

# Nutrient Composition of Deoiled Sandal Seed Meal: Minerals and Amino Acids

K. H. SHANKARA NARAYANA\* and K. PARTHASARATHI, Sandal Research Centre, Bangalore-560003, India

## ABSTRACT

Deoiled seed meal prepared from the decoated seeds of sandal (*Santalum album* Linn) contains 52.5% of protein and about 5% mineral constituents. The proteins are rich in essential amino acids. The deoiled sandal seed meal could be of utility as a feedstuff for farm animals.

## INTRODUCTION

Sandal (*Santalum album* Linn) is a species which fruits profusely. The annual availability of sandal seed is nearly 20,000 tons in the states of Karnataka and Tamilnadu, the main sandal growing states in India. The seeds contain about 50 to 60% of a drying oil (1-3) rich in santalbic glyceride (4-6). The seed oil can be made to react with zinc chloride, yielding a dark plastic solid which, when dissolved in benzene, forms an ideal base for insulation tapes (7). Resins like colophony and copal can be dissolved in the oil at 180-200 C, yielding an excellent orange colored varnish (7) possessing good insulating properties. The santalbic acid of the glyceride in the oil was found to form with dimethyl sulfate a molecular inclusion complex possessing detergent action (8). In this paper the amino acid and mineral composition of the deoiled sandal seed meal are reported, and the potential utility of the meal as a feedstuff for farm animals is suggested.

## MATERIALS AND METHODS

For analysis, a composite sample of the deoiled sandal seed meal, prepared from 500 g of decoated seeds obtained from 10 mature sandal trees (50 g from each tree), was used.

The crushed decoated seeds were extracted with petroleum ether (boiling range 60-80 C) in a soxhlet for 24 hr, powdered and the powder passed through a sieve (180 micron). The sieved powder was used for the analysis of amino acids and minerals.

Following the procedure of Laidlaw and Smith (9), the acid hydrolysis of the deoiled seed meal (20 g) was done using 6N HCl. The amino acids in the hydrolysate were adsorbed on an Amberlite IR-120 resin column and eluted using 2N ammonia. The ammonia solution of the amino acids was evaporated under vacuum (20 mm Torr), the residue was dissolved in 0.01N HCl and the amino acids analyzed using the amino acid analyzer (Model LKB-4400) along with standard amino acids.

The protein content (52.5% on the basis of air dry weight) of the deoiled seed meal was determined by the standard Kjeldahl procedure, using the conversion of N to protein constant of 6.25. Mineral constituents ( $K_2O$ , CaO, MgO, ZnO, CuO, MnO,  $Fe_2O_3$  and  $P_2O_5$ ) were determined following standard procedures (10,11).

## RESULTS AND DISCUSSION

Table I shows the relative percentage amino acid composition of the deoiled sandal seed meal hydrolysate. Table II gives the data showing the mineral composition of the meal in comparison with some common feedstuffs (12). It can be seen that the proteins in the deoiled meal are particularly rich in (i) phenylalanine and leucine, essential amino acids,

\*To whom correspondence should be addressed.

TABLE I

Relative Percentage Amino Acid Composition of Deoiled Sandal Seed Meal Hydrolysate

Amino acid	Relative percentage
Arginine	6.6
Lysine*	4.5
Histidine*	1.7
Phenylalanine*	8.7
Tyrosine	6.0
Leucine*	12.7
Isoleucine*	5.5
Methionine*	1.6
Valine*	6.3
Cystine	0.3
Alanine	4.0
Glycine	9.1
Proline	6.9
Glutamic acid	11.7
Serine	4.7
Threonine*	2.6
Aspartic acid	6.5

\* Essential amino acid.

TABLE II

Percentage Mineral Constituents in Deoiled Sandal Seed Meal in Comparison with Some Common Feedstuffs

Description	$K_2O$	CaO	MgO	MnO	ZnO	CuO	$Fe_2O_3$	$P_2O_5$
Deoiled sandal seed meal	1.80	0.84	0.57	0.003	0.008	0.002	0.023	1.80
Cottonseed <sup>a</sup>	1.01	0.44	0.51	—	—	—	0.030	1.20
Oat hay <sup>a</sup>	2.27	0.43	0.20	—	—	—	—	0.35
Wheat hay <sup>a</sup>	1.29	0.22	0.66	—	—	—	—	1.75
Sorghum hay <sup>a</sup>	2.24	0.61	0.41	—	—	—	—	0.51
Guinea grass <sup>a</sup>	3.40	1.81	0.53	—	—	—	—	0.57

<sup>a</sup>Reference 12.

The deoiled sandal seed meal contains 8.4% N and 5.1% ash.

and (ii) glycine and glutamic acid, among the non-essential amino acids. The seed meal is richer in protein content (52.5%) than some common oilseed cakes (13) (groundnut, 48.3%; safflower, 34.1%; gingelly, 35.1%, and linseed, 28.4%). The mineral composition of the meal is also quite comparable to that seen in some common feedstuffs (Table II). The meal is very rich in phosphorous.

