

A STUDY OF LIPIDS OF OAT VARIETIES

by

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**THIS BOOK
CONTAINS
NUMEROUS PAGES
WITH DIAGRAMS
THAT ARE CROOKED
COMPARED TO THE
REST OF THE
INFORMATION ON
THE PAGE.**

**THIS IS AS
RECEIVED FROM
CUSTOMER.**

INTRODUCTION

Recorded about 4000 years ago, developed as a domestic crop between 1500-500 B.C., oats were extensively established in western Europe at the beginning of the 17th century. In 1602, oat seeds are said to have been carried across the Atlantic Ocean to be planted first in North America (10, 13, 61).

Oat milling is a major industry for the production of breakfast cereals. Rolled oats and oatmeal are high in good quality protein and linoleic acid rich fat (86, 2). They also are the richest source of calcium, phosphorus, iron, and thiamine among cereal foods (10, 13, 49, 53). Their low sodium content makes them eventually useful as a nutrient for sodium-poor diets (5). These high nutritive attributes are partially due to the fact that they are made out of oat groats (shelled oats). The bulk of the bran, the aleurone layer, and the germ, which are rich in proteins, vitamins, and minerals, remain with the portion used as human food (13, 53).

The technique of air classification of cereal flours has been receiving particular attention during the last two decades in the hope that new unique and more useful products can be obtained from cereals. A wide variety of flour fractions of considerable value has been obtained from hard and soft wheat flours and from winter and spring wheats, and constituted a field of intensive investigation (26, 54, 55, 69,

77, 82, 85, 87, 91 among many others). Air classification also was performed on flours from corn, sorghum, rice, and defatted soybeans (85). No work has yet been reported concerning the response of oats to air fractionation.

This study deals with the determination of free and bound lipids of four oat varieties, of their groats and their air-classified fractions, and with the characterization of these lipids by thin-layer adsorption chromatography. Furthermore, it was an attempt to examine the behavior of oat flours upon air classification by following the eventual shifts occurring in the gross composition of the air-classified fractions as compared to the original flours.

REVIEW OF LITERATURE

Botanical Classification of Oats

Brownlee considered the treatise "L'avoine" by Denaiffe and Sirodot from 1902 as the most comprehensive monograph on oats (10). The botanical classification of the oat plant was summarized by Stanton in 1936.

As outlined by Matz (61), the oat plant is classified as follows:

Phylum:	Spermatophyta
Class:	Angiospermae
Subclass:	Monocotyledones
Order:	Graminales
Family:	Poaceae (Graminaceae)
Tribe:	Avenae
Genus:	Avena

Most of the oats produced today belong to the species *Avena sativa*. The red oat *A. byzantina* is grown in regions

considered too warm for satisfactory growth of the common oat.

According to Stanton's description, the oat roots are small, numerous, and fibrous and penetrate the soil to a depth of several feet. The oat plant produces from 3 to 5 hollow stems varying from 1/0 to 1/4 mm. in diameter and from 2 to 5 ft. in height. The leaves average about 10 mm. in length and 5/8 mm. in width. The panicles or heads are either tree-like or banner-like. The grain is produced on small branches, in spikelets varying in number from 20 to 150 per panicle. Each spikelet contains 2 or 3 florets or grains. The spikelet is loosely enclosed within the outer glumes (chaff). The kernels, except in the naked oat, are tightly enclosed in the lemmas or inner glumes and palea. The lemma or hull ranges in color from white, yellow, grey, and red to black. The kernel (caryopsis), without its adhering glumes, is very slender and constitutes 65 to 75% of the total weight of the whole grain.

Several hundred varieties of oats may be differentiated on the basis of botanical characteristics.

Culture and Distribution of Oats

Oats are best suited to cool, moist climates, but the diversity of varietal types makes successful production possible over a range of climatic conditions wider than for any other cereal (61).

Oats are the less selective grain among other cereals as far as soil is concerned. Ogrotechnical aspects of oat culture were discussed by Findley (31) and by Coffman (15).

Nitrogen is often the limiting nutrient in oat culture, mainly on the less fertile soils. Supplies of phosphates, potassium, and calcium are needed for oats as for other crops. Oats apparently are able to thrive at lower levels of these nutrients than are corn, wheat, or other cereals. They are subject to defective growth on manganese-lacking soils. Iron, copper, zinc, boron, and molybdenum also are essential minor nutrients for oats. Oats seem to require more moisture to produce a given amount of dry matter than any other cereal except rice (61). Pollination, fertilization, and embryology of oats were described by Bonnett (9). Hygroscopic moisture of this grain exposed to atmospheres of different relative humidity was studied by Coleman and Fellows (16). Specific heat studies on oats were made by Haswell (34).

A broad survey of world oat production is given by D. W. Kent-Jones (50). The chief oats producing countries are the United States, Canada, U.S.S.R., Germany, England, Finland, and France in the northern hemisphere and Argentina, New Zealand, and Australia in the southern hemisphere.

In the United States, the largest area of oat production lies north of the line where average June temperatures are below 65° F. (15). The southern limit of effective culture seems to be around the 30° of latitude in the northern hemisphere (61).

About 90% of the oat crop is used as feed. It ranks third as the most important grain crop in the United States, being exceeded only by corn and wheat (13). In terms of bushels, the average oat crop is about as large as the average wheat crop; however, a bushel of oats weighs only around 32 pounds while a bushel of wheat weighs 60 pounds.

The per capita consumption of oats in the United States occupies the fourth place after rice, corn, and wheat.

Quality Factors and Grading System of Oats

According to the Official Grain Standards, grain offered for sale as "oats" must have at least 75% of *A. sativa*, *A. hyzantina*, or a mixture of these species. Oats are divided into the five classes of white, gray, red, black, and mixed oats. White oats comprise the bulk of the oats grown in the United States and are the preferred type for milling (13).

Each class is further subdivided into grades which are based on the characteristics summarized by Matz (61). Some of the factors affecting the quality of the oat kernel, as summarized by Carroll (13), are: 1) condition of grain, 2) moisture content, 3) extraneous material, 4) percentage of hull, and 5) size of kernel. Milling oats now average 37-38 pounds test weight.

Structure and Gross Composition of Oats

As pointed out by Brownlee (10), the first drawing of

the longitudinal section of the oat kernel was made by A. L. Winton in 1932. The hystology of the oat grain was described by Bonnett (9).

The oat grain is comprised of the caryopsis or kernel called "groat" (75% ave. content) and the enclosing hull or husk formed from the lemma and palea (53, 61). The gross physical structure of the oat groat is similar to that of wheat and barley kernels; however, it is covered by numerous hairlike protuberances. The three major components of the groat are the bran, endosperm, and germ. Starting from the outside of the groat, the bran layer consists of the epidermis (fruit coat or pericarp), seed coat (spermoderm or testa), the hyaline layer (perisperm), and the alleurome layer.

The alleurome is a single layer of cells, and is morphologically a part of the endosperm; it surrounds the starchy endosperm only and ceases where the latter abuts on the scutellum. The endosperm is composed mainly of starch cells. The germ in groats is longer and narrower than the germ of wheat. Associated with the germ is the scutellum, a shield-shaped organ situated between the embryo (germ) and the endosperm; its function is considered to be the mobilizing the stored food reserves in the endosperm and transmitting them to the embryo when the grain germinates. The scutellum is the seat of most of the vitamin B contained in the grain. The germ and scutellum are richest in fat and protein. The epidermis contains the main part of lignin. Cellulose is found in the

hyaline layers, alleurone cell walls, endosperm cell walls, and germ cell walls (61). The proportions, as percentages by weight, in which the various parts occur in oat grains are (53): hull 25%, pericarp + testa + alleurone 9%, endosperm 63%, germ (embryo + scutellum) 1.2% and 1.6%, respectively.

The gross chemical composition of oats and oat products, according to data reported by Matz (61) from "Facts on Oats," released by the Quaker Oats Company is:

Component (dry weight basis)	Dry milling oats	Finished groats	Oat meal
Moisture, %	7.5	7.5	8.1
Crude protein, % (N x 6.25)	13.0	17.0	16.8
Crude fat, %	5.5	7.7	6.7
Crude fiber, %	11.8	1.6	3.9
Ash, %	3.7	2.0	2.2
Nitrogen free extract, %	66.0	71.6	70.5
Phosphorus, %	0.367	0.501	0.445
Iron, ppm.	51.0	47.0	51.0
Calcium, ppm.	700.0	570.0	530.0
Manganese, ppm.	34.0	34.0	48.0
Thiamine, mg./100 gm.	0.65	0.77	0.78
Riboflavin, mg./100 gm.	0.14	0.14	0.17
Niacin, mg./100 gm.	1.15	0.97	1.25
Pantothenic acid, mg./100 gm.	0.86	1.36	1.19
Folic acid, mg./100 gm.	0.034	0.057	0.06
Choline chloride, mg./100 gm.	107.00	120.00	120.00
Pyridoxine, mg./100 gm.	0.20	0.12	...

These single figures are of limited value since a wide range of figures for each chemical constituent is encountered when a series of samples is analyzed (53).

Lipids in Oats

Among the main attributes of the chemical composition of the oat kernel is its high fat content as compared to other

cereals. The very first published work on cereal oil studies was made on wheat by Topley in 1861 (4). König, in a study of the fat of various plants in 1874, obtained the following information on oat seed oil:

Oleic acid,	60.5%
Solid acids,	36.7%
Glycerine,	2.8%
M.p. of solid acids,	55-56°C.

In 1890, Stellwag, in a study of various feeds, also extracted oats with ether and benzene and found that ether-extracted fat contained a slimy substance. He performed determinations on both filtered and unfiltered lipid extracts and found a lecithin content of 0.76% on the former and 2.87% on the latter. Oat fat was further investigated by Moljavko-Wesotzki (1894), using petroleum ether as a solvent. Berry (1920) found that free fatty acids in oat oil were negligible, but hydrolysis occurred in proportion to the time between grinding and extraction. Paul, in 1921, extracted oats with petroleum ether and filtered the extract through kaolin and kieselguhr. On keeping the oil stoppered for three months, the acid value was little affected, but the odor was that of a rancid oil. Chemical and physical constants of the oils of oats, as given by the various investigators up to 1921, are summarized by Ball (4).

In 1927, Amberger, in Germany, published a study on the composition of the oat lipid glycerides (2). Amberger pointed out the characteristic distribution of fat in the oat kernel as compared to other cereals. While in other cereals the oil is mostly concentrated in the peripheral and in the germ

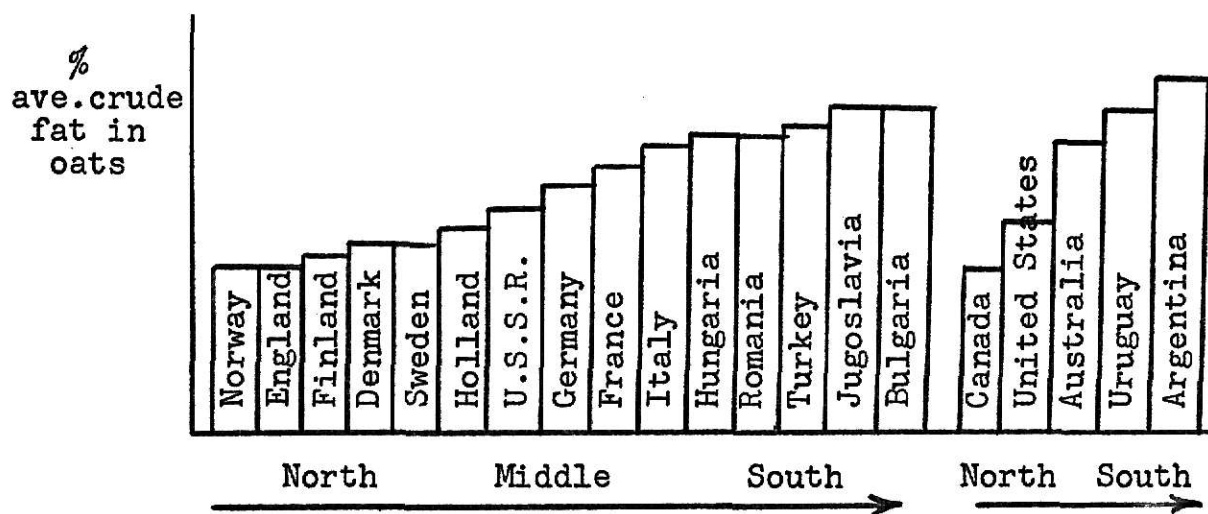
layers, the oat has its fat distributed throughout the whole kernel; the main part of it, however, is contained in the germ. Amberger emphasized the particularly high fat content of oats with respect to other cereals. His work revealed the peculiar quality of oat fat, namely, its high content in linoleic acid.

Hutchinson and Martin, in England (43), emphasized the varietal character of oat lipid content. Oat lipid content depends chiefly upon kernel content which is itself a varietal characteristic. The free fatty acid content, however, is mainly dependent upon the environment during cultivation, harvesting, and storage rather than on the variety (3-10% for sound kernels). After milling, the free fatty acid content rises to much higher levels within a few days.

In Prague (Czechoslovakia), a series of studies on oats and the chemistry of oat products has been published since 1959 (47) (70, 71, 72, 73). The composition of saturated and unsaturated fatty acids of different parts of the oat grain was investigated by a fractionated extraction with solvents of increasing polarity: hexane, followed by a mixture of ethanol and benzene. Extracts were dried, using anhydrous Na_2SO_4 , filtered, the solvent distilled off and the last residues of solvent removed in an oven at 105°C . Fractionation of solid fatty acids (lead salt separation) was performed by GLPC after previous esterification with methanol. Linoleic acid content was highest in the whole grain extract.

A fraction of 10% of stearic acid was found contrary to previous data. Small amounts of palmitoleic, lauric, and myristic acids also were detected. Changes in lipid chemistry of oat flakes were studied during heat treatment under different conditions of time and temperature. The lipid content decrease was found to be a linear function of reaction temperature. Lipids of oat hulls also were investigated. Furthermore, the varietal factor, as determining the lipid content of oats, was established in agreement with Hutchinson's findings.

The genetic variability and the environmental influence upon the oats' fat content were studied by Stuke in Germany (86). Crude fat content, on a broad range of varieties and samples over a three-year period, was determined refractometrically and found to be relatively stable over the years. Hence, it was concluded that the environmental factor had little impact on fat accumulation, and lipid content was mainly varietally determined. On the other hand, comparing the mean oats' fat content from different Eurasian and American countries, an increase of crude fat content was found with a trend from North towards South. Within variety, the highest variability was found in *avena sativa*. An increase of fat content with the number of chromosomes was observed. A highly significant negative correlation was found between 1000 kernel weight and crude fat content. Crude fat vs. crude protein, however, showed a poor correlation.



Lindberg et al. (59), in Sweden, studied the fatty acid composition of ripening oats. The main part of the fat synthesis occurs in a short and early period of ear formation. A decrease in C_{18} mono- and triunsaturated and an increase in C_{18} diunsaturated fatty acids was observed as ripening proceeded.

Aylward et al. (3) isolated phosphatidic acids and phosphatidyl inositol from oats, barley, and rye. The presence of a serine containing phospholipid also was ascertained which was considered a phosphatidyl serine without being identified. The fatty acid components of these phospholipids were identified by GLC. The yields suggested that phosphatidyl inositol and phosphatidic acids together comprise about 5% of the total phospholipids contained in oat lipids.

Novzhilova (66) found that oat lipids contain about 0.19-0.30% sterols and up to 75 mg. % tocopherols.

According to data recorded by the Quaker Oats Company, Matz (61) summarized the lipid composition of oats as follows:

Lipids (ether extract, dry basis)			Fatty acids	
Whole grain	%	5.4	Palmitic	% 10.0
Groats	%	7.6	Oleic	% 58.5
Hulls	%	0.62	α -linoleic	% 17.2
Germ	%	11.2	β -linoleic	% 13.9
Germ-free groats	%	5.8		
Germ plus bran coats	%	7.4-9.0		
Endosperm	%	6.2-6.7		

The amount of lipid extracted depends upon the solvent. Considering the diethylether extract as 100, the equivalent figures for fat extracted by other solvents are: Petroleum ether, 97; CCl_4 , 104; CHCl_3 , 110; Benzene, 113; Acetone, 113; Ethanol, 128. Oat oil contains about 45% polyunsaturated fats and is an excellent source of linoleic acid which is essential for human diet.

Indeed, De Groot (27) found striking blood cholesterol lowering effects of rolled oats fed to rats. When carried out with human volunteers, a 12% decrease in cholesterolemia was observed after a three-week diet (300 g. daily intake of an experimental bread containing 140 g. of rolled oats). Serum cholesterol rose to initial levels after two weeks of normal diet. This effect was attributed to the high content of polyunsaturated fatty acids in oats and was confirmed by a further experiment performed with defatted oats.

Proteins

The usual data presented as protein in oats are the

Kjeldahl determined nitrogen contents multiplied by 6.25. This factor is not universally accepted. When so calculated, the protein of a typical example of milling oats averages about 13.1%, of hulls about 4.5%, and of finished groats about 16.9% dry basis.

