

117
BIOCHEMICAL STUDIES ON SOYBEAN OIL

BY

Nadia Yehia Ahmed Attia

B. Sc. Agriculture

Thesis

Submitted in Partial Fulfillment of The
Requirements For The Degree of

MASTER OF SCIENCE
Agricultural Biochemistry

Faculty of Agriculture
Ain Shams University

1975

APPROVAL SHEET

Name : Nadia Yehia Ahmed Attia

Title: Biochemical Studies On Soybean Oil

M. Sc. Thesis Approved by:

Prof. Dr. H. H. El-Sayed

Prof. Dr. M. M. El-Sayed

Prof. Dr. A. A. El-Sayed

Date: 2 / 7 / 1975.



ACKNOWLEDGEMENT

The present investigation was suggested and kindly supervised by Dr. M.A.M. Kamal, Prof. of Biochemistry, Faculty of Science, and Dr. S.I. El-Hinnawy ,Prof. of Biochemistry, Faculty of Agriculture ,Ain Shams University, to whom their help, continuous interest and constructive suggestions were highly acknowledged.

The kind help and continuous co-operation offered by Dr. K. Roushdy Hussein, lecturer of Biochemistry, Faculty of Agriculture, Ain Shams University, was quite helpful and beneficial.

<u>C O N T E N T S</u>		Page
I - INTRODUCTION		3
II - REVIEW OF LITERATURE		
A - Extraction of the oil.		
B - Solubility of soybean oil.		
C - Storage and autoxidation.		
D - Gas liquid chromatography of fatty acids and its methyl esters.		
E - Thin layer chromatography of the triglycerides.		
F - Phosphorous content.		
III - MATERIALS AND METHODS		
- Preparation and Extraction		21
- Physical and chemical properties		22
Refractive index - specific gravity -		
Acid value - Saponification value -		
Iodine value - Unsaponifiable matter		
Peroxide value - Thiobarbituric acid test - Thiocyanate method.		
- Solubility of soybean oil		29
- Identification and determination of fatty acids in soybean oil by GLC.		30
- Thin layer chromatography TLC		32
- Determination of Phosphorous		38

	Page
IV - RESULTS AND DISCUSSION	40
Part I : a. Efficiency of Different Organic Solvents in the extraction of Oils from soybean seeds	40
b. Effect of the Solvent of Extraction on some of the Physical and Chemical Properties of the oil produced.	45
c. Solubility of Soybean oil	51
Part II: Stability of soybean seed oil	57
Part III: a- Fatty acid components of soybean seed oil.	64
b- Thin layer chromatography	72
c- Determination of phosphorous	78
V- SUMMARY AND CONCLUSION	80
VI - REFERENCES	85

INTRODUCTION

The soybean ,an immigrate crop from China had been spread over many countries in the world over the past 40 years. Soybean seeds as high content of excellent protein ranging from 39 to 44 percent and moderate content of oil ranging from 18 to 22 percent which is useful for edible and facilitate its complete importance for the nutritional point of view.

Although Soybean oil does not quite measure up in quality to cotton, peanut, corn and some other vegetable oil crops, research has modified these deficiencies to such an extent that over 50% of all the fats and oils consumed in the U.S.A. are soybased. The soybean has resulted in the development of a world wide marketing and processing technology covering the seeds and its two main products. i.g. oils and proteins. The meal and oil fractions are finding their way into more and more highly sophistications for human use.

Soybean have made a great advanced in the past 40 years and hold a dominant position in the oil seeds industry. The price of soybean oil and meal has markedly influenced the price of other oil seed products. There is a tendency among investigators and producers of other oil seeds to feel they are in competition with soybean and perhaps they are but in a broader since they are all

in competition with each other. Soybean have an advantage over other oil seeds because of their low cost of production and their easy adaptability to solvent extraction, moreover it is an important contribution to the animal feed industry.

