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REFINED PROTEIN PRODUCTS FROM RAPESEED

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I do not think it would be wise of me to bore you at this time with the usual song and dance about the world population explosion, food shortage, starvation, malnutrition, and bridging the protein gap. But I am sure you will be amused to hear in 1970 the opening sentence of a paper published by Osborne and Mendel (1917) "The demand which the international shortage of food has created for cheaply produced and easily obtainable sources of all nutrients, and particularly of suitable proteins and fats, has directed attention (anew) to the possibilities of the soybean." I could have easily substituted rapeseed for soybean and used the same sentence as my opening remark today.

Over the past 15 years, there has been a significant commercial development in the utilization of soybeans and other oilseeds. This has been the introduction of refined protein products for use in either conventional processed foods or in totally new food items. Since the original technology for the production of these refined protein products has been mainly developed for soybeans, I will briefly describe the soy products and summarize the processes used in their production.

1. Soybean flours and grits: Soy flour is defined as the screened, graded product obtained after expelling or extracting the oil from selected, sound, clean and dehulled soybeans. The term "flour" generally signifies that the material has been ground finely enough to pass through a 100-mesh screen. The term "grits" refers to particles of larger size described in terms of U. S. standard screens which are:

Coarse	No. 10 to No. 20
Medium	No. 20 to No. 40
Fine	No. 40 to No. 80

There are five general types of soy flour (or grits) available commercially:

1.1 Defatted soy flour containing less than 1 percent of residual fat (ether extract)

1.2 Low-fat soy flour containing between 5 and 6 percent of residual fat and produced either as a result of partial removal of the oil or by adding soybean oil back to the defatted soy flour.

1.3 High-fat soy flour containing about 15 percent fat usually produced by adding soybean oil back to defatted soy flour.

1.4 Full-fat soy flour containing all of the oil originally present in the raw soybeans, usually from 18 to 20 percent.

1.5 Lecithinated soy flour which is a type of low- or high-fat soy flour in which lecithin is added to defatted soy flour at a specified level, usually in a range up to 15 percent (Anon. 1966). In general soy flours and grits contain 45 to 60 percent protein (depending on the fat content). Estimated production per year in the U.S.A. is 200 million pounds, excluding pet foods and specialty animal feeds. The selling price is in the neighborhood of 7 to 8 cents per pound.

2. Soy protein concentrates. Soy protein concentrate is defined as, "the product prepared from high quality, sound, clean, dehulled soybeans by removing most of the oil and water-soluble non-protein constituents and shall contain not less than 70 percent protein (N x 6.25) on a moisture-free basis". (Anon. 1966).

Soy protein concentrates are manufactured from hexane-defatted flakes or flour by three processes which differ only in the means utilized to immobilize the main protein fraction while separating the low molecular weight carbonhydrates, mineral matter and other minor constituents.

2.1 Water extraction process. In this process the proteins of the defatted soybean flour are immobilized by denaturation with moist-heat. The carbohydrates and other constituents are then extracted with water. The residue is pressed to remove excess liquor, then broken, dried and ground. This type of product would have no functional characteristics other than some ability to absorb water and fat. Its use would be restricted to incorporation in meat patties and meat loaf products.

2.2 Aqueous alcohol extraction process. This process depends on the fact that the protein components are insoluble in aqueous alcohol of about 60 to 80

percent concentration; while the carbohydrates and other components are. Protein concentrates prepared by this method will have lighter color than those produced by the water extraction process, but their functional characteristics will not be appreciably different. This type of product will be largely used in the baking industry and in some comminuted meat products.

2.3 Acid leach process. This method makes use of the long-known fact that the main soy proteins have minimum solubility in aqueous acid solutions at their isoelectric point of about pH 4.5. The residual material may be neutralized with food-grade sodium hydroxide to pH 6.7 to 7.1 and the resulting sodium proteinate may be spray-dried directly or may be first heated and bleached. Considerably more functional value is obtained with this method.

The yield of dried concentrate from any of these 3 processes is about 60 to 70 percent, based on the weight of defatted flour. Estimated production of soybean protein concentrates in the U.S.A. is 25 million pounds per year. Selling price is 18 to 24 ¢ per pound.