As summarized by Matz (61), the amino acid composition of oat proteins (expressed on a 16% N₂ dry matter basis) is: alanine 6.2, arginine 6.4, aspartic acid 3.7, cystine 1.8, glutamic acid 20.0, glycine 2.4, histidine 2.2, isoleucine 4.8, leucine 7.0, lysine 3.4, methionine 1.6, phenylalanine 5.0, proline 5.7, serine 4.5, threonine 3.3, tryptophan 1.3, tyrosine 3.8, and valine 5.8. As compared to wheat, lysine and arginine are considerably higher in oats.

The predominate factor determining the nitrogen level in oats seems to be, as in wheat, the level of available nitrogen in the soil in late spring and early summer prior to heading and during development of the grain (44). Ninety-five per cent of the total nitrogen of the oat grain is contained in the groat, where it varies from 1.4-3.9%. The husk contains only 3-10% of the total nitrogen in the grain. Compared to wheat, whole oats are slightly poorer while the groats are decidedly richer in protein content.

Hutchinson found a significant inverse correlation between nitrogen and fat content within variety (44).

Hischke et al. (39) described seven pure varieties of oats grown under similar environmental conditions (Grayslake,

Ill.). The protein content of their groats ranged from 18.1-22.2%.

Carroll (13) described a new high protein variety of oats called "*Avena sterilis*." This species grows wild in Israel. Its protein content was found to be higher than 25%. The highest protein oat grown commercially in the United States is the variety Garland, of 22.6% groats protein, developed at the University of Wisconsin. Most wild species of oats have a different number of chromosomes and cannot be readily crossed with United States cultivated oat types. "*Avena sterilis*" has the same number of chromosomes as the United States cultivated "*Avena sativa*" and can be readily crossed. Such a jump in protein would make oats an even better food for children and adults.

Csonka (20) emphasized that nitrogen content may serve as a guide in the selection of proper varieties, but should not be the final factor in judging protein quality. The low biological value of cereal protein is due mainly to its low lysine content (18). Osborne and Mendel, in 1914, showed that maintenance but not growth of animal is possible without lysine (62). According to Rose, the daily requirement of lysine is 0.8 g. This amount is already contained in the daily wheat bread consumed (18). According to Petitpierre A. Holet, the addition of lysine to wheat does not provide the optimum wheat quality.

There seems to be a poor lysine utilization of wheat

protein due to changes determined by heat throughout the baking process. During treating, reducing sugars or other aldehydes may react with functional nitrogen groups not protected by peptide linkages. The bond thus formed cannot be hydrolyzed by the animal enzymes (11). This phenomenon, known as the browning reaction, has a twofold effect: 1) lysine is most affected through its ϵ -amino group, and since it is frequently the limiting amino acid, the loss in protein value will be proportional to the amount of lysine lost; 2) even if lysine is not limiting, the blocking of the ϵ -amino group of lysine or the guanido group of arginine may interfere with overall protein digestibility. Trypsin splits a protein molecule at the peptide bond containing the carbonyl of either lysine or arginine, provided their ϵ -amino group is unsubstituted.

Since oats are relatively high in lysine, when used as a sole source of protein, they show the highest value in standard rat feeding tests when compared to wheat (18). The importance of oats as a nutrient is diminished only by its lack of gluten which makes it unsuitable for processing into bread (18, 48). On the other hand, it seems likely that wheat protein is damaged to a greater extent through baking it than are oat proteins when only cooked.

Cunningham et al. (21) compared the properties of wheat protein with analogous proteins of oats, barley, and rye, in order to get an insight into the nature of dough, and hence

to find a chemical explanation of its viscoelastic properties. Crude but analogous preparations of "gluten" were obtained from each of these cereals. The main differences between these various proteins were the cohesiveness and elasticity of the wheat, barley, and rye "glutens" as compared to the claylike oat "gluten." Total nitrogen content was highest in wheat gluten, followed by oat protein. Apparently, variations in physical characteristics of glutens cannot be attributed to variations in total nitrogen. The amide nitrogen content was highest in the wheat gluten, lowest in oat "gluten," and intermediate in barley and rye "glutens." These findings were interesting in view of the hypothesis of polymer cross-linkages occurring through the interaction of amide groups. Precipitation of proteins from these cereals with salt was described in a further study by the same authors (22).

Frey (32) emphasized the poor nutritive quality of the alcohol soluble proteins of corn, wheat, and oats because of deficiency in lysine and tryptophane. Csonka pointed out that about 16% of the oat protein is soluble in 70% ethanol (20); this ratio appears to be constant in oats in both the high and the low nitrogen-containing species, while in corn the alcohol soluble fraction (zein) increases, leading to a poorer protein quality (32). Thus, the total protein of high protein corn is less nutritionally balanced than in low protein corn. This ratio increases also with the advancing maturity of corn and with the accumulation of nitrogen in the grain. In oats,

this alcohol soluble N/total N ratio decreases during the initial stages of seed development (92) and remains rather steady regardless of high or low total nitrogen level of the variety. Only environmental variations associated with season and planting area tend to change the proportions of certain amino acids in the oat protein with the variation of total nitrogen content (33). McElroy, in 1919, found that the lysine content of nine samples of oats ranging in protein content from 9.4 to 18.9% remained constant (39). Weber et al. (39) reported a significant difference in growth response when various oat varieties were fed at isonitrogenous levels to rats; he established a lack of correlation between lysine content and growth rate.

Hischke (39), studying seven pure oat varieties, concluded that the lysine and glycine level decreased with respect to total protein content with protein level increase, thus not confirming the statements by Frey. Glutamic Acid, according to Hischke, was directly correlated to sample protein content; no significant results were detected from his feeding experiments.

Evans (30) studied the nutritive value of oat proteins through feeding experiments on rats.

Burrill and Schuck (12) studied the adequacy of the intact oat protein as compared with the amino-acid equivalent on humans.

Kuether (57) found rolled oats well utilized by human

subjects, while wheat flakes were inferior to unprocessed whole wheat.

Smirnova (80) made an evaluation of oat flour according to amino-acid composition and digestibility of proteins. She found the digestibility and essential amino acid content of oat flour twice as high as in barley flour.

Carbohydrates

As pointed out by Matz (61), compared to other cereals, oats are characterized by a low carbohydrate content.

Whole oats yield about 65% N-free extract on a dry basis. Starch and other carbohydrates constitute about 90% of this material. Reducing sugars in the extract are less than 0.1%, while total sugars are often near 1.4%.

Whole oats contain about 14% pentosans, mainly araban and xylan. The highest concentration of pentosans is in the hulls. Therefore, oat hulls are an important raw material for the manufacture of furfural.

The fiber of oats, found mainly in the hulls, consists basically of cellulose, hemicellulose, and lignin.

Krznarich (56) found that the hemicelluloses from oat hulls were composed of polyoses and polyuromide material.

MacMasters (61) found that the amount of starch extractable from whole oats ranges between 33 and 43%. The starch granules are very irregular in shape, assuming mainly a polyhedral configuration. Furthermore, ovoid and hemispherical

particles, and spindle-shaped particles, characteristic of oats, are occasionally seen.

The average size of starch particles varies according to the location within the kernel. The largest and most loosely packed granules are found in the cheek near the crease, while smaller particles are found in the horny endosperm.

Oats contain the smallest starch particles of all cereal grains; their size ranges from 3μ to 10μ . The individual granules develop in clusters of about 60μ in diameter, which fill most of the central space within the endosperm cells. These compound clusters are usually broken up during processing of the kernel, yielding the irregularly shaped granules occurring in most oat starch preparations.

Minerals

The principal mineral content of oat products is summarized by Joslyn (unpublished data from the Quaker Oats Company) on dry matter basis (49):

	Whole oats	Groats	Rolled oats
Calcium, %	0.043-0.117	0.052-0.065	0.049-0.055
Phosphorus, %	0.340-0.490	0.510-0.550	0.460-0.500
Iron, ppm.	42-84	49-54	43-48
Manganese, ppm.	20-96	38-46	45

Morgan (65) studied the content of major elements like Ca, Mg, Na, K, P, and Cl and trace elements like Cu, Co, Mn, and Zn in oat and barley grain grown in Wales for three years.

Variations in their contents were discussed in relation to current dietary recommendations for farm livestock.

Greaves and Hirst found a 70% variation in total phosphorus content in oats grown in different localities. Inorganic phosphorus varied from 5.5 to 12% of the total phosphorus content in oats grown in Utah (35).

Bergner and Wagner found the following mineral content in German oats and their products (5):

	Oats	Groats	Rolled oats (oat flakes)
Boron, g./100 g. d.m.	75	73-79	72-82
Fluorine " "	63	50	48
Iodine " "	12	6	5
Sodium, mg./100 g. d.m.	9	7	7
Potassium " "	396	335	337

They also studied the distribution of minerals through various component parts of the oat kernel.

Generally, the mineral content varied with the origin of oats and depended upon soil treatment, climate, degree of maturity, and variety. They emphasized the high iron content in the oats. Their fluorine content corresponds to that of fruits and vegetables; however, it is too low to constitute a supply source (daily requirement, 1.5 mg.). Some silica was detected in the oat kernel composition.

Vitamins and Antioxidants

Kent summarized the vitamin contents in oats as follows (53):

Vitamin B ₁ (thiamin)	5.7 mg./g.
Vitamin B ₂ (riboflavin)	1.3 "
Niacin (nicotine acid)	0.4 "
Panthothenic acid	9.0 "
Pyridoxin (vitamin B ₆)	1.2 "
Folic acid	0.2 "
Choline	980.0 "
Vitamin E (total tocopherol)	1.7-3.2 mg./100 g. (D.M.B.)

Pulkki and Puntula (76) found the distribution of thiamine in hulled oats rather different as compared to wheat and rice. The outer surface layers of the oat kernel (pericarp) contain much more thiamine than the whole kernel; its amount is 10-15 times higher than in the corresponding material from wheat and rye. Thiamine, however, seems to be more evenly distributed in germ and endosperm than in other cereals. Thus, products made from oat endosperm would be comparatively good sources of thiamine even though the vitamin rich outer surfaces are removed by polishing.

Hunt et al. (42) studied the environmental factors influencing the thiamine, riboflavin, niacin, and panthothenic acid content of wheat, corn, and oats.

The total tocopherol level in oats is comparable to that in wheat (53). Novzhilova found a range of 9.8-75 mg. tocopherol in 100 g. oat fat (66).

The use of antioxidant properties of oats as an additive to clear wheat flour for the improvement of its storage properties was described by Dr. Shellenberger (81). Daniels et al. isolated an antioxidant from oats in 1961 (23). Thin layer chromatography of this material revealed at least six components

which were partially separated by column chromatography. Alkaline hydrolysis of the mixture of these compounds gave 29% caffeic acid, 10% ferulic acid, 4.5% glycerol, and 50% long-chain hydroxy fatty acids. Two dimensional TLC detected 24 phenolic antioxidants (24). Characterization and eventual structures of these compounds are discussed by Daniels and Martin in their latest publications (25).

Processing and Enzymes in Oats

Oats Processing. The difference between oat and wheat processing is the consequence of the different structure and composition of the two cereal grains (53).

1. The adherent husk of the oat grain is tough and fibrous; it must be removed in a special dehulling process, during the manufacturing of edible products. The chaff of wheat, which corresponds morphologically with the husk of oats, comes away easily during threshing.

2. The fat content of the oat kernel is 2-5 times as high as that of wheat, and the oat kernel contains an active lipase.

3. The bran of the oat groat is relatively thin and pale as compared to that of the wheat grain, hence oatmeal and rolled oats are manufactured from the entire groat and the bran is not separated from the endosperm. Removal of the groat pericarp is necessary in infant food manufacturing.

4. Since oat flour cannot be used for bread making, the

processing of oats is designed for products suitable for porridge making, oatcake baking, ready-to-eat breakfast cereals, and infant foods.

Oatmeal Milling. The milling of oats is primarily an operation in which the hulls are separated from the groats, and the groats milled into the end products of oatmeal, rolled oats, or oat flour (13). The entire groat is used normally for the manufacturing of the final product whether it be oatmeal, rolled oats, or oat flour. The physical appearance of the oat products may be different; however, the chemical composition will be essentially the same.

a. Cleaning. Oats as received from the elevator are designated as green oats. Milling oats are obtained by removing all undesirable materials through a series of separations and gradings according to length, width, and density.

b. Drying is the next operation. Nowadays, pan dryers are in use. They are normally 10-12 feet in diameter and placed one above the other in stacks of 7 to 14. Each pan is steam-packeted and open on the top. Oats enter at the center of the top pan and are slowly moved to its edge by sweeping arms in order to prevent burning. The oats drop from the top pan to the center of the pan below where the process is repeated; the drying cycle is about one hour long and the temperature seldom exceeds 90° C. The purpose of drying is to loosen the hull from the groat, to impart a toasted flavor, and to inactivate the lipolytic enzymes of the oats. After drying,

the oats are colled and then are ready for processing into milled oat products.

c. Hulling is actually the first stage of the oat milling process. Today, the impact huller is used most frequently. The oats enter the center of a high-speed rotor with fins which throw the oats against a rubber liner fixed to the housing of the machine. The liner reduces the breakage during impact and also assists in efficient separation of the hull from the groat. The discharge from the huller consists of groats, free hulls, meal and fines which are separated from the groats by aspiration.

d. Steaming, cutting, and flaking. The groats may be steamed as whole groats or cut into smaller pieces (2-4 per groat), depending on the end product to be manufactured. Old-fashioned rolled oats are made by steaming and flaking the whole groat.

Quick-cooking rolled oats are made by steaming and flaking the cut groats.

Steaming usually is done using live steam at atmospheric pressure. The steaming softens the groats and permits flaking with a minimum amount of breakage. It also reduces any enzyme activity which may have survived the drying step.

e. Flour production. Oat flour may be made by grinding either the groats (steamed previously and dried) or rolled oats on impact-type grinders. For some uses, oat flours with a lighter color are required. This may be accomplished by

cutting the groats and removing some of the higher protein fractions by aspiration.

Enzymes and Processing of Oats. The enzyme lipase present in raw oats is located almost entirely in the pericarp of the groat (43, 53). Lipase hydrolyzes fats into glycerol and f.f.a. Oat fat is distributed throughout the endosperm, germ, and aleurone layer; the last two being particularly rich sources. There is little, if any, fat in the pericarp.

In the intact kernel, the lipase and fat thus do not come into contact with each other, being located in different parts of the kernel. Sound oat kernels normally contain up to 10% of the fat in the form of f.f.a.

In the process of oat milling, the lipase from the pericarp is brought into contact with the oat fat in the endosperm, germ, and aleurone, and the process of f.f.a. release is enhanced to an extent dependent upon moisture content, temperature, and fineness of grinding. Lipase activity is low at the low moisture content and low temperature, but is appreciable at 10% or higher moisture content and is optimal at 36° C. (in wheat) (94).

Thermic deactivation of oat lipases was studied by Jan Pokorny et al. in Prague, Czechoslovakia, who found that complete inactivation of the enzyme occurs only at a temperature of at least 120° C. in dry oats. Addition of water diminishes the resistance of the enzyme (71).

Kent (51) emphasized the importance of a pre-kilming

process of steaming the raw oats, called process of stabilization. Lipolysis of different commercial fats by oat flour was studied by Widhe and Onselius in Sweden (94). Heiss (38) emphasized the importance of steaming for the prevention of bitter and astringent off-flavors of oat products which may occur during storage. He considered the peroxidase activity as a criterion for the control of the steaming process on account of the high temperature stability of this enzyme. He developed a routine test for the determination of peroxidase activity and included the determination of lipase activity only as an additional information.

Kent (51), however, emphasized the importance of not overdoing the steam treatment. The meal from heavily steamed oats tends to be abnormally granular and deficient in the fine floury fraction. He suggested the tetrazolium test (51, 53) for determining the limit of steaming when lipase inactivation occurred.

The use of palm kernel and coconut oil should be avoided in oatcake baking (51, 53) since the fatty acids in these oils are chiefly lauric and myristic; the former when released by lipase has a soapy taste.

The raising agent used in oatcake baking often is NaHCO_3 . The presence of free fatty acids in the oatmeal would lead to the formation of their sodium salts (soaps) and hence to a soapy flavor. Measures to avoid lipolytic activity at the Quaker Oats Company are mentioned by Carrol and Richards (13).

Usage of Oats and Their Products

Oat flour consumption has been increasing during the last years mainly for manufacturing of baby food and ready-to-eat breakfast cereals. Oat flours are used to some extent in baking mixes. Oatmeal cookies usually are made with rolled oats; in some instances, a mixture of oat flakes and oat flour is used. Steel cut oatmeal also is used in cookies to produce a nut-like texture and taste.

In some South American countries, oat flour is used as a base ingredient in beverage mixes.

Addition of small amounts of malted oat flour to wheat for improvement of diastatic and storage qualities was described by Dr. Shellenberger (81). Oat flour also was found to be effective in preventing rancidity in frozen sausage (13).