Vegetable oils are usually used for cooking or manufacture of margarine, and flavor enhancer such as salad applications. The steady increase in soybean oil consumption and the steady decrease in butter, lard and cotton seed oil usage are impressive. Since oil is so essential to the dietary and traditional meal of most of the world's population, and with a great deal of the population consuming only a fraction of its potential, the future of all fats and oils seems most secure.

The aim of this investigation is to study the major chemical and physical properties of the soybean oil produced locally. Such study necessitates the study of extraction of oil, as well as its stability towards autoxidation and rancidity. Also this identification of soybean fatty acids, triglycerides and its phospholipids will be tackled thoroughly. Such study will undoubtedly furnish the future use of soybean oil in Egypt to cover our continuous lacking demands of vegetable oils.

REVIEW OF LITERATURE

In fact there is a tremendous and voluminous work carried out on soybean oil so it will be represented in six parts to facilitate its presentation.

A- Extraction of the Oil:

Bailey (1945), stated that solvent extraction was relatively more advantageous in the processing of soybeans and other oil seeds.

Karnofsky (1948), noticed that the solvent extraction method was very complicated because of the total extractable materials varied in its quantity and composition, depending on the solvent and other factors. He added that the composition of the extractable materials were the glycerides in the first fraction followed by other fractions containing increasing amounts of the non-glyceridic materials. He also found that the rate of extraction depended markedly on several factors. It was quite clear from his results that the mechanism of extraction appeared to be a combination of two processes namely diffusion and dialysis.

Raghumathn (1953), referred to the solvent extraction process as an excellent tool for obtaining oils from the oily seeds.

Othmer and Jactionen (1959), noticed that the mechanism of extraction of soybean oil, in which they used different solvents, the amounts of the residual oil in cakes and the speed of extraction processes were exponential functions of the duration of extraction.

Hsia and Cho (1959), used ethyl acetate to extract oil from soybean cake. The extracted oil was measured to determine the yield and the thickness of the soybean cake which was inversely proportional to the yield of extraction. They found the extraction was complete when a cake 0.28 mm. thick had been extracted (soybean/ ethyl acetate weigh ratio 1 : 3), the yield did not change with temperature in the range of 10-15°C indicating that room temperature could conveniently be used and fifteen minutes were nearly sufficient for complete extraction for the definite thickness of the cake.

Arnold and Choudhury (1960), studied the rate of extraction of soybean with pure hexane, pure benzene and two varieties of commercial hexane - commonly used in oil extraction at 135 - 140 °F. They found that extraction of soybean by pure hexane was relatively slower than by the other two commercial hexanes, and during the first 60 minutes of extraction by benzene the process was comparatively rapid than by the other solvents. At the end of

60 minutes of extraction the amount of oil extracted by benzene was slightly more than that extracted by pure hexane but definitely less than that extracted by the commercial hexane.

Arnold and Choudhury (1962), extracted soybean oil from flakes in a laboratory scale, in glass extraction apparatus with four hydrocarbon solvents and the amount of the extracted oil was determined at 10 minutes intervals for 60 minutes of extraction. They observed that the extraction rates of solvents increased in the following order, isopentane, pentane, isohexane and hexane and the quality of the oil extracted by the solvents showed no significant differences. They also reported that there was little difference in the quality of the oil produced by different solvents when they used 90, 95, 98, and 100% ethanol in the extraction of soybean flakes.

Grishina and Kaznetov (1969), used a mixture of chloroform and ethanol (2:1) for the extraction of oil from soybean seeds, and the results were compared with those for the extraction of oils from the same sources by pressing. They concluded that the present method gave slightly better results.

Gulbran et al (1970), described a method for the

extraction of soybean oil. The extraction method described utilized a percolating solvent (benzene and toluene) at low pressure (550 - 600) mm Hg with 100% yield in oil. They found that extraction is faster and more complete than in the classical process and operate at lower temperature giving a clear micella.