3. Soy protein isolates. Isolates are prepared from defatted soybean flakes or flour, which have had minimal exposure to moist heat in order to retain maximum water dispersibility of the protein. The protein is extracted with an aqueous medium which may vary in pH from near neutrality to an alkaline pH. The process variables such as liquid:solids ratio, pH, time and temperature are usually selected to obtain optimally economic, and not necessarily maximum yields. The aqueous extract is separated from the residue by some suitable means such as screening, filtering, or centrifugation. The pH of the clarified extract is then adjusted to about pH 4.5 with food-grade acid to precipitate the major proteins. The precipitated protein is washed with water, concentrated to a slurry or cake containing up to 30 percent solids. The concentrated slurry is either dried directly or is first neutralized with alkali then dried. Most protein isolates are sold in the neutral or sodium proteinate form since this product is water-dispersible and may be readily incorporated in many food products or formulations for maximal functional value. Spray-drying is most often used rather than drum or roller-drying.

Soy protein isolates contain 90 to 95 percent protein and sell

for about 35 to 40 cents per pound. Estimated production in the U.S.A. is 15 million pounds per year. Average yield is 30 to 40 percent based on the weight of defatted flour.

This was then the basic technology which had to be applied to rapeseeds to produce refined protein products similar to those from soy. Certain characteristics of the rapeseed, however, made the application of this technology not as straightforward as one might have liked it to be. The first is, of course, the presence of the goitrogenic and growth inhibitory factors (Daxenbichler et al, 1964) which had to be removed somehow from the protein fraction without being allowed to go into the major component of the seed, i.e., the oil. The other characteristic is the high fibre content of the non-oil fraction because of the size and nature of the rapeseed (Wetter, 1965). We are quite confident that the plant breeders will ultimately overcome both these difficulties but, for the time being, they had to be dealt with.

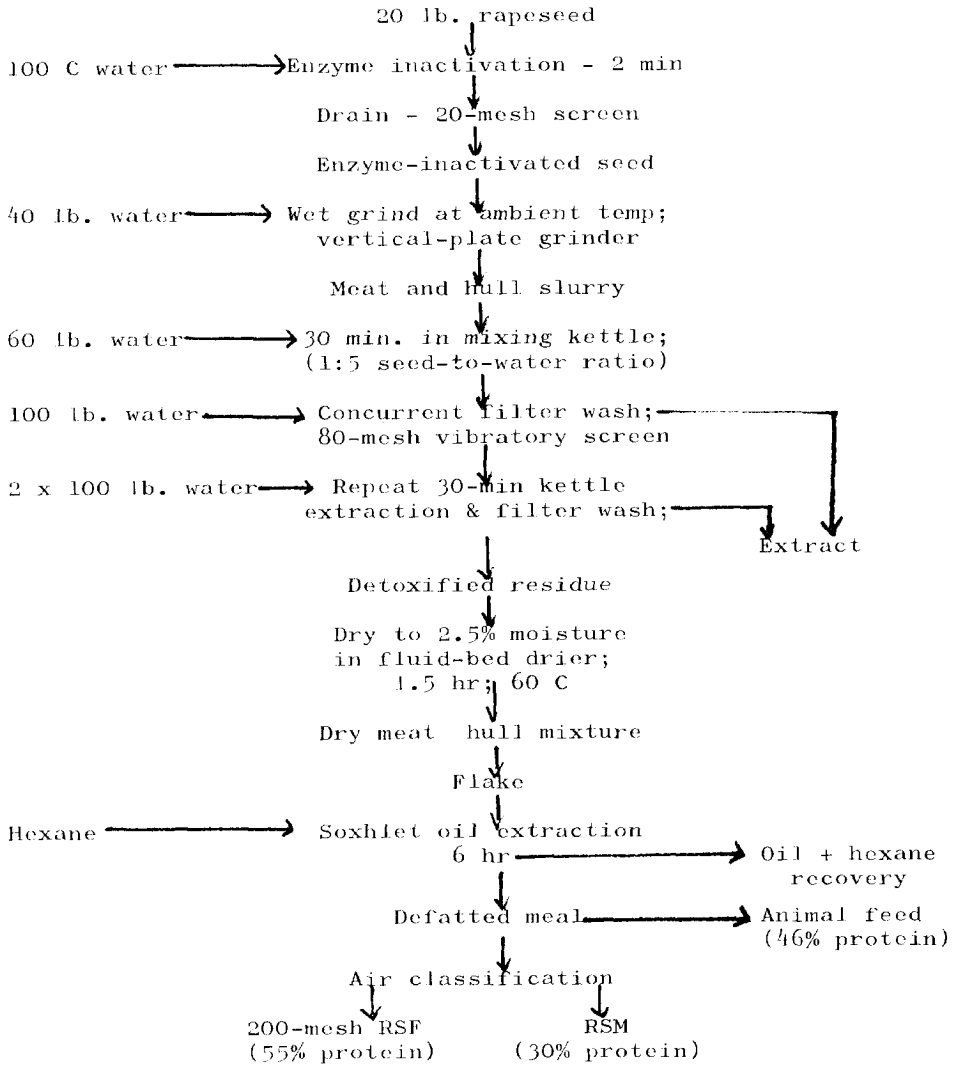
Early attempts towards detoxification of the rapeseed were naturally aimed a making its press-cake or meal more suitable for animal feeding, and we have to thank the animal nutritionists for pioneering this area. These early attempts have been thoroughly reviewed by Professors Bell and Belzile (1965). They concluded that, "methods of processing rapeseed in Canada result in production of myrosinase-free rapeseed meal containing unhydrolized thioglucosides. Such meal apparently is free of most of the undesirable properties if myrosinase is not reintroduced by other dietary ingredients or by intestinal bacteria. The potential antithyroid activity can be markedly reduced by heating, as revealed by the findings that most of the thioglucosides can be destroyed by two hours of either autoclaving at a steam pressure of 1.2 Kg/cm^2 or steam stripping at 110° C. Autoclaving resulted in severe damage to protein quality but steam stripping showed promise as a means of alleviating the risk of thioglucoside hydrolysis during digestion in the animal body." In 1967, Sallans and his co-workers at the Prairie Regional Laboratory of the Nation Research Council reported their study on the detoxification of rapeseed meal through catalytic decomposition of the thioglucosides by heating with salts of iron, copper or nickel (Sallans et al, 1967). A toxic, non-volatile nitrile was found to remain among the decomposition products in the meal.