The main byproducts of oatmeal milling are oat husk, oat dust, and meal seeds, which make about 70, 20, and 10%, respectively of the total milling by-products (53).

Oat husk consists mainly of true (botanical) husk with an admixture of up to 10% kernel material.

Oat dust is rich in kernel material, particularly bran, and contains about 16% groat hairs and about 11% husk fragments.

Meal seeds consist of fragments of husk and kernel in about equal proportions.

Oat dust and meal seeds are of rather good feeding value

and are used for animal feeding. Meal seeds also are used for a peculiar Scottish dish (a fermented extract).

Before 1939, oat husk was used as fuel for burning in oat kilns in England. During World War II, it was used for oat husk meal.

When mixed with other by-products of oatmeal milling, oat mill feed of low nutritive value for ruminants is obtained.

Oat husk is used industrially as a raw material for furfural production; as a filter-aid in breweries in admixture with ground malt and water; as a diluent or filler in linoleum; and as an abrasive in air-blasting for removing oil and products of corrosion from machined metal components (13, 53).

Air Classification

Cereal flours obtained by conventional milling contain

- 1) fragments of/or whole endosperm cells or larger particles made up of two endosperm cells. These are the largest particles containing starch granules, water soluble carbohydrates, protein matter forming a matrix around the starch granules, and some lipids disposed in this protein matter or closely surrounding the starch surfaces.
- 2) Further, discrete starch granules varying in size, loosened from the protein matrix or still carrying small remnants of the protein matrix, constitute a smaller particle size category.
- 3) A large number of discrete very small particles, varying greatly in shape, which are pure protein, constitute the fine fraction of flours (77, 40, 26, 36, 55).

While in the stacked sieve procedure, the particle size is the main determining factor; fractionation by air classification makes simultaneous use of size, shape, and density (44, 77).

In the conventional milling process, the parent flour is sifted starting with the largest sieve opening, the finest fractions passing through sieve openings of decreasing size. In air classification, the finest particles are removed first from the parent stock and the coarsest last, advantage being taken of the ease with which the air current blows the light particles aside (82).

If a stream of conventionally milled flour is allowed to fall on a spinning disk, all the flour will be thrown off the disk in about the same direction. If a stream of air is directed at right angles to the path of the flour coming off the disk, the lighter and smaller particles will be blown in a different direction. Hence, a flour particle introduced through the top of the separation chamber of an air classifier is subjected to two opposing forces: a) a centrifugal force proportional to the cube of its size and b) the air drag (centripetal) force proportional to the square of its size. For particles larger than the equilibrium between these forces or larger than the "cut" size determined by the conditions of operation, the centrifugal force is larger than drag force. Hence the coarse particles accumulate in the peripheral region of the chamber. For smaller particles, the air drag (the

centripetal force) is larger than the centrifugal force and hence the fine fraction travels in the air stream towards an exit at the center of the chamber, being collected in a cloth filter.

Each "cut" produces a "fine" and "coarse" fraction. The coarse fraction is further classified by readjusting the system towards increasing "cuts" by means of adjustable vanes and by changing the angle of the air stream. This operation corresponds to the variation of the sieve wire number.

The very fine fractions usually contain high concentrations of small protein particles. The intermediary fractions are of a rather starchy nature and the residual "coarse" fraction usually corresponds to the original flour.

By varying the speed of the disk and the velocity of the air, it is possible to control the size of particles separated by the air stream. Furthermore, this technique permits the manufacture of more homogenous flours from varying cereal varieties, the increase of protein content in certain flour fractions and its decrease from other ones, and the production of specially "tailor-made" flours for specific uses.

A wide variety of flour fractions was obtained from hard and soft wheats, from winter and spring wheats, and from corn, sorghum, rice, and defatted soybeans (69, 85, 82).

Most studies have been concerned with wheat flours, their protein and mineral matter distribution, and baking qualities (36, 40, 41, 55, 69, 77, 82, 85, 87, 91 and many others).

Data on lipids in air-classified flours are fragmentary. Houston (41) reported preliminary results of studies on simple and compound lipids in air-classified flours. Gracza (1965) studied the petroleum ether extract and Daftary et al. (26) determined total, free, and bound lipid content of air-classified flours.

MATERIALS AND METHODS

Four pure varieties of oats (*Avena sativa*) from the 1968 crop were studied. The Garry, Jaycee, and Clintford varieties were white spring oats grown on a farm near Grayslake, Illinois and obtained from the John Stuart Research Laboratories of the Quaker Oats Company (Barrington, Illinois). Each of these varieties was dehulled and aspirated at Barrington; however, they still contained substantial amounts of unhulled oats. The Andrew variety (gray oats) was obtained from Kansas State University and grown on the Agronomy Farm in north Manhattan, Kansas.

All samples were stored in the cold room prior to milling. The flours also were kept in the cold room to prevent enzymatic lipolysis and other deleterious effects which may occur at room temperature.

Milling of Oats

Milling of oat samples was made in the experimental facilities of the Milling Department at Kansas State University.

Separation of oats from groats was done on an indentation separator.

Hulling of oats was performed on a Forsberg dry impact huller according to the flow given on Fig. 1. After the first dehulling step (3000 rot./min.), the material was passed through a Kice aspirator where the hulls were removed from the system by aspiration and the unshelled oats recycled for hulling. After the second hulling step and aspiration, the material was introduced into a Brabender Lab-O-Fix Separator which removed the good groats from the system, directed the cracked groats towards another collector, and separated the still unshelled oats to be recycled for hulling. The hulling procedure thus was repeated four times in order to achieve the complete shelling of the whole batch.

Milling of groats into flour was made on a Ross Experimental Mill, according to the flow shown on Fig. 2.

The groats were introduced into a 14/14 roller system and hence passed onto a sieve system consisting of three stacked sieves: the upper screen - a 20 w (20 wires/linear inch), the middle screen - 32 w (32 wires/linear inch), and the lowest screen - 84 w (84 wires/linear inch). The coarse (remainder) from the 20 s (small) was passed to a 22/20 system, wherefrom the same sieve separation was repeated. The coarse of the 20 w screen passed hence to a 23/28 roller system to be again separated by the same sieve system. The 20 w coarse material was then directed to a duster unit where

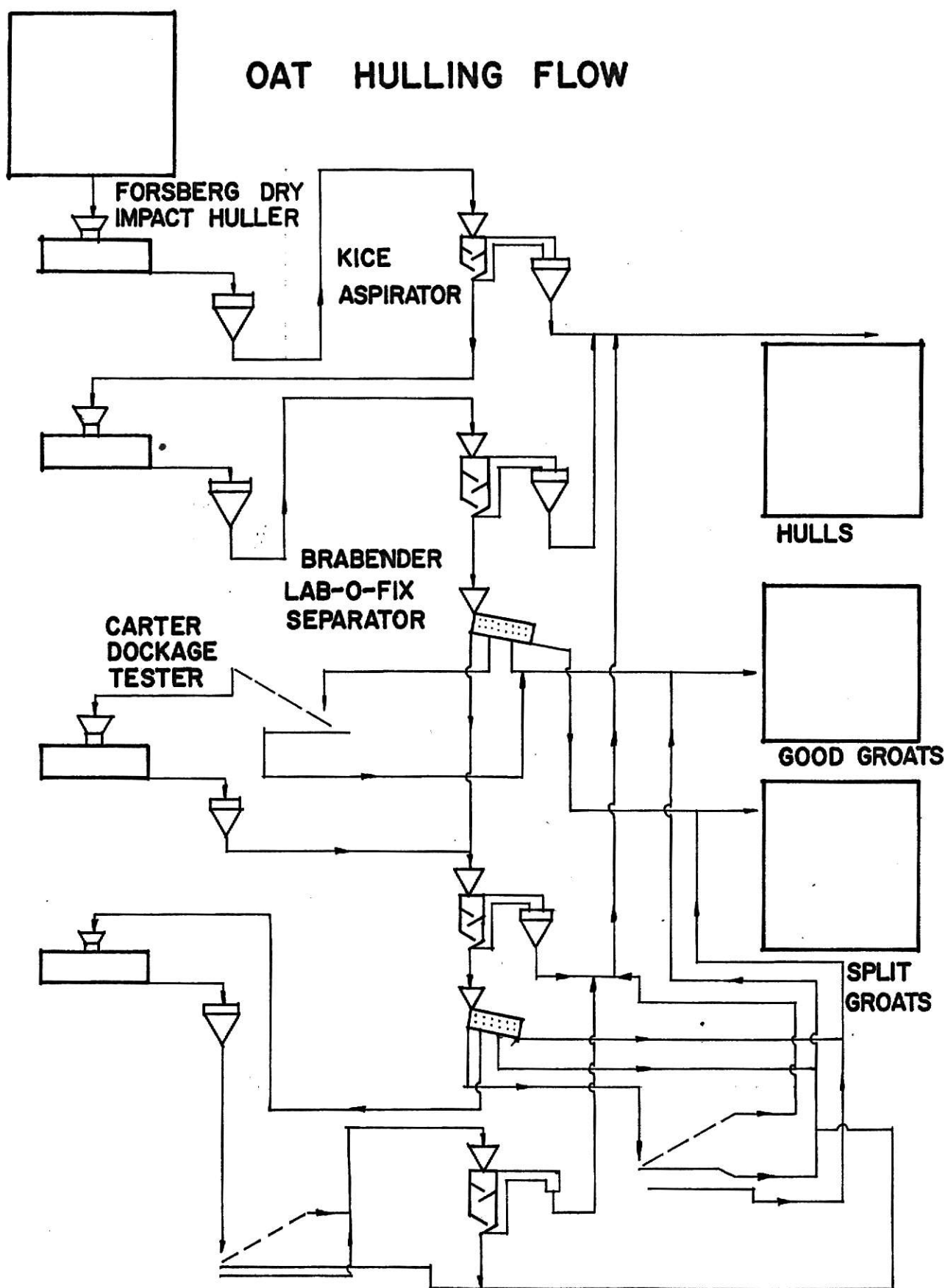
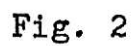


Fig. 1

34



separation into bran on one side and fines on the other side occurred. The fines were passed through an 84 w screen. The 84 w coarse fraction was passed to a 24/26 roller and hence separated into flour (84 w troughs) and shorts (coarse) by means of a duster.

The troughs of the 20 w screen were passed onto a 32 w screen, therefrom to the 84 w screen. The 32 w coarse was collected to follow a common path towards the 24/26 roller unit, whereupon they joined the 84 w remainder.

The 84 w troughs constitute the flour obtained by this milling procedure. A rather uniform extraction of a 65% yield was obtained for all four varieties described in this study.

Air classification of the 65% extraction flour obtained from the roller mill was performed on a Pillsbury Laboratory Model Turbo air classifier, according to the flow shown on Fig. 3. The operating "cut size" was varied by changing the feeding rate, by varying the angle of the air stream by means of adjustable louver curtains (vanes) through which the air passes on entering the separating chamber, by varying classifier rotation speed, and by adjusting the effective depth of the chamber, using decks. Each stage or "cut" produced a fine fraction which was removed and a coarse fraction which was further classified by readjusting the classifier.

The original flour was labeled as A flour. The first and second fine fractions (B and C) were obtained by using the same 100 lbs./hr. feeding rate, 5860 RPM speed, and 10' louver

AIR SEPARATION FLOW

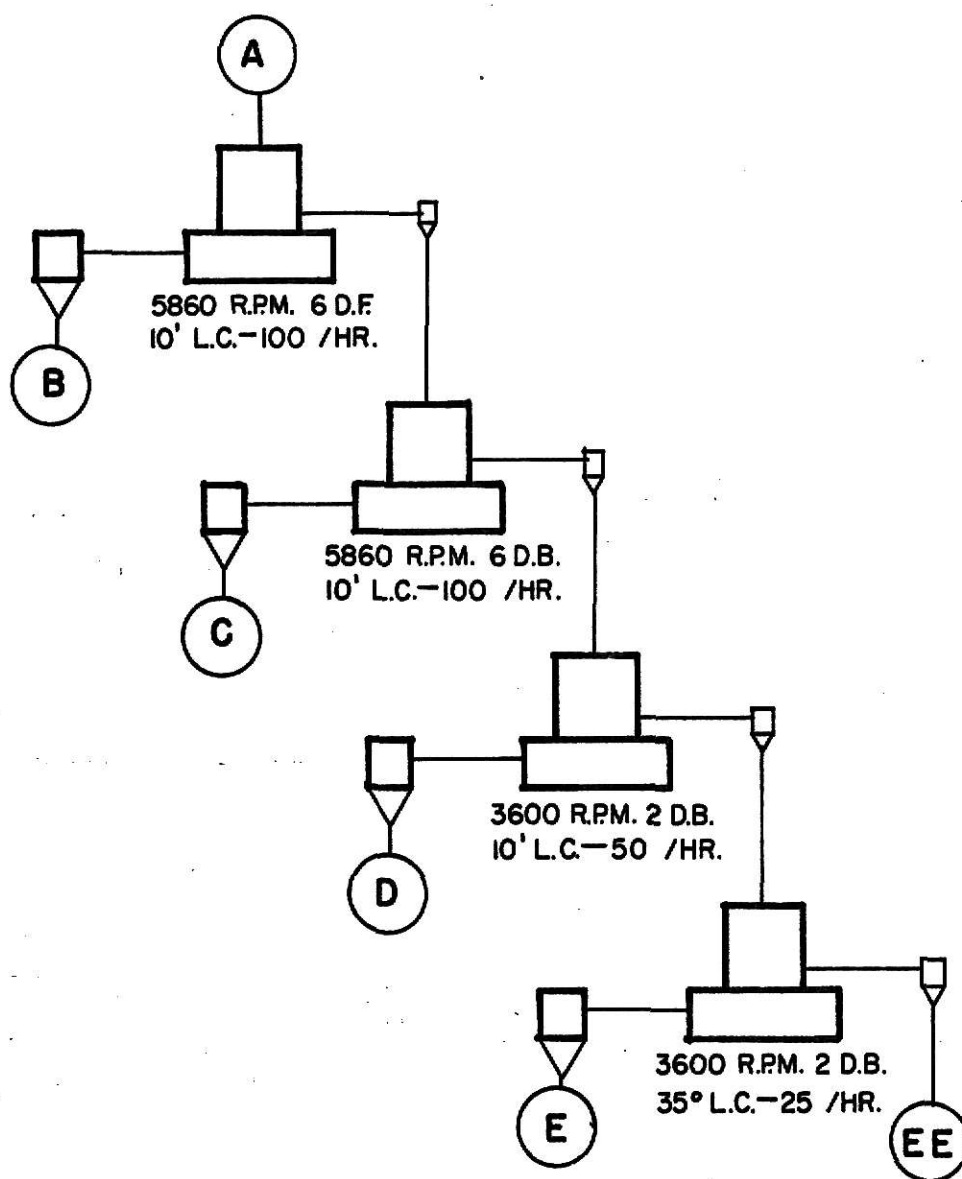


Fig. 3

curtain inclination. The intermediary fractions D and E were obtained by using a 3600 RPM speed, a 10' louver curtain inclination, and 50 lbs./hr. feeding rate for the former and a 35° louver curtain inclination with a 25 lb./hr. feeding rate, maintaining the same rotation speed for the latter. The coarse fraction was labeled EE. The same procedure was followed for all four varieties. Hence two fine, two intermediary, and one coarse fraction were obtained for each variety.

Analytical Determinations

The Fisher Average Particle Size. This parameter of the original and the air-classified flour fractions was determined on a Fisher sub-sieve sizer which employs the principle of permeability of porous beds. The procedure outlined by Croteau (19) was followed.

The Agtron Color. The color of the same samples was ^{as outlined} determined by Gillis (34). The "wet" Agtron was determined as described by Patton and Dishaw (68) in order to eliminate variations due to differences in degree of granularity and uneven distribution of the sample particles.

Moisture, Ash, Crude Fiber, and Kjeldahl Protein. These determinations were made on the oat and groat samples ground on a micro-Willey mill against a 40-mesh screen, on as milled, and on air-classified fractions, according to the AOAC (1966) methods. Carbohydrates (nitrogen free extract) were determined

by difference.

Proteins also were determined by a dye-binding method, according to Udy (88, 67), following the prescriptions and using the equipment furnished by the UDY Analyzer Company, Boulder, Colorado. The principle of this method is based on the reaction given by the basic groups contained in the protein molecules with the dissociated sulfonic acid groups of the dye orange G, at low pH, leading to the formation of an insoluble dye-protein complex.

"Orange" **G** is the coupling product of diazo benzene and 2-naphtol-6,8-disulfonic acid. The method also is based on the assumption that the proteins of a given cereal are distributed in a regular manner among varieties. The method consists of thoroughly mixing a 0.72-g. sample of oat flour with a certain amount of dye solution. A quantity of dye proportional to oat protein present in the sample becomes bound by the protein. Following removal of oat material by filtration, the amount of unreacted dye is determined by reading its increased transmission (extinction); this, in turn, is calibrated in terms of protein

$$\text{Crude protein content} = f(\text{transmission})$$

The oats and groats samples were ground on a UDY Analyzer Mill. Kjeldahl protein determinations were performed on these samples simultaneously. The air-classified and "as milled" flours were used as they were. The dye reagent, furnished by

the UDY Analyzer Company, was prepared according to the manufacturer's prescriptions. Conversion of reading results into protein content was made according to the conversion table for oats furnished by the UDY analyzer. Determinations were made in triplicate. The results in crude protein were then compared with the data obtained by the Kjeldahl method.