B- Solubility of Soybean Oil:

The new world process of extracting edible vegetable oils from oligeneous seeds was the solvent extraction process due to its higher yield than the hydrolic press process.

Roa et al (1955), studied the solubility of soybean oil in aqueous ethanol solution at various temperatures and found that the critical solution temperature (C.S.T.) increased with the moisture content of the alcohol and consequently the relationship was linear.

Roa et al (1957), investigated the solubility of 14 vegetable oils in different concentrations of aqueous 2-propanol at various temperatures. They concluded that there was a general relationship between fatty acid contents of the oils and critical solution temperatures.

Ramalingan (1959), reported that at ordinary temperatures absolute alcohol did not prove to be an efficient

solvent for vegetable oils since the solubility was even less than 10 grams of oil per 100 grams of alcohol. He also noticed that the addition of an efficient solvent to the alcohol, like n-hexane, increased the solubility of the oil and the solubility temperature were lowered.

C- Storage and Autoxidation:

Seiichi and Akio (1955), studied soybean oil and other vegetable oils which were heated at 60 - 220°C at a pressure of 6 - 8 mm. Hg under H₂ and after which they determined their physical and chemical characteristics, they noticed that the Ultra Violet Spectra showed that some changes occurred i.e. (conjugated dien) increased in the soybean oil.

Ichiro et al (1960), stored deodorized and decolorised soybean oil at 20, 50, 75, or 100 °C. They found that when the deodorized oil was stored, color reversion occurred earlier at higher temperatures, and then promptly the color faded, however the maximum color (lovibond red) was similar at 50 - 100 °C which was stable at 20 °C. The change of velocity of color reversion relative to the temperature was similar to the change of initial velocity of oil as expressed by peroxide value or oxygen. They also found that the color of decolorized oil was stable at 50° and faded at 75°C and 100° (more rapidly at 100 ° than 75°), when

the decolorized oil was heated above 100 ° (180-270°) the color faded more rapidly at higher temperatures under reduced pressure, but it faded only slightly in the air when heated above 25° color faded less in longer time.

Evans et al (1960), in their work on soybean oil found that the increase in the non-volatile carbonyl constant of freshly deodorized oils was proportional to the peroxide value of the oils before deodorization. The rate of loss of flavor and oxidative stability of the oil was in relation to the extent of carbonyl development.

Hoffman (1961), investigated the neutral volatile products obtained by stripping oxidized soybean oil by means of gas liquid chromatography. It was possible to isolate the flavor carrier proper and to complete the analysis to a point at which the structure 3-cis hexanal seemed highly probable due to the oxidation of linolenic acid. Synthesis and subsequent comparison with the neutral products confirmed the correctness of the assumption n-hexanal and 3-transhexanal, and this fractions plays no significant role in the reversion falvor.

Alf Tanger (1962), applied the spectrophotometric benzidine acetate method for aldehydes to palm and soybean oils. He stored the oils at 50° for a week and 4 to 13 weeks

at room temperature and judged for flavor by a test panel. He reported that there was no correlation between flavor deterioration and initial aldehyde values for oils stored up to 4 weeks, and oils of highest aldehyde values stored for 13 weeks had lowest test scores and measureable amounts of peroxides were found in 40% of these samples.

Pier Giovanni Garoglio (1964), noticed that in one year of experiments that soybean, peanut, cottonseed, sunflower and olive oils when analyzed every 7 days for peroxide and iodine values and oxygen content in container atmosphere, he noticed that low humidity, diffuse light, inert atmosphere and resistant materials for containers were the most suitable variables to improve conditions for storage of these oils.

Rutkowski et al (1965), studied both hydrolysis and oxidation of crude and degummed soybean oil. They found that hydrolysis on oil which has not been degummed is rapid and spontaneous during storage, degumming reduces hydrolysis even when the oil temperature increased during storage and when it is mixed with water.

Hill and Hamond (1965), studied the flavor components of soybean oil in the early stages of autoxidation. The flavor components were isolated by distillation. The dis-