Belzile and Bell (Bell and Belzile, 1965) also reported that hot water (90° C) extraction of rapeseed meal did not reduce its original toxicity.

With this background information, work was initiated in 1966, at the Food Research Institute, Canada Agriculture, to investigate

FIGURE 1

PROCESS FOR PREPARATION OF FLOUR AND MEAL FROM RAPESEED



the technical feasibility of producing a white, bland, defatted rapeseed flour free of both toxicity and fibrous seed coat. The results of this work have been reported by Eapen et al (1968, 1969) and by Tape et al (1970). The process developed for the production of rapeseed flour for human consumption is shown in Figure I. The main features of this process is the inactivation of the enzyme to immobilize the thioglucosides, then leaching the intact thioglucosides with water at ambient temperature (20°C) and finally the air classification of the defatted meal to obtain edible rapeseed flour containing 55 percent protein.

While this work was being reported from Ottawa, similar work on rapeseed meal detoxification was being conducted at the University of California by a group of workers from Chile (Ballester et al., 1970). They used samples of rapeseed meal from commercial sources to test two types of treatments: steaming and water extraction. Water extraction for 12 - 14 hours followed by a second extraction for 1 - 3 hours, both at room temperature, reduced oxazolidinethione contents by 84% and isothiocyanate contents by 77%. Van Etten (1969) used the same water leaching procedure to detoxify crambe seeds. In a more comprehensive investigation Van Etten and his co-workers (1969) at the Northern Regional Research Laboratory found that extraction of crambe seeds meal with aqueous acetone removed all toxicity regardless of hydrolysis or the pathway followed in the hydrolysis of the major thioglucoside of crambe, or in other words, whether the thioglucoside was still intact had undergone autolysis to give rise to the oxazolidinethione, or had given rise to nitriles.

It would, therefore, appear that aqueous extraction of flaked rapeseed in which the enzyme had been inactivated does wash out the intact thioglucosides thus eliminating the toxicity from the subsequently resulting meal. The disadvantages of this procedure include, of course, the additional steps of water-washing and subsequent drying of the wet seeds before extracting the oil. The other disadvantage is the removal of solids from the seed during aqueous extraction, Table I.

Lowering the fiber content in the high-protein rapeseed flour necessitates removal of the seed coats, cuticles or hulls either before oil extraction or after. Removal of these hulls before oil extraction, with the equipment available to us at the time, was impossible. It was, therefore, necessary to resort first to screening and then to air-classification. The Centri-Sonic Air Classifier manufactured by the Bauer Bros. Company of Springfield, Ohio, U.S.A., gave the best results and we are indebted to that company for their assistance and cooperation. The same air classifier, incidently, also gave the best results

TABLE I

AVERAGE YIELDS AND PER CENT REMOVAL OF SOLIDS AT EACH STEP OF PROCESS*

Process Step	Dry Wt. of Solids (Aver. of 12 lots)	Standard Deviation (12 lots)	Loss of Solids from Previous Step
	lbs		%
Echo seed	18.74	0.19	-
Enzyme inactivated	18.49	0.51	1.3
Ground seed	18.25	0.14	1.3
Aqueous extracted	14.26	0.10	21.8
Defatted	7.79	0.18	45.7
Rapeseed flour	3.89	-	-
Rapeseed meal	3.89	-	-

* from "Production of Rapeseed Flour for Human Consumption", by N. W. Tape, Z.I. Sabry and K. E. Eapen, CIFT Journal (in press).