Lipids. Lipids were determined by a fractionated extraction with solvents of increasing polarity.

Free Lipids. Free lipids were determined on all aforementioned samples by extraction with petroleum ether Skelly F (b.p. 35-60° C.) in a Goldfish extractor (12-14 hrs. on 5 g. samples). The extracts were not clear; the Jaycee oats gave a rather turbid extract; and the other varieties' extracts were less turbid. Literature described this occurrence for oats quite long ago (4, 70, 71). Filtration would have led to loss of some compounds. Quantitative comparison between cleared and uncleared extracts showed no difference. Determinations were sometimes repeated 6-8 times, showing rather satisfactory reproducibility. Since the extracted lipids were to be subjected to TLC, no clearing was made on extracts in order to avoid losses or artifacts of compounds. The lipid residue was stored at 4° C. for TLC.

Bound Lipids. The petroleum ether extracted samples were air dried in their thimbles and hence subjected to water-saturated butanol extraction for the determination of bound lipids. The extraction was made in a laboratory Stein mill

(The Fred Stein Laboratories, Atchinson, Kansas) by treating the material twice with 100 ml. of W.S.B. The first extraction with 100 ml. solvent was made 3 times: 4 min., 2 min., and 2 min. with 1 min. intervals; a 2-minute interval was left between the first and the second extraction. The second extraction was made 3 times for 2 min., with 1 min. intervals and 2 min. interval before opening the blender. The combined extracts were filtered and evaporated almost to dryness under reduced pressure in a glass apparatus at 45-50° C. The extracts were kept in a desiccator under vacuum over P_2O_5 at 4° C. for about 2 days, and hence extracted with petroleum ether (Skelly), cleared by centrifugation (to remove non lipid compounds), and evaporated under reduced pressure at about 40° C.

A similar procedure was used by Daftary (26), C. Chiu (14), and other people in this department. The water-saturated buthanol was prepared by mixing analytical grade 1-butyl-alcohol (normal) with excess distilled water; the upper layer was used for extraction after at least 24 hours from the preparation of the mixture. Determinations were made in duplicate or quadruplicate. The extracted lipids were stored at 4° C. for TLC.

Thin-layer Chromatography. Glass plates (20 x 20 x 0.4 cm.) were coated with a 250 layer of silicagel G for TLC, according to Stahl (E. Merck, A. G. Darmstadt, Germany) with a commercial spreader (C. A. Brinkman Co., Great Neck, N. Y.)

(84, 60, 89). The plates were activated for 3 hours at 130° C. and allowed to cool in a desiccator (14, 26, 75).

The extracted lipids were applied at a level of 100 g per spot. They were dissolved in petroleum ether (Skelly F) and adjusted to a uniform concentration of 20 $\mu\text{g}/\mu\text{l}$ (14, 26), and applied to migrate in the direction of the spreading of the coating. The solvents used for one dimensional ascending adsorption development were: chloroform for non polar lipids (14, 26, 75) and a mixture of chloroform-methanol-water (65:25:4) (chloroform mixture) for polar lipids (89, 60, 75, 14, 26).

All solvents were analytical grade. The chromatography chambers were lined with filter paper in order to insure saturation of the chamber space with solvent vapors (12 hrs.) (89, 60). Fresh solvents were used for every series of assays. Length of run was 15 cm. Time of run for non polar lipid fractionation was 1 hr; for polar lipids, 1½-2 hrs. Visualization of spots was made on air-dry plates after about ½ hr. from their development.

The non-polar fractions were made visible by exposure to iodine vapors. Bright yellow to brown spots on a white background were given by unsaturated compounds; after evaporation of iodine, the plates were sprayed (very slightly) with a chromic H_2SO_4 reagent (8, 84, 60) or with a 50% v/v H_2SO_4 solution. In the case of the former, the plates were heated at 175-180° C. for 20 minutes. In the case of the H_2SO_4 spray,

the plates were heated at 130° C. for 15-20 min., observing the colors of the spots from 100-130° C. Plant sterols were hence identified (6, 60). The charred chromatograms were then photographed under ultraviolet light (60, 8, 84, 75, 26, 14).

The polar compounds were sprayed first with a ninhydrin reagent (58, 89, 60, 84) for detection of free amino groups. The sprayed plates were heated for a few minutes at 100° C., revealing purplish pink spots in the presence of free amino group containing compounds. Once recorded, the plates were hence subjected to a Molybden blue containing spray, specific for any phosphorus containing compounds (blue spots on a white background instantaneously) (29). The spots were again signed on the plate and hence heated at 130° C. (10 min.). The H_2SO_4 contained in the molybdic reagent revealed thus all the lipid fractions (yellow-brown spots on a blue background). The plate was hence photographed under ultraviolet light. Other specific reagents used were the Dragendorff reagent specific for cholin phosphatides (84) (orange spots on a white background) and an α -naphtol containing reagent specific for sugar-containing compounds (glycolipids - blue spots on a white background after heating at 100° C. for 3 min.) (84, 46).

Lipids separated by TLC were tentatively identified by use of specific sprays and by comparing R_f values with those of pure compounds.

Among the non polar compounds, a mixture of reference compounds containing tripalmitin, 1,3-dipalmitin, 1,2-dipalmitin, and 1-monopalmitin; triolein - from the Applied Science Laboratories, State College, Pennsylvania and plant (soybeans) sitosterol- β from the Sygma Chemical Company were used.

For the tentative identification of polar compounds, a mixture containing α -glyceryl-phosphate, lyso-lecithin, and palmitic acid and a mixture containing lecithin, cerebrosides, and monogalactosyl glycerides were used. The pure compounds applied were pure phosphatidyl choline (plant lecithin), phosphatidyl serine, and phosphatidyl ethanolamine - all products from the Applied Science Laboratories, State College, Pennsylvania.

The plates usually were examined under ultraviolet light and photographed mainly when charred with H_2SO_4 50% v/v.

RESULTS AND DISCUSSION

The Oat and Groat Samples

The physical and chemical characteristics of the samples of oats investigated are summarized in Table 1. .

The Andrew variety has the lowest test weight (32.3 lbs./bu.), while Jaycee is the heaviest variety (43 lbs./bu.). The gross composition data are expressed on a 14% moisture content weight basis.

The protein content of the oats varies from 11.2% (Garry)

Table 1. Chemical and physical characteristics of oat samples.¹

Variety	Moisture		Proteins		Lipids		Crude: Carbo- fiber:hydrates ²		Test	
	Ash	(Nx6.25)	Udy	Free	Bound	Per cent	lbs./bu.	designation	weight	Grade
Andrew	9.7	3.75	12.8	11.8	4.55	2.54	11.36	51.0	32.3	No. 3 gray oats
Garry	10.0	2.67	11.2	11.2	4.12	1.15	11.08	54.8	36.0	No. 1 white oats, heavy
Jaycee	9.8	2.67	12.8	12.9	5.35	2.09	9.53	54.1	43.0	No. 1 white oats, extra heavy
Clintford	10.7	2.69	11.7	11.1	4.01	1.99	9.63	56.0	41.2	No. 1 white oats, extra heavy

¹Expressed on a 14 per cent moisture basis.

²By difference.

to 12.8% (Andrew). The ash and crude fiber content is highest in the Andrew oats, which seems to correspond to the lowest test weight of this species. The crude fiber content is higher (11.4%) in the lighter varieties and lower (9.5%) in the heavier ones.

The ash content is rather constant (2.67%) except for Andrew (3.75%). A considerable variability is observed in the lipid content of the four varieties. The total lipid content (free + bound) ranges between 5.27% (Garry) and 7.44% (Jaycee). The free lipid content is highest in the Jaycee variety (5.35%) and lowest in the Clintford and Garry varieties (4.2%).

The highest proportion of bound lipids (35%), with respect to total lipids, corresponds to the highest protein-containing species, while the lowest proportion of bound lipids (21.8%) corresponds to the lowest protein containing variety (Garry).

The gross composition of the groats obtained by dry impact hulling of the oats is summarized in Table 2.

The Andrew variety shows an almost 50% protein level increase as compared to the whole oat grain, which may be explained by the high crude fiber and ash content and hence low test weight of the latter. The protein content (18%) of this species is about 30% higher as compared to the other varieties.

The correlation coefficient obtained between Kjeldahl protein values and proteins determined by the dye-binding

Table 2. Chemical characteristics of groats.¹

Variety	Mois-	Ash	Protein	Udy	Lipids	Crude:	Carbo-
	ture		(Nx6.25)		Free:Bound	fiber:	hydrates ²
	Per cent						
Andrew	11.4	2.04	18.0	15.2	5.79	2.47	1.45
Garry	9.9	2.00	15.0	12.9	5.72	2.09	2.48
Jaycee	8.6	1.90	16.0	14.3	7.45	2.40	2.07
Clintford	10.1	1.82	15.0	13.7	6.09	2.28	2.19

¹Expressed on a 14 per cent moisture basis.

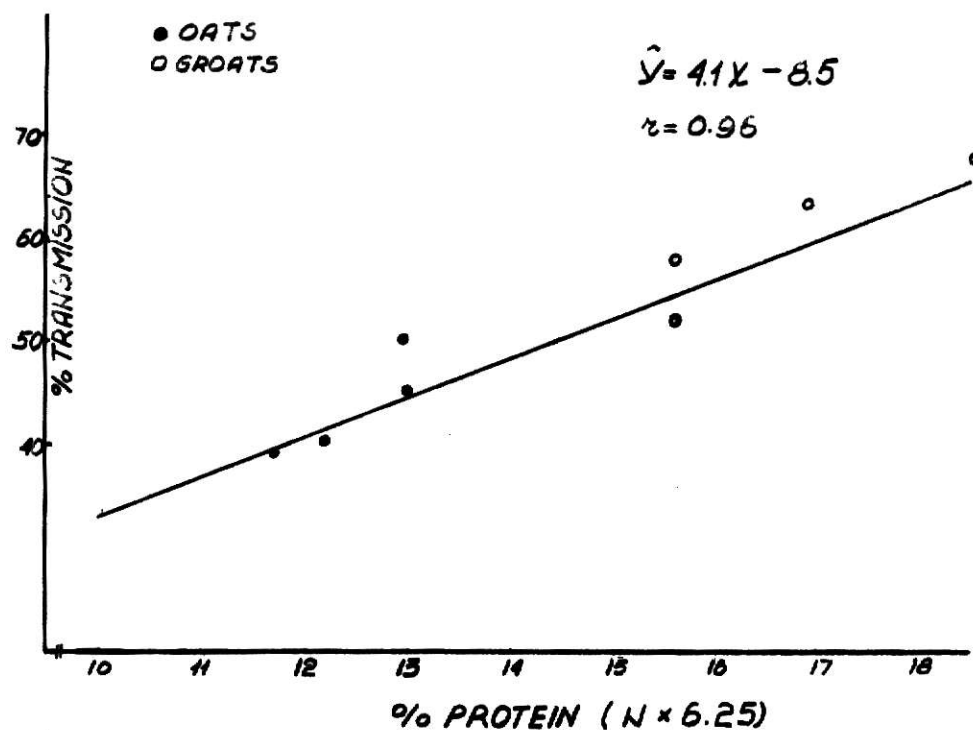
²By difference.

technique on both oats and groats data was highly significant ($r = 0.96$). The regression line passing through the origin and its equation are shown on Fig. 4.

The ash content reveals no variability (2.0%). The crude fiber content is highest in the Garry variety. The lipid content is highest in the Jaycee variety (9.8% free lipids + bound lipids), the lowest fat level being found in the Garry variety (7.8%). Again, the highest proportion of bound lipids (with respect to total lipids (30%)) corresponds to the highest protein variety, while the other varieties contain about 25% bound lipids as compared to the total lipid content.

Air-Classified Flours

Table 3 summarizes some physical characteristics of the "as milled" and air-classified flour fractions. Tables 4 and 5 summarize the variation of each component level through air



† Fig. 4. Dye binding data vs. Kjeldahl values.

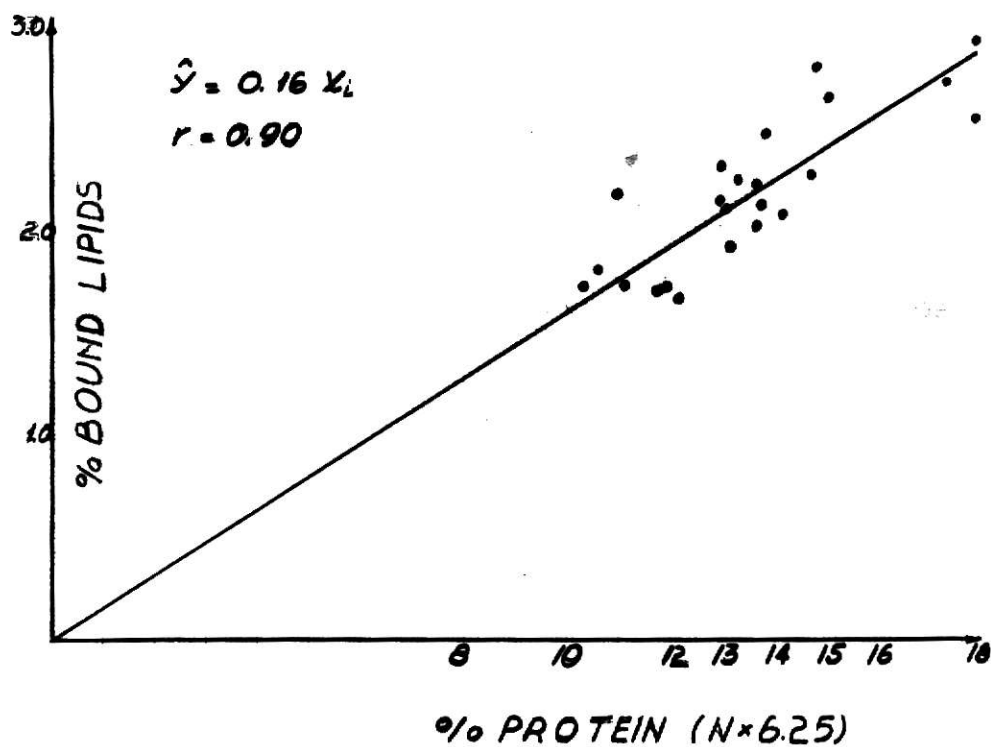


Fig. 5. Bound lipids vs. protein content.

Table 3. Physical characteristics of the "as milled" and air-classified oat flour fractions.

Variety	:	Yield percentage						
Fraction	:	Andrew	:	Garry	:	Jaycee	:	Clintford
A		100.0		100.0		100.0		100.0
B		4.6		1.3		0.7		1.7
C		19.2		9.0		6.3		23.3
D		23.2		19.7		20.7		21.7
E		13.0		29.7		28.6		17.6
EE		40.0		40.3		43.7		35.7

Variety	:	Fisher average particle size μ						
Fraction	:	Andrew	:	Garry	:	Jaycee	:	Clintford
A		11.8		14.0		14.2		12.4
B		5.8		5.3		5.6		6.2
C		5.3		6.7		7.2		6.5
D		7.7		8.0		8.2		8.4
E		12.4		10.9		10.0		10.4
EE		18.0		16.9		15.6		19.6

Variety	:	Agtron color							
	:	Andrew		Garry		Jaycee		Clintford	
Fraction	:	dry	"wet"	dry	"wet"	dry	"wet"	dry	"wet"
A		45.0	22.0	32.5	11.0	29.5	17.0	34.5	20.5
B		46.5	27.5	47.0	23.0	39.0	-	34.0	21.0
C		49.5	32.0	36.5	24.5	36.0	33.0	42.5	32.0
D		47.5	27.0	44.0	23.0	45.5	34.0	46.0	27.5
E		35.5	19.5	33.5	-	35.5	30.0	34.5	19.0
EE		14.5	0.0	7.0	0.0	14.0	0.0	18.0	10.0

Table 4. Chemical characteristics of the "as-milled" and air-classified oat flours.¹

Variety Fraction	Moisture percentage			
	Andrew	Garry	Jaycee	Clintford
A	11.6	10.6	9.6	11.9
B	10.0	10.2	11.3	10.6
C	9.6	9.7	9.7	10.0
D	10.6	10.3	10.1	10.6
E	11.0	10.9	10.1	10.8
EE	10.8	10.7	9.9	11.2

Variety Fraction	Ash percentage			
	Andrew	Garry	Jaycee	Clintford
A	1.00	1.15	1.52	0.95
B	1.24	1.05	1.35	0.96
C	0.92	0.89	0.89	0.70
D	0.92	0.92	0.95	0.79
E	0.79	0.86	0.91	0.75
EE	0.88	1.54	1.91	1.16

Variety Fraction	Crude fiber percentage			
	Andrew	Garry	Jaycee	Clintford
A	1.07	1.35	1.33	1.16
B	1.43	0.57	0.71	1.06
C	1.14	1.04	1.05	1.05
D	1.06	0.90	0.53	1.06
E	0.82	0.96	0.76	1.15
EE	1.42	1.73	1.90	1.16

Variety Fraction	Carbohydrates percentage (by difference)			
	Andrew	Garry	Jaycee	Clintford
A	52.2	63.9	59.5	65.1
B	56.4	59.1	-	62.9
C	17.2	62.6	62.4	63.2
D	51.5	63.1	61.0	63.7
E	68.2	66.0	62.7	66.3
EE	62.3	63.5	56.2	66.5

¹Expressed on a 14 per cent moisture basis.