The status of minerals, proteins and amino acids in the deoiled sandal seed meal, in comparison with that of the common feedstuffs, points to the potential utility of the meal as a good feedstuff for farm animals.

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## Antioxidant Effects of Chlorophyll and Pheophytin on the Autoxidation of Oils in the Dark. II. The Mechanism of Antioxidative Action of Chlorophyll

YASUSHI ENDO\*, RIICHIRO USUKI<sup>1</sup> and TAKASHI KANEDA<sup>2</sup>, Department of Food Chemistry, Faculty of Agriculture, Tohoku University, Sendai, Japan 980

### ABSTRACT

To understand the mechanism of the antioxidant effect of chlorophyll on the autoxidation of oils in the dark, antioxidant activities of several derivatives of chlorophyll were compared. Antioxidant activities were observed in chlorophyll derivatives such as protoporphyrin methyl ester and its magnesium chelated compound. Porphyrin seems to be an essential chemical structure for the antioxidant activity of chlorophyll. Chlorophyll did not decompose the hydroperoxides, but reduced free radicals such as 1,1-diphenyl-2-picrylhydrazyl. Electron spin resonance spectrum of the  $\pi$ -cation radical was recorded during the oxidation of chlorophyll in methyl linoleate solution. These observations suggest that chlorophyll may act as a hydrogen donor to break the chain reaction.

### INTRODUCTION

In a previous paper (1), we reported the antioxidant activities of chlorophyll (CHL) and pheophytin (PHY) on the autoxidation of methyl linoleate (ML) in the dark. The antioxidant effect of CHL also was observed in triglycerides such as rapeseed and soybean oils. In this paper, to understand antioxidant effects in CHL and PHY on the autoxidation of oils in the dark, the following studies were carried out:

- (i) The relationship between structure and antioxidant effects of CHL derivatives;
- (ii) The reaction of CHL and PHY with hydroperoxides and 1,1-diphenyl-2-picrylhydrazyl (DPPH), and
- (iii) The electron spin resonance (ESR) spectrum of CHL in ML during autoxidation

Based on the results obtained, we discuss the possible mechanism of the antioxidant effect of CHL on the autoxidation of oils in the dark.

### MATERIALS AND METHODS

#### Materials

The preparation of CHL, PHY and ML has been described previously (2).

Protoporphyrin methyl ester (PRO) was obtained by esterification of sodium protoporphyrin purchased from

\*To whom correspondence should be addressed at Department of Food Chemistry, Faculty of Agriculture, Tohoku University, 1-1 Amamiyamachi-Tsutsumidori, Sendai, Japan 980

<sup>1</sup>Present address Shokei Women's Junior College, Sendai, Japan.

<sup>2</sup>Present address Koriyama Women's College, Koriyama, Japan.

Nakarai Chemical Ltd. Magnesium chelated porphyrin methyl ester (Mg-PRO) was prepared after inducing magnesium (Mg) in PRO by the degradative Grignard reaction (3).

Pyrrole and magnesium chloride (MgCl<sub>2</sub>) were purchased from Tokyo Kasei Industries Ltd. and Wako Pure Chemical Industries Ltd. (Tokyo, Japan), respectively.

Methyl linoleate hydroperoxides (MLHPO) were prepared from autoxidized ML by silicic acid column chromatography with a series of n-hexane-diethyl ether mixtures as solvent system.

#### Autoxidation

Some derivatives and structural constituents of CHL were diluted with n-hexane-diethyl ether solutions, except MgCl<sub>2</sub>, which was diluted with methanol, and added to one gram of ML in a small beaker ( $\phi$  27 mm). These samples were incubated at 30 C in the dark. In addition to CHL A, PHY A, PRO and Mg-PRO as typical CHL derivatives, MgCl<sub>2</sub> as Mg ion and pyrrole as one structural component of porphyrin were used, respectively, for oven tests. Addition levels of derivatives and structural constituents of CHL were  $2.2 \times 10^{-7}$  mol/g ML, except for pyrrole, which was of  $8.8 \times 10^{-8}$  mol/g ML. Peroxide value (PV) and carbonyl value (CV) of each sample were determined after autoxidation.

#### Degradation Test of Methyl Linoleate Hydroperoxides

1% (w/w) CHL A and PHY A were added to 10% (w/w) MHLPO in ML solution in a small beaker and then incubated at 30 C in the dark.

#### Reduction Test of 1,1-diphenyl-2-picrylhydrazyl

CHL A and PHY A ( $4.4 \times 10^{-5}$  and  $1.1 \times 10^{-4}$  M) in carbon tetrachloride (CCl<sub>4</sub>) solutions were added to  $4.0 \times 10^{-4}$  M DPPH in CCl<sub>4</sub> solution, and absorbance at 516 nm of DPPH of these mixtures was monitored during incubation at room temperature (4).

#### ESR Measurement

200  $\mu$ l of  $10^{-5}$  mol CHL A in benzene solution and  $10^{-5}$  mol iron chloride (FeCl<sub>3</sub>) as an oxidative initiator were mixed in 300  $\mu$ l of ML. After vigorously shaking this mixed solution, its ESR spectrum was measured at room tempera-