TABLE II

PROXIMATE COMPOSITION AND MUSTARD OIL CONTENT OF RAPESEED FLOUR AND MEAL*

Product	Composition (Dry-Wt. Basis)						
	Protein	Fat	Fiber	Ash	NFE	Mustard Oil	
						Isothio.	Oxazol.
	(Nx6.25) %	%	%	%	%	mg/gm	mg/gm
RSF[1] (Jan 69)	54.7	8.3	8.0	9.5	19.5	nil	0.002
RSF (Apr 69)	54.9	9.1	5.9	7.3	22.3	Trace	0.003
RSF (Aug 69)	52.1	10.9	8.8	9.8	18.4	Trace	0.003
RSM[2] (Jan 69)	34.2	4.3	30.2	4.7	26.6	nil	0.001
RSM (Apr 69)	28.5	4.3	24.6	3.6	39.0	Trace	0.003
RSM (Aug 69)	31.4	5.2	27.1	4.9	31.4	Trace	0.003

1,2 Rapeseed flour and meal respectively.

* from "Production of Rapeseed Flour for Human Consumption" N. W. Tape, Z. I. Sabry and K. E. Eapen, CIFT Journal (in press).

in an independent study run outside the Department of Agriculture to find a process for upgrading commercial rapeseed meal by reducing the fibre content. Proximate composition of the rapeseed flour and meal obtained by air classification is shown in Table 2. Present work in this area at the Food Research Institute resulted in a slight modification in the procedure for preparation of rapeseed flour. We can now satisfactorily separate the hulls from the meats using a single disc attrition mill manufactured by the Bauer Bros. Co. of Springfield, Ohio U.S.A. - Again, I must acknowledge the assistance we received from this company in the development of a special set of plates or discs to make possible the complete separation of meats from hulls with minimum percentage of whole seeds or objectionable fines. After passing the seeds through the attrition mill, the hull-meat mixture is air-classified to a hull-free fraction and a hull fraction. The hull-free fraction is then used in the production of rapeseed flour as described before (Figure I) for the production of flour from whole seeds. The hull fraction constitutes about 35 percent of the weight of the original seed, and contains fines from the meats. Its composition, the possibility of its use as a high-energy feed, as well as other factors affected by this modification have yet to be determined.

This is then the first refined protein product from rapeseed, which may be compared with the well-known soy flour. Preliminary incorporation of rapeseed flour in different food items have been tried recently in our laboratories, using soy flour as a reference. Unfortunately statistical analysis of the sensory evaluation tests have not been completed yet. Slides taken of the different products seem to indicate that in most cases at least the appearance and physical characteristics of the products do not differ very much. (Aref and Noel, 1970).

The other refined protein product from rapeseed which has received quite a bit of attention is the rapeseed protein isolate According to accepted terminology, a protein isolate should contain from 90 to 95 percent protein. For unknown reasons, no interest in rapeseed protein concentrates containing 70 to 75 percent protein seems to exist a present.

The first attempts at the isolation of rapeseed protein are probably those of Pokorny and his group in Poland between 1963 and 1967 (Pokorny and Rutkowski, 1967). In this work, they determined the effects of the different factors involved in protein isolation such as pH, concentration, ratio of solids to liquid, temperature and contact time. In 1968, Shaikh and his co-workers in Pakistan (Shaikh et al, 1968) extracted defatted

TABLE III

SPECIES AND VARIETY DIFFERENCES IN NITROGEN SOLUBILITY INDEX OF THE MEAL, YIELD AND PROPERTIES OF THE ISOLATED PROTEIN, AND WHEY NITROGEN CONTENT[*]