Table 5. Protein and lipid content of the "as milled" and air-classified fractions.¹

Variety	: Free lipids percentage (petroleum ether extract)			
Fraction	: Andrew	: Garry	: Jaycee	: Clintford
A	5.03	5.71	7.76	5.23
B	6.41	5.13	-	5.22
C	5.73	6.28	6.34	5.22
D	4.82	5.59	7.18	5.35
E	3.82	5.05	6.46	4.97
EE	4.82	5.70	8.26	5.16

Variety	: Bound lipids percentage (water-saturated buthanol, following petroleum ether extract)			
Fraction	: Andrew	: Garry	: Jaycee	: Clintford
A	2.25	1.64	2.11	1.70
B	2.53	2.70	-	2.20
C	2.89	2.07	2.30	2.00
D	2.78	2.23	2.45	1.90
E	1.78	2.17	2.13	1.72
EE	2.30	1.60	2.60	1.72

Variety	: Protein percentage							
Fraction	: Andrew	: Garry	: Jaycee	: Clintford	: Andrew	: Garry	: Jaycee	: Clintford
	: Kjcl.: Udy	: Kjcl.: Udy	: Kjcl.: Udy	: Kjcl.: Udy	: Kjcl.: Udy	: Kjcl.: Udy	: Kjcl.: Udy	: Kjcl.: Udy
A	14.7	14.1	12.2	11.8	13.8	13.8	11.9	11.5
B	18.0	15.4	17.4	16.3	13.0	12.2	13.7	11.9
C	18.1	16.9	13.1	13.0	13.0	12.0	13.7	12.7
D	14.9	14.6	13.3	12.7	13.9	11.9	13.2	12.3
E	10.6	9.7	11.0	10.1	13.0	11.2	11.1	10.3
EE	14.2	14.2	11.8	11.8	15.1	15.6	10.3	9.9

¹Expressed on a 14% moisture basis.

fractionation for all flour varieties. The histograms on Fig. 6 (a, b) and Fig. 7 (a, b) illustrate these variations for each variety; the broken lines represent the level of the examined parameter, found on the "as milled" flour; the abscissa represents the yield percentage obtained for each fraction; the y axis shows the level of the examined parameter, as found for each air-classified fraction of the given variety.

In examining the oat flours' response to air fractionation, the high fat content of the oats endosperm, as compared to other cereal grains (2, 53, 7), should be kept in mind.

Yield percentage obtained for the fine fractions seems to reflect this very fact. The yield percentage in fine fractions B + C was highest in the Andrew and Clintford varieties (around 25%) while it was only 7% in case of the Jaycee variety. The yield in coarse (residual fraction) was rather constant for all varieties (40%) (Table 3, histograms Figs. 6, 7).

The Fisher average particle size is expressed in μ of average diameter of particles contained in a given fraction and is an index of specific surface of the given fraction (Gracza, 1959) without actually indicating the true particle size distribution of the given flour.

In this experiment, the parameter varies from 5-7 μ for the fine fractions, from 8-12 μ for the intermediary fractions, between 12-14 μ for the "as milled" flours, and from 16-20 for the coarse fractions.

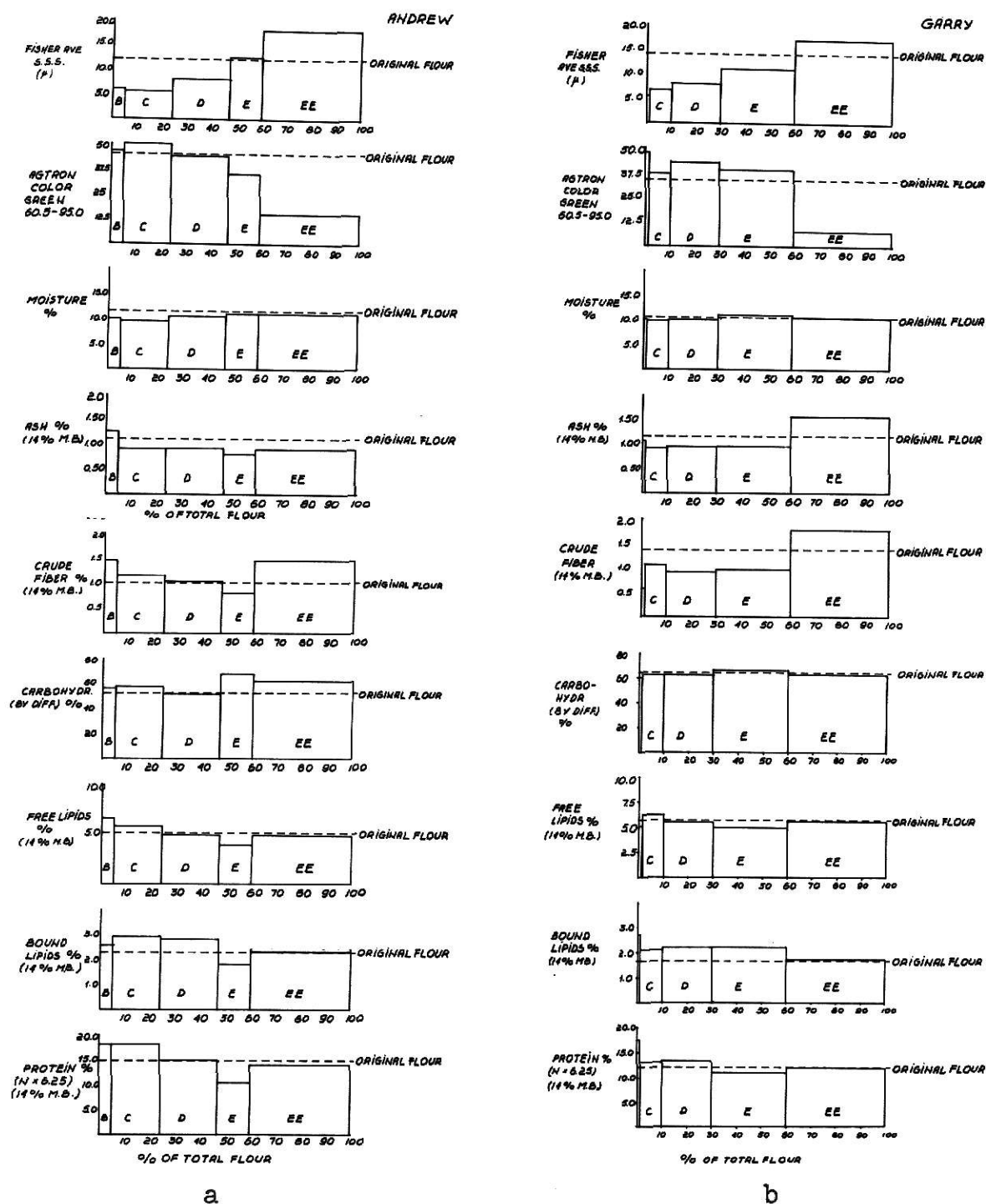


Fig. 6. Variation of gross composition of air-classified flours. (a) Andrew oats; (b) Garry oats.

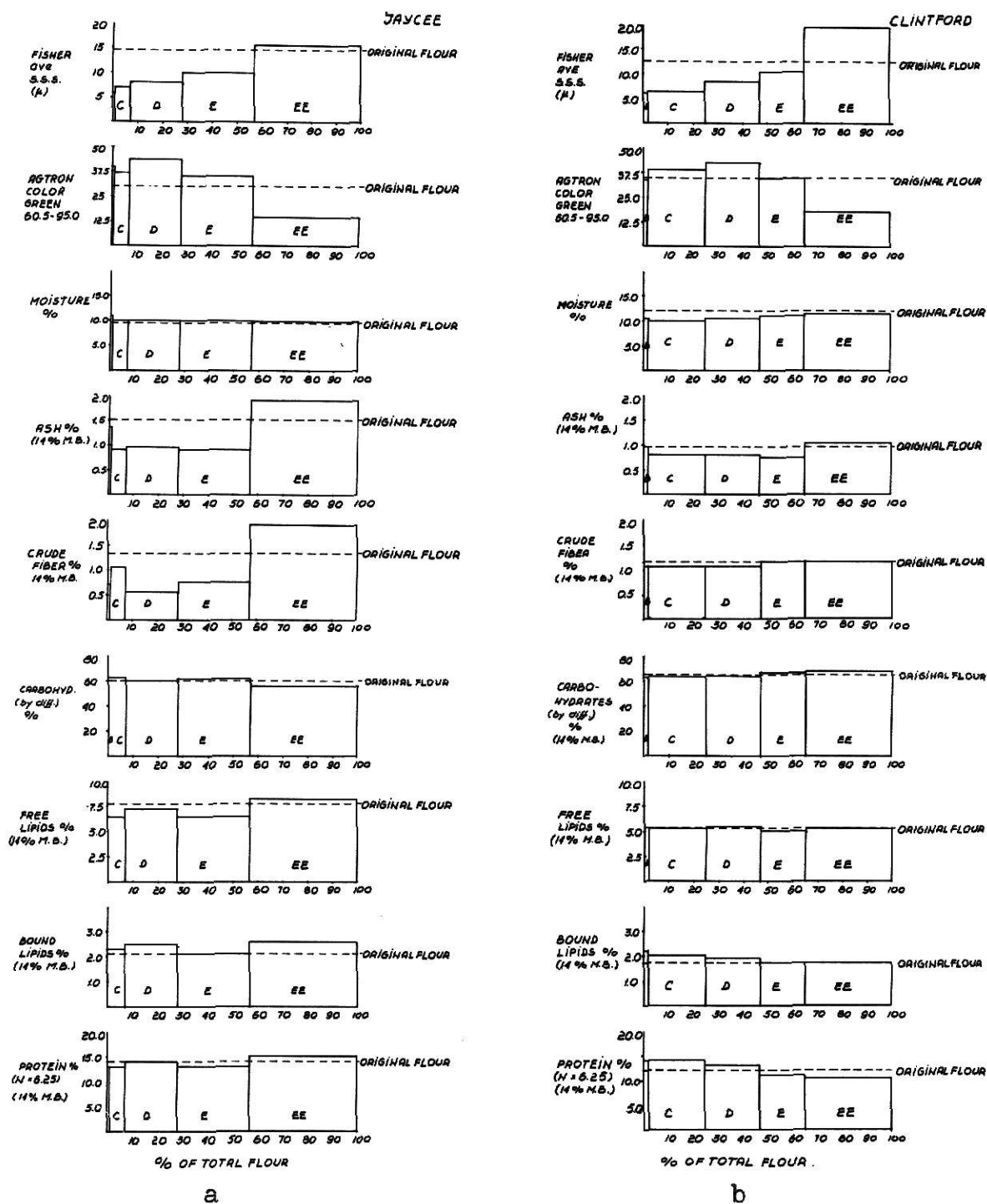


Fig. 7. Variation of gross composition of air-classified flours. (a) Jaycee oats; (b) Clintford oats.

The moisture content (Table 4) of the Fine B and C fractions is lower than that of the parent flours. This may be expected as a result of air handling of small particles which have a larger specific surface. The Jaycee variety makes an exception, showing in the B fraction a higher moisture content than the "as-milled" flour. This may be due to the stickiness of the high fat containing species which possibly led to a clustering of flour particles.

The Agtron color reading is a function of a monochromatic reflectance comparison of the sample and a reference screen. An increase in reflectance is found with the decrease of particle size. Jaycee and Clintford showed the highest reflectance on the D fraction, with an average Fisher diameter of 8μ . The lowest reflectance was found on the coarse fractions as would be expected. The Garry variety showed a substantially lower reading than the other varieties which may be expected from its highest crude fiber content as compared to the other varieties (Table 2).

The results of the "wet" Agtron readings showed a more uniform peak of reflectance for the C fraction in all varieties. The coarse fractions were not readable by the "wet" Agtron procedure, except for the Clintford variety which was the lowest crude fiber containing species. The correlation coefficient between Agtron color and ash content on the whole set of data ($r = 0.6$) was not significant for an $\alpha = 0.05\%$. When examined per variety, a highly significant negative

correlation was found for both the Jaycee and the Clintford varieties ($r = -0.86$). The correlation between "wet" and "dry" Agtron was significant ($r = 0.7$). The negative correlation coefficient ($r = -0.45$) between "wet" Agtron and ash was not significant at the $\alpha = 0.05\%$ level.

The ash content appears increased (by 24%) in the fine fraction only in the Andrew flour (hystogram, Fig. 6a) where this increase accompanies a positive protein shift. This increase may be due to a high amount of cell wall material, which separates in the finest fraction; it also may be due to the presence of protein sheaths higher in ash than the small starch granules or it may be due to very small fragments of bran and to other dusty material.

The ash content drops in the coarse fraction of the Andrew flour by 20% with respect to the original flour. This may be due to the increase in starch granules on one hand (20% carbohydrates increase, Table 4) and also to a considerable depletion of protein and mineral materials from the coarse fraction in favor of the fine B and C fractions. In all the other varieties, the maximum ash content is found in the coarse fraction probably as a result of the accumulation of bran and aggregates containing higher ash level cell wall material; on the other hand, the yield in fine fractions was much lower in these flours than in case of the Andrew variety, which may account for some ash which eventually would have accompanied the fine particles.

No significant correlation was found between ash and particle size ($r = 0.41$) on the whole set of data. When examined per variety, only Garry showed a significant $r = 0.76$. Ash vs. crude fiber gave a high correlation in the Jaycee variety ($r = 0.916$), and in the Garry variety ($r = 0.76$). No significant correlation was found between ash and protein content ($r = 0.23$) as would be expected for other cereals. The only variety with a significant $r = 0.76$ was Jaycee, where most likely, the stickiness of the material pulled the compounds together, leading them to a shift towards the coarse fraction.

Crude fiber was highest in all coarse fractions as would be expected as a result of bran accumulation. It was, however, increased in the fine B fraction of the Andrew variety, where the higher yield obtained for this fraction probably involved bran dust particles; this also was seen from its increased ash content.

Carbohydrates calculated by difference showed a peak in the E fraction for all varieties as would be expected. They also presented a positive shift along with protein in the fine B-C fractions of the Andrew variety (as inferred earlier from the low protein shift). A minimum of carbohydrates is found in the B fraction of the Garry variety which showed the highest protein shift. The Jaycee flour seems to have released starchy granules all along the process of fractionation where a minimum of carbohydrates is found in the coarse fraction.

Protein. The protein content was computed by using the factor 6.25 for converting the Kjeldahl N values (Table 5). The correlation coefficient for the data obtained by the dye binding method for all air-classified samples vs. Kjeldahl protein was not significant ($r = 0.4$). This discrepancy is most likely due to the clustering of fine particles which hinders physically the contact between particle surface and dye reagent. The tendency of the oat flour fine particles to clustering seems to be the consequence of the high fat content of the oat endosperm. However, when used according to manufacturer's prescriptions, as mentioned before, on oats and groats the correlation is highly significant. Also when used on air-classified fractions, with the fine fractions omitted, r improves to 0.94 which, however, could not be considered a fair evaluation.

The hystrogram in Fig. 6 a indicates a positive protein shift of 20% from the parent flour to the fine fractions B and C in the Andrew oats. This low protein shift, as compared to shifts produced in other cereal flours, may be due again to the high fat content of the endosperm of the oat grain and on the other hand, it may be due to a dilution with very fine starch particles.

Kent emphasized the preponderance of compound starch granules in the oat endosperm (53). These granules may contain up to 80 individual granules which range in size between $2-10\mu$. According to Rozsa, Ward et al., the protein and starch

particles side by side, have identical flow dynamic characteristics, i.e., common settling time (77). A critical cut by flow dynamic separation will treat protein matter twice the size of starch granules alike and grade them together into the same fraction of a separation.

The coarse fraction in the Andrew flour is slightly lower in protein content than the parent flour wherefrom protein depletion and shift towards the B and C fractions may eventually explain this occurrence.

The Garry variety showed a 40% protein shift towards the B fraction; however, the yield of this fraction was very low (Histogram, Fig. 6b).

The Clintford flours revealed a 20% shift towards B, C, and D, which led to a slight drop in protein level in the coarse EE fraction as in the Andrew oats.

The Jaycee variety had the highest protein content in the coarse fraction. No protein shift was observed in any fine fraction. Probably, being the highest fat containing species, an accumulation of intact cell clusters occurred in the coarse fraction and no protein has been released during the process. It seems that only starch granules were released during the fractionation (Fig. 7a). The lowest protein content was found in the E fraction in all varieties, which is a starchy fraction.

