid Res.*, **2**, 152–60.

Lee, A-R., Chang, W-L., Lin, H-C. & King, M-L. (1987). Isolation and bioactivity of new tanshinones. *J. Natural Products*, **50**, 157–61.

Loliger, J. (1983). Natural antioxidants. In *Rancidity in Foods*, ed. J. C. Allen & R. J. Hamilton. Applied Science, London, pp. 94.

Luo, H., Gao, J. & Zheng, J. (1988*a*). Relationship between structure and antibacterial activities of tanshinones and related compounds. *J. China Pharmaceutical Univ.*, **19**(4), 263–5.

Luo, H. W., Hu, X. J., Wang, N. & Ji, J. (1988*b*). Platelet aggregation inhibitors from *Salvia miltiorrhiza Bunge*. *Acta Pharmaceutica Sinica*, **23**(11), 830–4.

Paquot, C. (1979). *IUPAC Standard Methods for the Analysis of Oils, Fats and Derivatives*, 6th edn, Pergamon Press, Oxford.

Waltking, A. E. & Zmachinski, H. (1970). Fatty acid methodology for heated oils. *J. Amer. Oil Chem. Soc.*, **47**, 530–4.

Wang, Y. (1984). Research of natural antioxidants. MSc dissertation, Zhengzhou Grain College, China.

Zhang, K-Q., Bao, Y., Wu, P., Rosen, R. T. & Ho, C-T. (1990). Antioxidative components of tanshen (*Salvia miltiorrhiza Bunge*). *J. Agric. Food Chem.*, **38**, 1194–7.