Species and variety	Nitrogen content of meal[**]	Nitrogen solubility index	Isolated protein from meal			Nitrogen content of whey
			Yield of product	Nitrogen content	Color of product	
	%	%	g/100 g meal	%		% of total
Soybean, dehulled						
Portage	7.7	81.2	36.4	14.6	cream	10.3
Altona	8.2	86.2	40.2	15.0	cream	12.3
Rape						
Argentine	7.1	82.3	25.2	13.7	brown	32.3
Target	6.7	83.4	25.3	13.7	brown	30.3
Oro	6.4	85.7	26.6	12.8	brown	31.3
Turnip rape						
Polish	6.2	81.3	24.5	13.4	brown	27.4
Echo	6.1	83.2	26.3	12.9	brown	25.8
Zero erucic	6.9	79.5	24.5	13.6	brown	29.2
Flax						
Redwing	7.1	94.8	40.0	12.6	tan	23.2
Redwood	6.8	96.2	36.3	12.7	tan	27.2
Noralta	6.7	91.1	36.8	11.2	tan	29.4
Sunflower, dehulled						
Commander	10.3	92.4	52.6	14.5	green	17.2
Advent	10.0	95.9	54.0	14.1	green	19.4
Peredovik	9.3	95.3	52.3	13.4	green	19.8

[*] from: Isolated Proteins from Rapeseed, Flax and Sunflower Meals, F. W. Sosulski and A. Bakal, CIFT Journal, 2:28-32, 1969.

[**] Dry basis

rapeseed meal with aqueous sodium hydroxide at pH from 8 to 11, and precipitated the protein at pH from 4 to 6; pH 10 for the extraction and pH 4 for the precipitation proved most satisfactory. Sosulski and Bakal (1969) isolated protein from rapeseed, flax and sunflower meals and compared these isolates to soy protein isolates prepared in the same manner. Their results are shown in Table III. There was some concern at the time about the brown color of rapeseed protein isolates, but I understand that Sosulski and his group subsequently succeeded in eliminating the coloration from their isolates (Sosulski, 1970). Finlayson and his team at the Prairie Regional Laboratory of the National Research Council conducted the most extensive studies on the characterisation of rapeseed protein (Finlayson et al, 1969). Of special interest in the preparation of refined protein products from rapeseed is their finding that 25 percent of the defatted rapeseed meal nitrogen was water-soluble, that 45 percent of the meal nitrogen was soluble in sodium pyrophosphate at pH 7 and that about 70 percent of the meal nitrogen was soluble in 1 M sodium chloride at 5°C for 3 hours. In a recent report yet to be presented, Owen and his coworkers (Owen, et al, 1970) describe a pioneer study, conducted in Chile, for the production of a protein isolate containing 84 percent protein which is free of toxic compounds, and of a feedstuff residue containing 25 percent protein with a 10-fold decrease of goitrogens.

Present work in this area in our laboratories involves the use of rapeseed flour prepared from hull-free seeds as the starting material for the preparation of protein isolates. Jones (1970) is studying the effect of aqueous acetone extraction, reducing agents and -SH blocking agents on characteristics and yield of protein isolates. Results so far indicate it would be possible to recover up to 43 percent of the crude protein in the starting material in the form of isolates.

Another area we plan to be active in at the Food Research Institute is a study of the functional properties of rapeseed protein. It has been proven over the years that we do not eat or drink the different items we consume because of their nutritional value. The classical example is, of course, alcoholic beverages. I have yet to meet the person who normally drinks whiskey because it is a source of readily available energy. Likewise, nobody is going to use rapeseed protein because of its high PER or NPU. We are going to use it in our foods if it is proven to have suitable functional properties. A functional property of any food component or ingredient may be defined as, "The normal or characteristic property of that component or ingredient which allows it to perform a special task when used in the preparation of a food product or when naturally present

in a food raw material." So we use gelatin, for example, in several food preparations because it is capable of forming gels with relatively very large amounts of water. We use soy protein concentrates in comminuted meat products because of their juice holding capacity. We use corn syrup in candies because it controls granulation, and so on.

Food proteins are usually denatured upon processing into food products. The degree to which the amino acid residues are exposed on denaturation under various processing conditions is dependent upon the type of bond stabilizing the native, 3-dimensional molecular structure. Quinn (1970), therefore, intends to use protein bond-specific reagents in the extraction and solubilization of proteins from rapeseed; and in the determination of the types of intermolecular interactions in solution. The proteins will be divided into classes according to reactivity towards the bond-specific denaturants. The classes will then be compared as to functional properties such as water-binding, fat emulsifying, gelling, thickening, whipping, heat coagulability and so on. We hope this work will give us information on the natural functional characteristics of rapeseed protein and, if necessary, the means to modify them.

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