Lipids. The distribution of the lipids in the air-classified flour fractions is summarized in Table 5 and illustrated

on the hystograms in Figs. 6 and 7. The original flour A and the residual fraction EE are comparable total (free + bound) lipid content. The fine fractions B and C generally follow the protein shift, mainly as far as bound lipids are concerned. The lowest lipid content is always observed in the starchy E fraction.

The Jaycee variety, due to its particularly high lipid content, shows a different pattern in air fractionation; almost no B fraction was obtained and, apparently, only starchy depletion occurred during this process. Hence all the lipid- and protein-containing particles concentrated in the coarse EE fraction which appears higher in both lipid and protein level as compared to the original flour.

The proportion between free and bound lipids is illustrated in the hystograms on Fig. 8. The black portion represents the bound lipid content, the clear portion corresponds to the free lipids. The broken and dotted lines represent the corresponding levels of the "as-milled" flours.

The highest proportion of bound lipids is found in the Andrew flours, as mentioned before. The Garry and Clintford varieties of similar protein level (which was lower than in the Andrew variety) showed a lower content of bound lipids. The Jaycee variety (the highest lipid-containing species) showed the lowest bound lipid content.

No significant correlation between free and bound lipids was found. A rather high correlation ($r = 0.90$) between bound

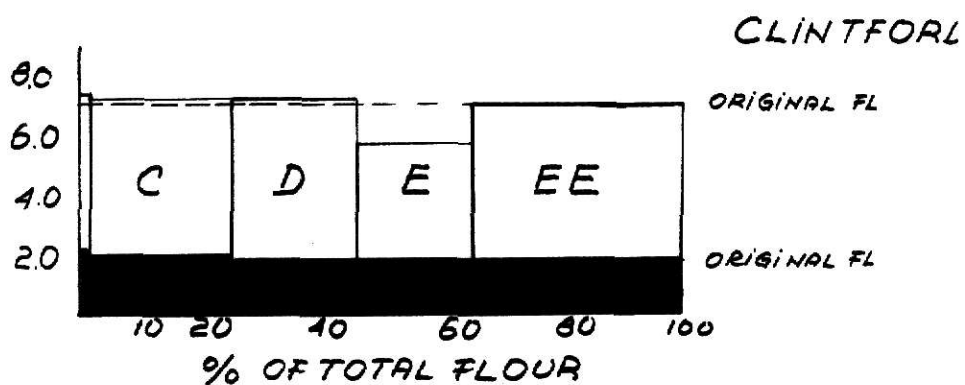
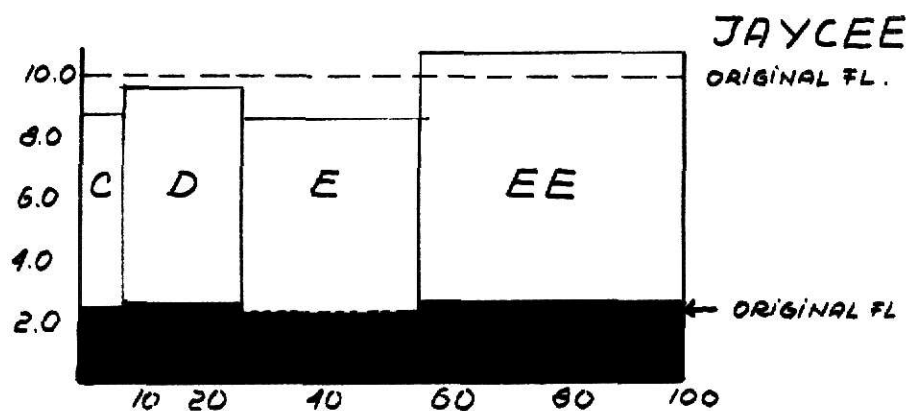
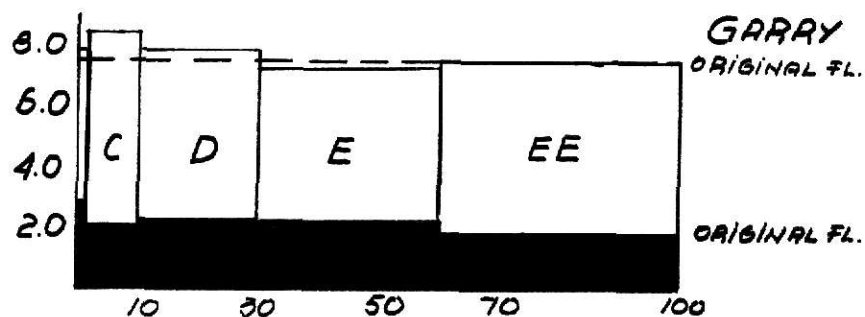
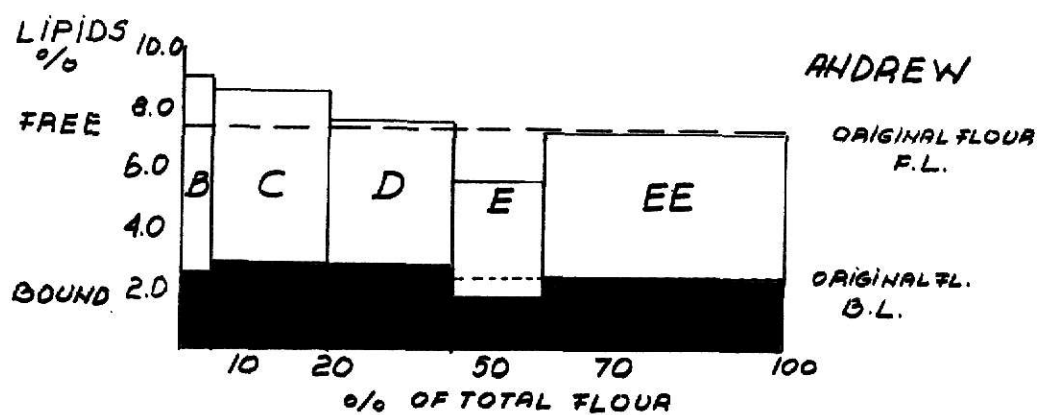


Fig. 8. Free and bound lipid content of air-classified flours.

lipids and protein level was observed, with the regression line passing through the origin (Fig. 5). No significant correlation between free lipids and protein content was found.

Thin-layer Chromatography. The TLC of the lipids is shown in Figs. 9-20. The fractionation of non polar lipids shows that triglycerides, traces of free fatty acids, and 1,3-diglycerides and 1,2-diglycerides were present mostly as free lipids, extractable with petroleum ether. Small amounts of triglycerides and diglycerides were present in a bound form in the oats and groats (the lowest amount was found in the Jaycee variety by visual comparison). Traces of triglycerides in bound form were detected in the coarse fraction of the air-classified flours; diglycerides in very small amounts also were found in bound form in the intermediary and coarse fractions of the Clintford variety (Fig. 19). In the case of groats and air-classified fractions, the spot corresponding to 1,3-diglycerides appeared to be a double layer, containing some plant sterols in addition to the mentioned diglycerides.

Both the petroleum ether extract and the butanol, following petroleum ether extract, contained considerable amounts of polar lipids. The concentration of polar lipids in the water saturated butanol extract was substantially higher than in the petroleum ether extract. The lowest concentration of polar lipids was found in the extract obtained from ground oats.

The following classes of polar lipid compounds were

tentatively identified in the water-saturated buthanol extracts:

- a) neutral, non polar compounds, eventually containing sterol esters; however, not identified.
- b) monogalactogyl diglycerides in the W.S.B. extract and triglycerides in the pet. ether extract.
- c) some type of plant cerebrosides according to the reference compounds and specific reaction (α -naphtol +); the possibility of their occurrence in cereal grains was described by Houston (41).
- d) digalactogyl glycerides.
- e) phosphatidyl ethanolamines.
- f) unknown.
- g) lecithins.
- h) phosphatidyl serines.
- i) lysolecithins.
- k) sugars (free).
- l) aminoacids.

No substantial qualitative differences were detected between the four oat varieties, their groats, or their air-classified fractions. By visual comparison, the EE (coarse fraction) revealed a lower content in polar lipids (except in the Jaycee flours). The air-classified fractions of the Clintford oats seemed to have one more phosphorus-containing lipid compound as compared to the other varieties.

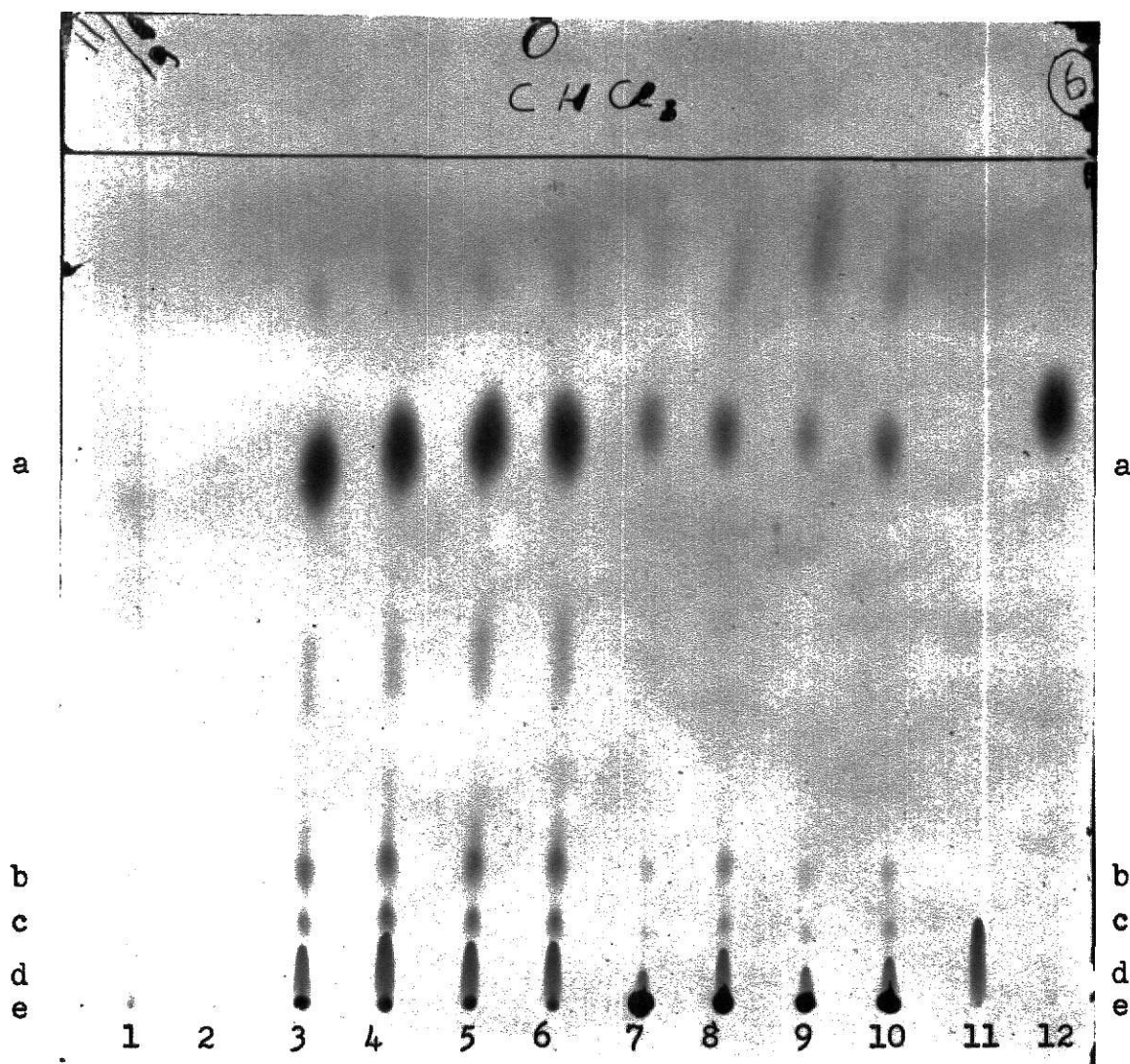


Fig. 9. TLC of free and bound non polar lipids of oat samples. From left to right: 1) reference mixture containing a) tripalmitin, b) 1,3-dipalmitin, c) 1,2-dipalmitin, d) 1-monopalmitin; 2) oleic acid, 3-6) petroleum ether extract of four oat varieties in the order: Andrew, Garry, Jaycee, Clintford; 7-10) Water Saturated Butanol following petroleum ether extract of the same samples in the same order; 11) as 2; 12) triolein. Reference lipids were applied at a 25 γ /spot level, oat lipids at a 100 γ /spot level. Developed with chloroform spots charred with H_2SO_4 50% v/v. Picture taken under UV light. Tentatively identified as: a) triglycerides, b) 1,3-diglycerides, c) 1,2-diglycerides, d) free fatty acids, e) unresolved polar compounds.

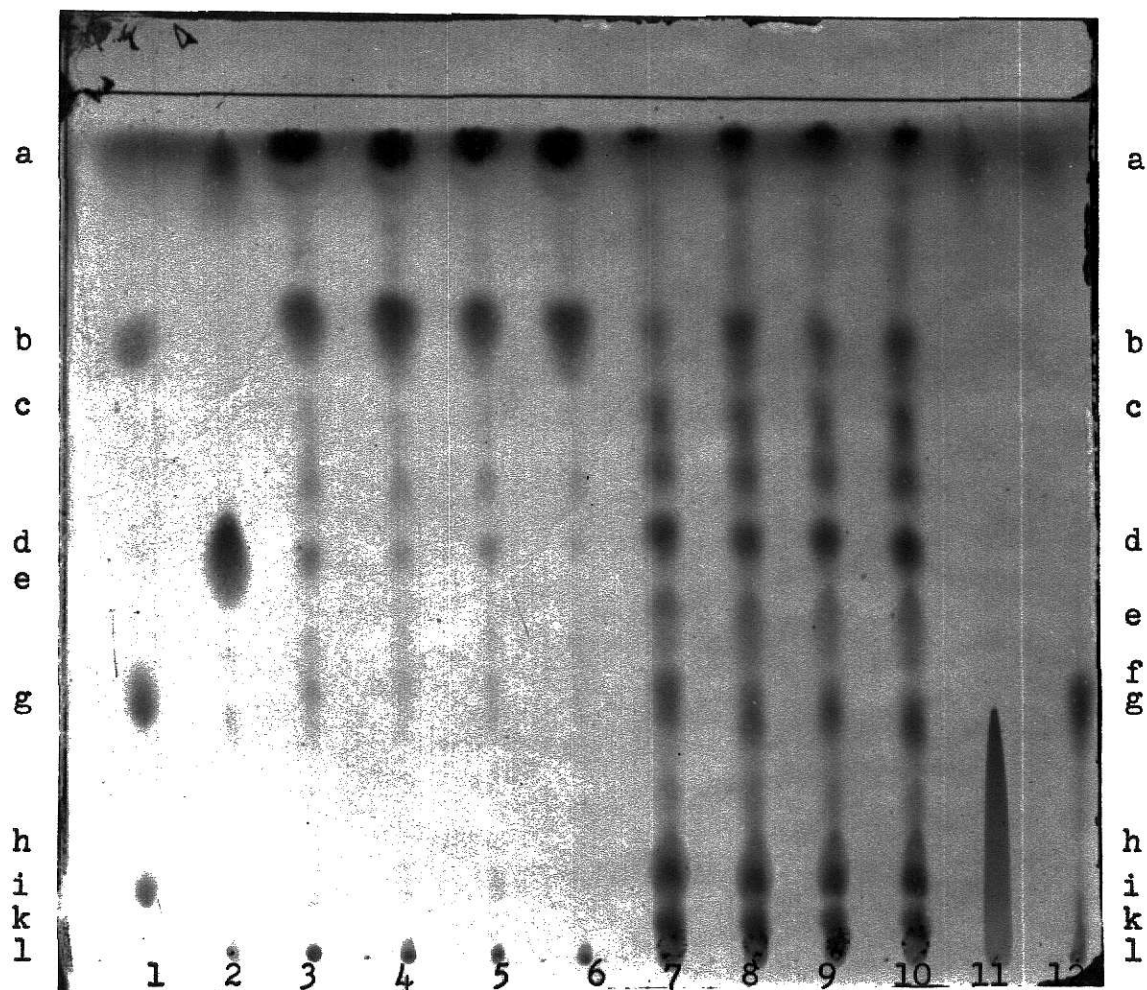


Fig. 10. TLC of free and bound polar lipids of four oat varieties. From left to right: 1) mixture of 3 reference compounds (b: palmitic acid, g: lecithin, i: lysolecithin), 2) phosphatidyl ethanolamine, 3-6) petroleum ether extract of four oat varieties in the same order as in Fig. 9; 7-10) Water saturated butanol following petroleum ether extract of the same samples, 11) phosphatidyl serine, 12) lecithin. Reference lipids applied at a 25 γ /spot level, oat lipids at a 100 γ /spot level. Developed with chloroform mixture ($\text{Cn}_3\text{Cl}:\text{Cn}_3\text{ON}:\text{H}_2\text{O}$; 65:25:4). Tentatively identified as: a) non polar compounds, b) monogalactosyl glycerides (right) and triglycerides (left), c) plant cerebrosides, d) digalactosyl diglycerides, e) phosphatidyl ethanolamines, f) unknown, g) lecithins, h) phosphatidyl serines, i) lysolecithins, k) sugars, l) amino acids.

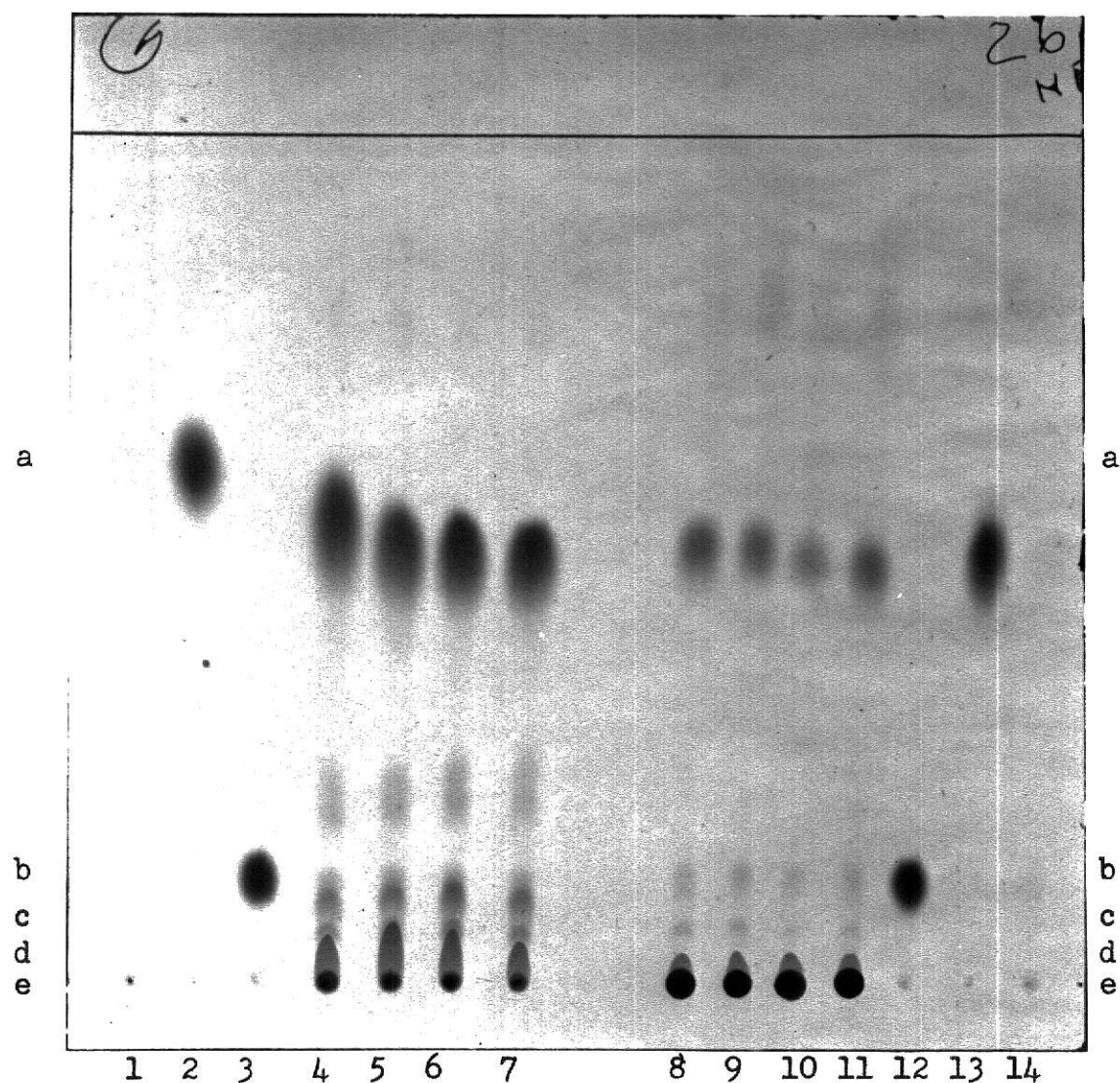


Fig. 11. TLC of non polar lipids of goat samples. From left to right: 1) reference mixture containing: a) tri-palmitin, b) 1,3-dipalmitin, d) 1,2-dipalmitin, e) 1-mono-palmitin, 2) triolein, 3) plant sitosterol, 4-7) goat lipids from four oat varieties extracted with petroleum ether, order as in Fig. 9; 8-11) goat lipids extracted with W.S.B. following petroleum ether, 12) plant sitosterol, 13) triolein, 14) reference mixture as 1). Tentatively identified as in Fig. 9; however, the b spot is visualized as a double layer containing also plant sterols. Other conditions as in Fig. 9.

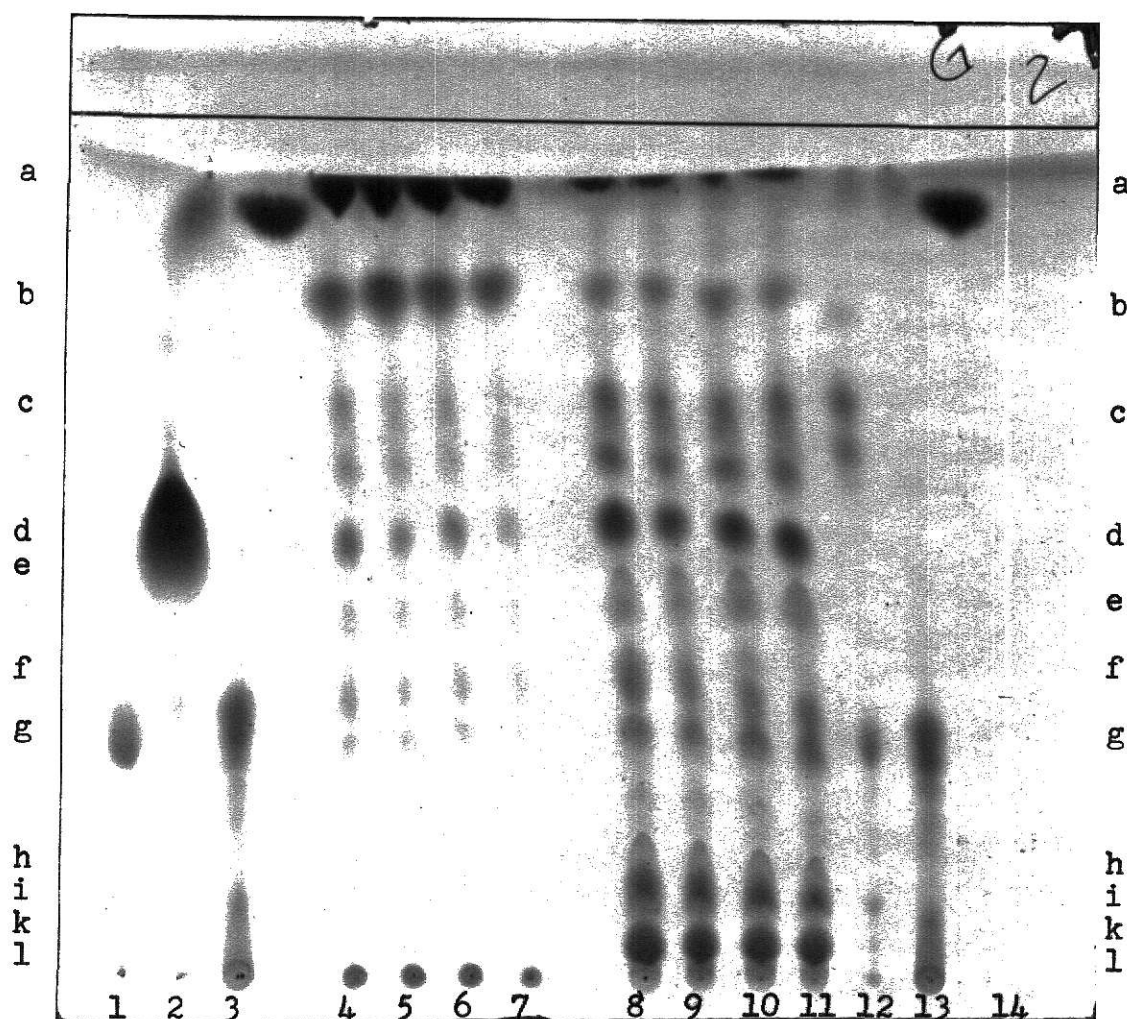


Fig. 12. Comparing polar lipid content in petroleum ether and W.S.B. following petroleum ether extracted lipids of four oat groat varieties. From left to right: 1) lecithin (g) and lysolecithin (k); 2) phosphatidyl ethanolamine, 3) lecithin, 4-7) four oat groat varieties in the same order as in Fig. 9 (petroleum ether extract), 8-11) the same samples extracted with W.S.B. following petroleum ether, 12) mixture of reference compounds containing (c) cerebrosides, (g) lecithin, 13) lecithin. Developed with chloroform mixture as in Fig. 10. Tentatively identified as: a) unresolved non polar compounds, b) monogalactosyl glycerides (right); triglycerides (left, c) plant cerebrosides, d) digalactosyl diglycerides, e) phosphatidyl ethanolamines, f) unknown, g) lecithins, h) phosphatidyl serines, i) lysolecithins, k) sugars, l) amino acids.

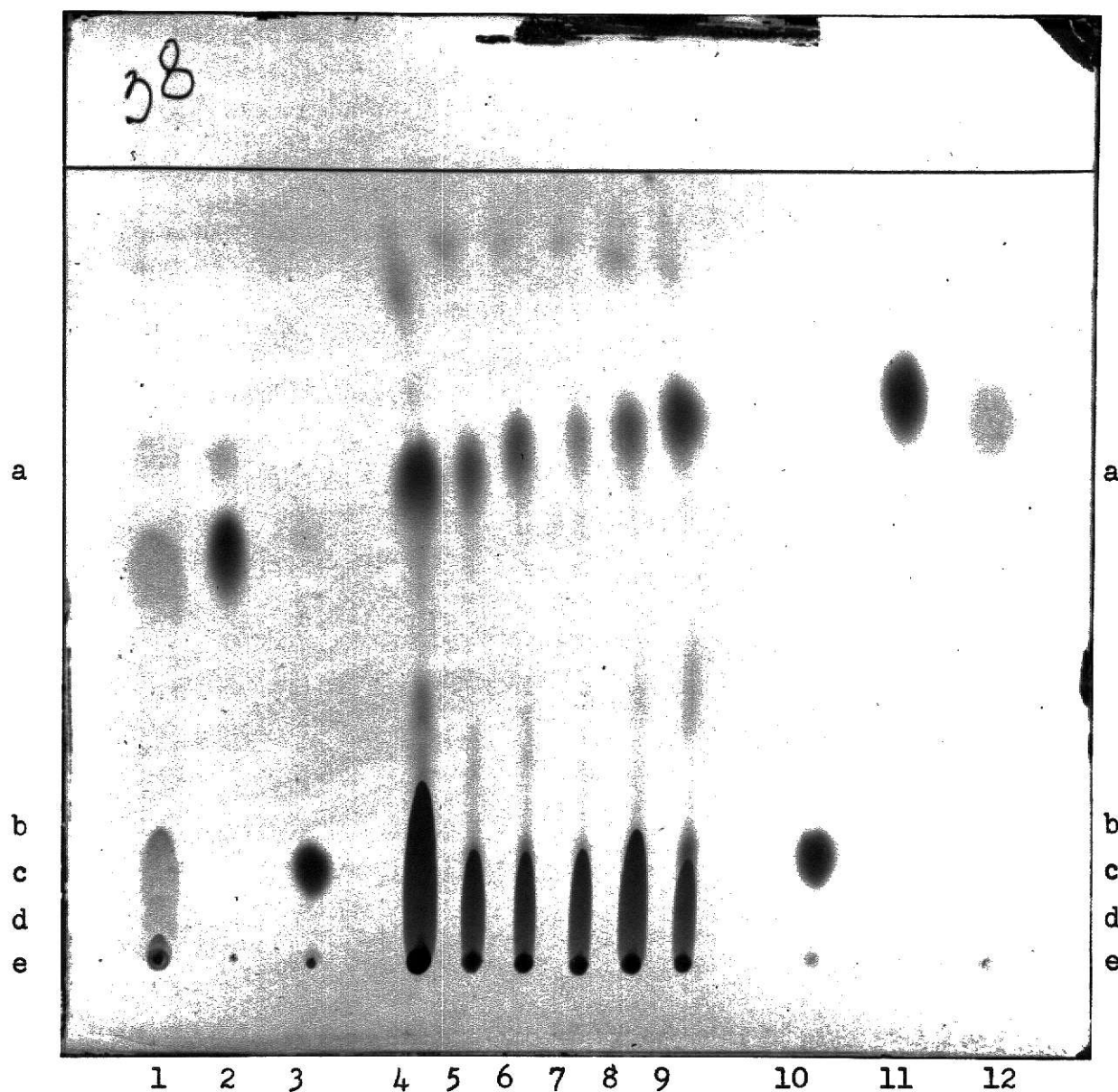


Fig. 13. Comparing non polar lipids extracted with petroleum ether from air-classified Andrew flours. From left to right: 1-3) as in Fig. 11; 4) original flour, 5) fraction B, 6) fraction C, 7) fraction D, 8) fraction E, 9) fraction EE, 10-12) as 3-1. Developed with chloroform. Tentatively identified as in Fig. 9.

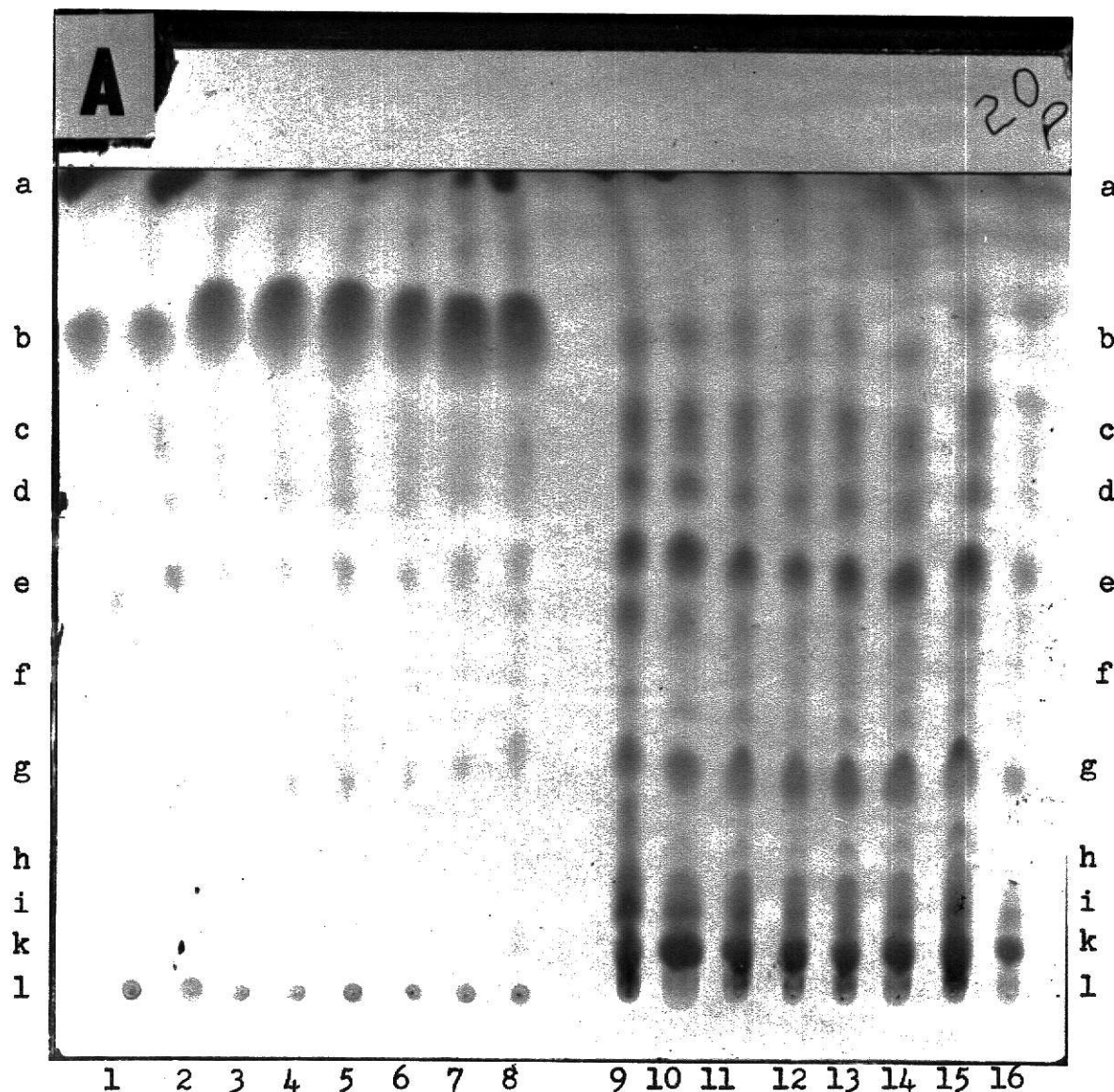


Fig. 14. Comparing polar lipid content of Andrew oats, groats, original, and air classified flours.

From left to right: 1) oat lipids, 2) groat lipids, 3) original flour, 4-8) B, C, D, E, EE fractions--extracted with petroleum ether; 9-16) in similar order, polar lipids extracted with W.S.B. following petroleum ether. Developed with chloroform mixture. Tentatively identified as in Fig. 12.

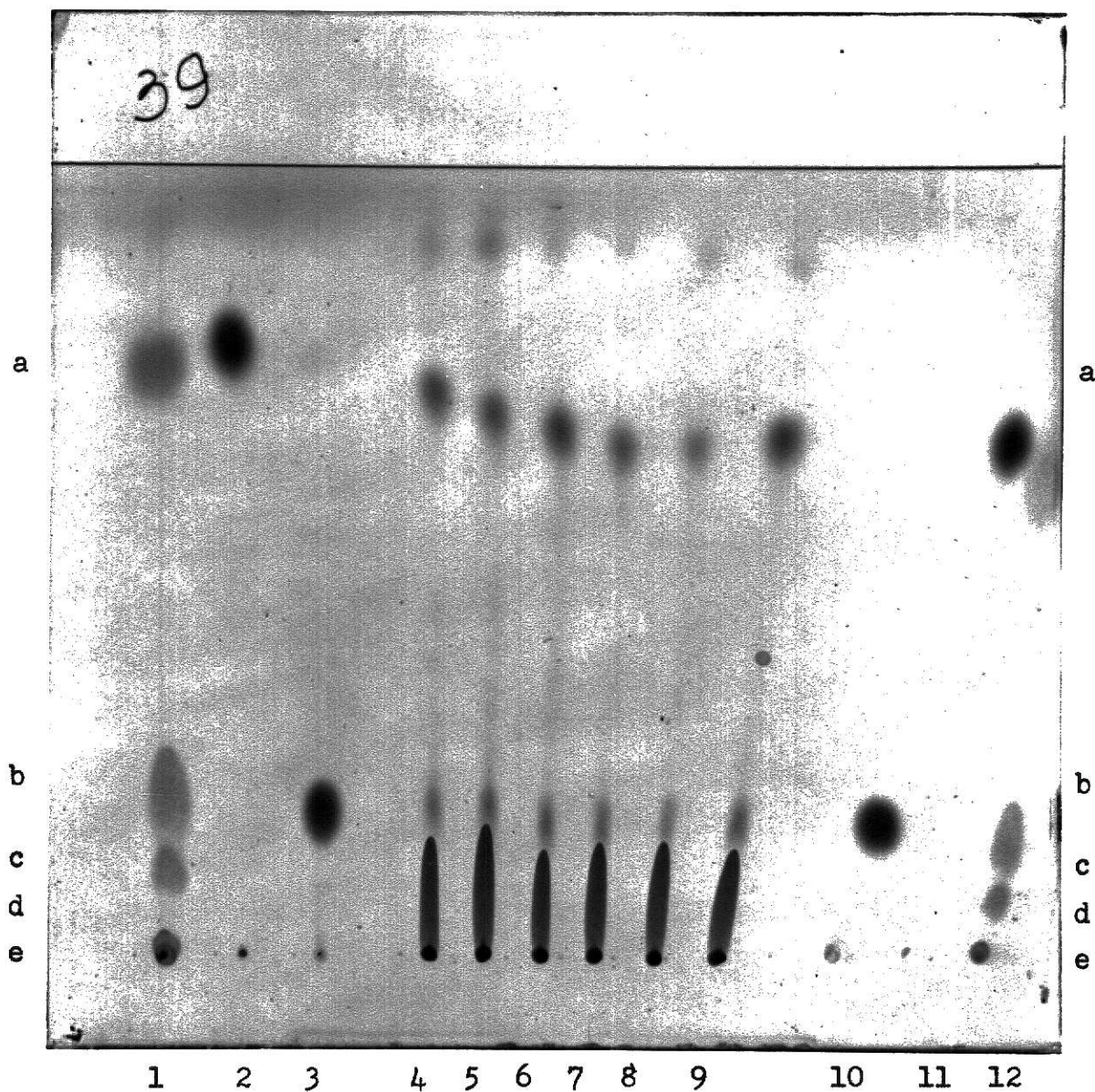


Fig. 15. Comparing non polar lipids in Garry original and air-classified flours.

Sample sequence as in Fig. 13. Developed with chloroform. Tentatively identified as in Fig. 11.

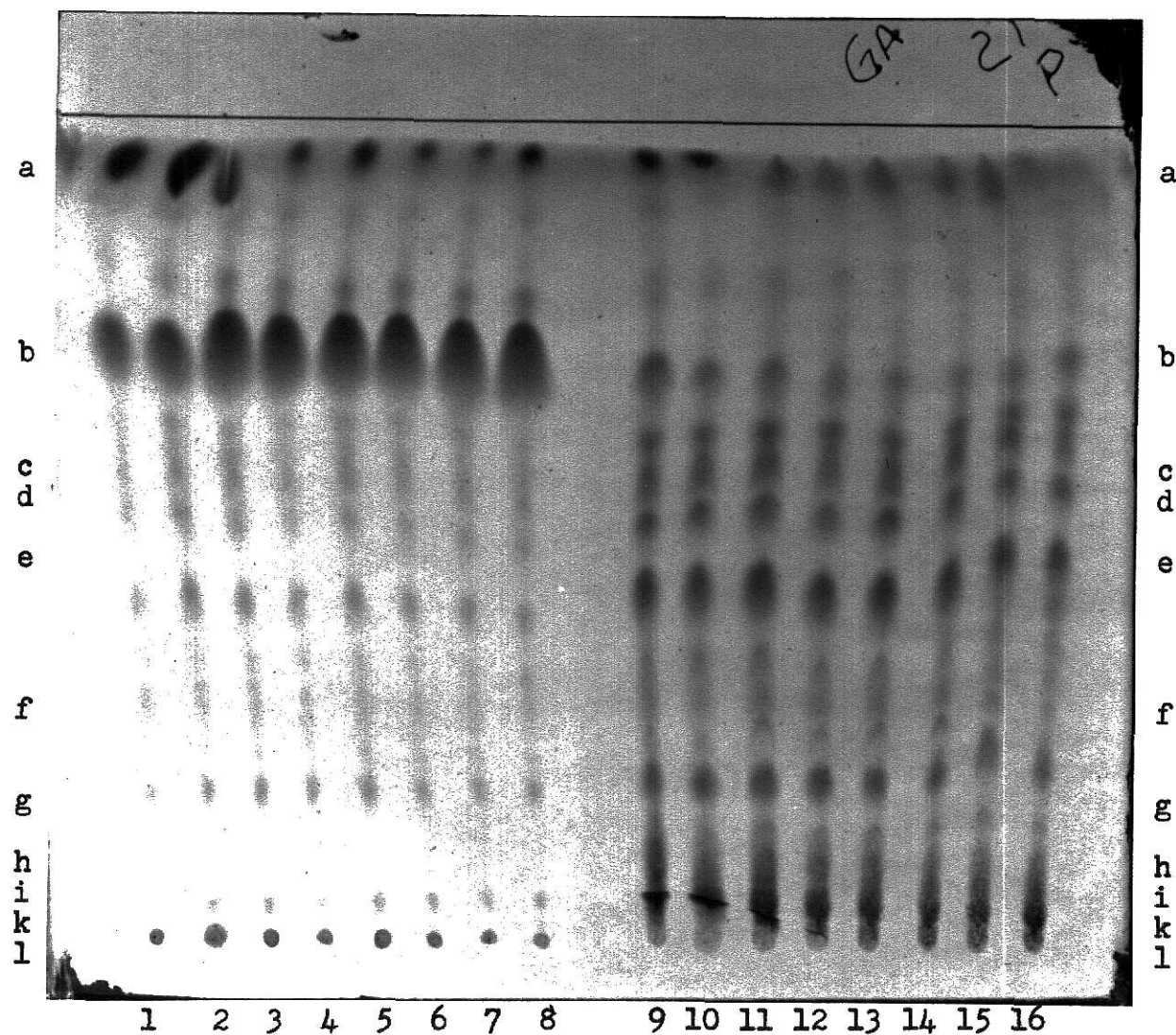


Fig. 16. Comparing polar lipids in samples extracted from oats, groats, original and air-classified flours of the Garry oats.

Sample sequence as in Fig. 14. Developed with chloroform mixture. Tentatively identified as in Fig. 12.

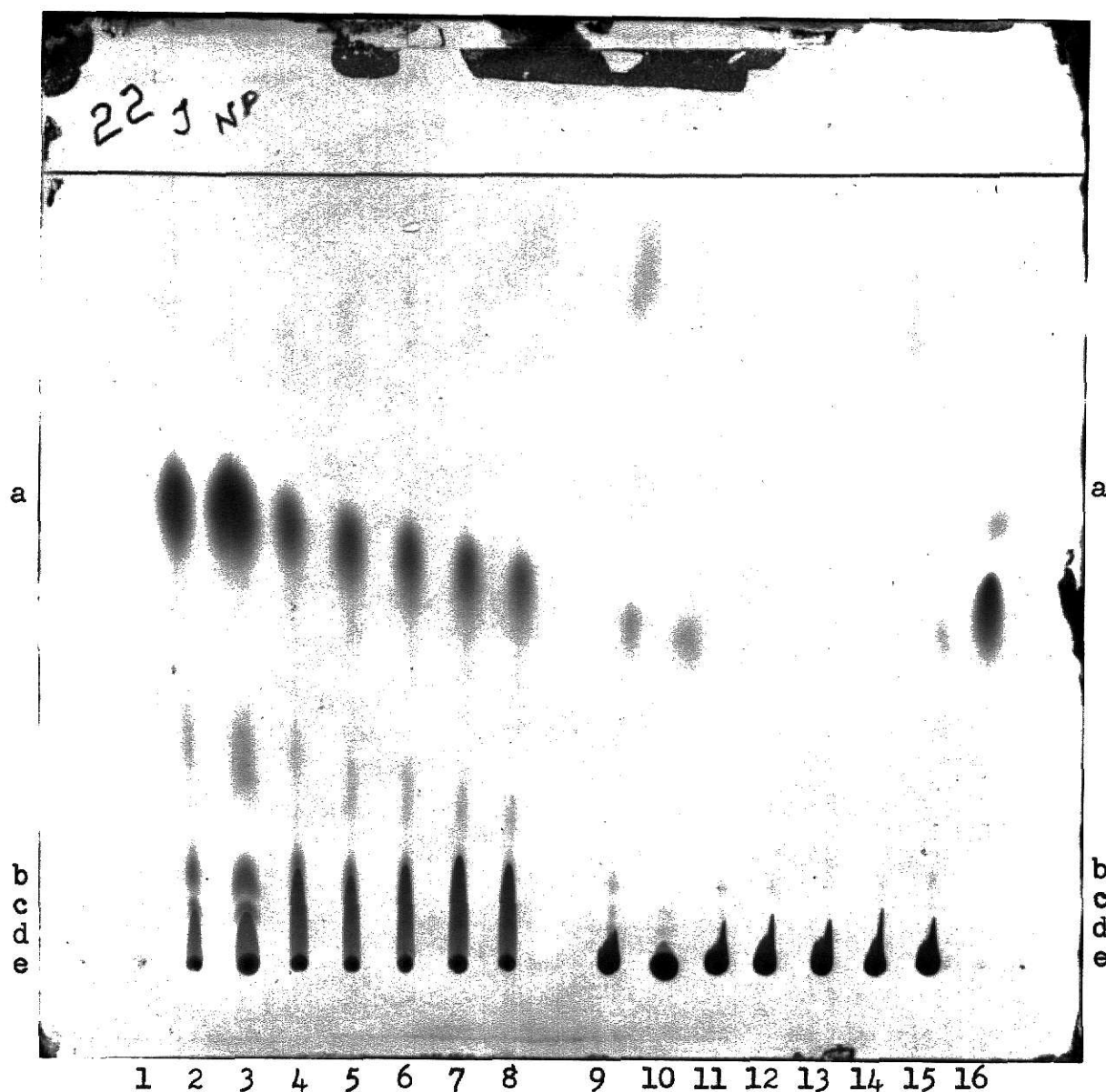


Fig. 17. Comparing non polar lipids in samples extracted from oats, groats, original and air-classified flours of the Jaycee oats. LEFT: Petroleum ether extract; RIGHT: W.S.B. following petroleum ether.

Sample sequence as in Fig. 14. Only 1) is the reference mixture used in 1) Fig. 9; 16) is triolein. Fraction B is missing. Developed with chloroform. Tentatively identified as in Fig. 11.

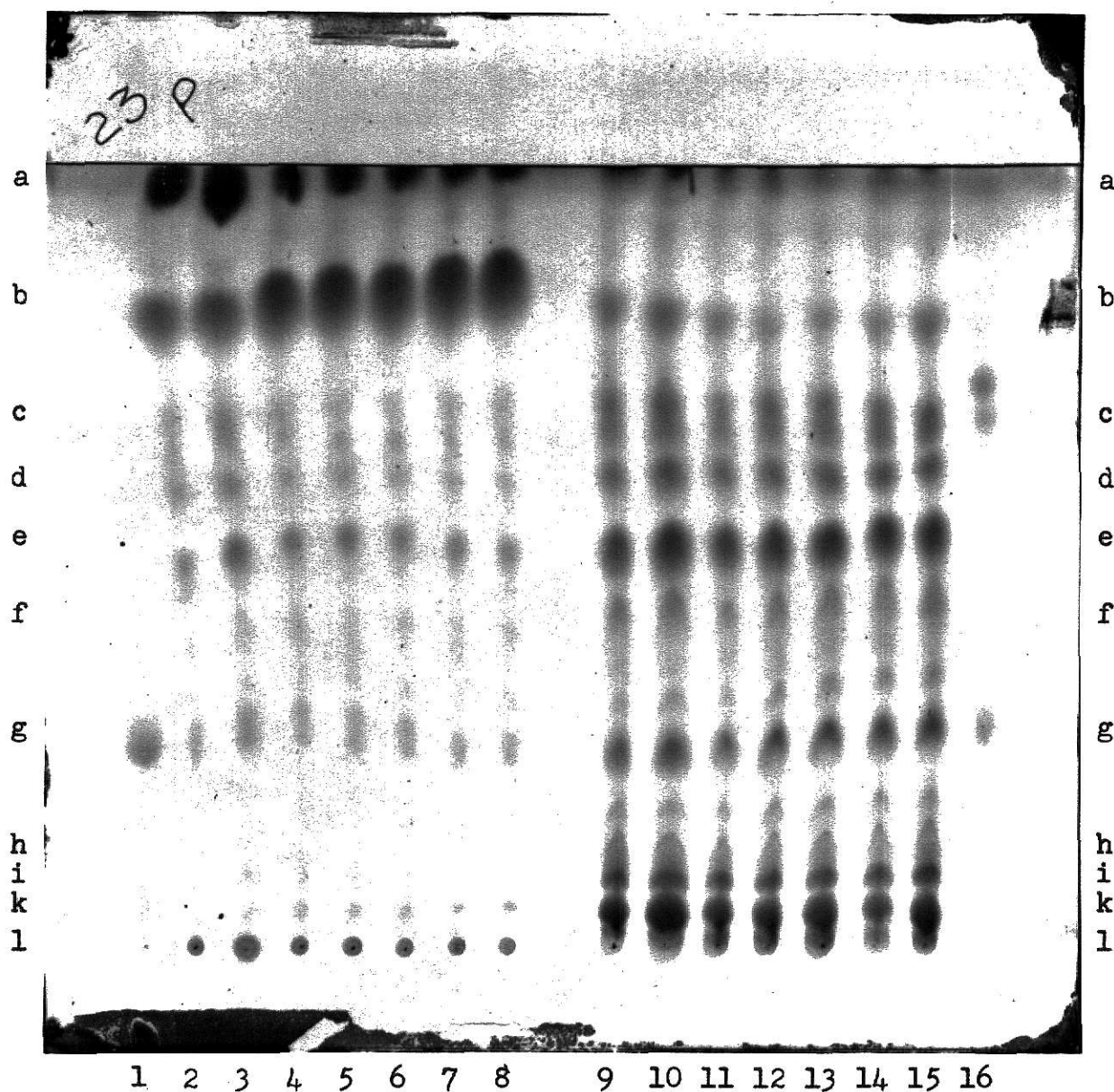


Fig. 18. Comparing polar lipid content of Jaycee oats, groats, original and air-classified flours.

From left to right: 1) reference mixture containing: lecithin (g) and lysolecithin (i); 2-15) as in Fig. 17, 16) reference mixture containing cerebrosides (c), lecithin (g), and lysolecithin (i). Tentatively identified as in Fig. 12.

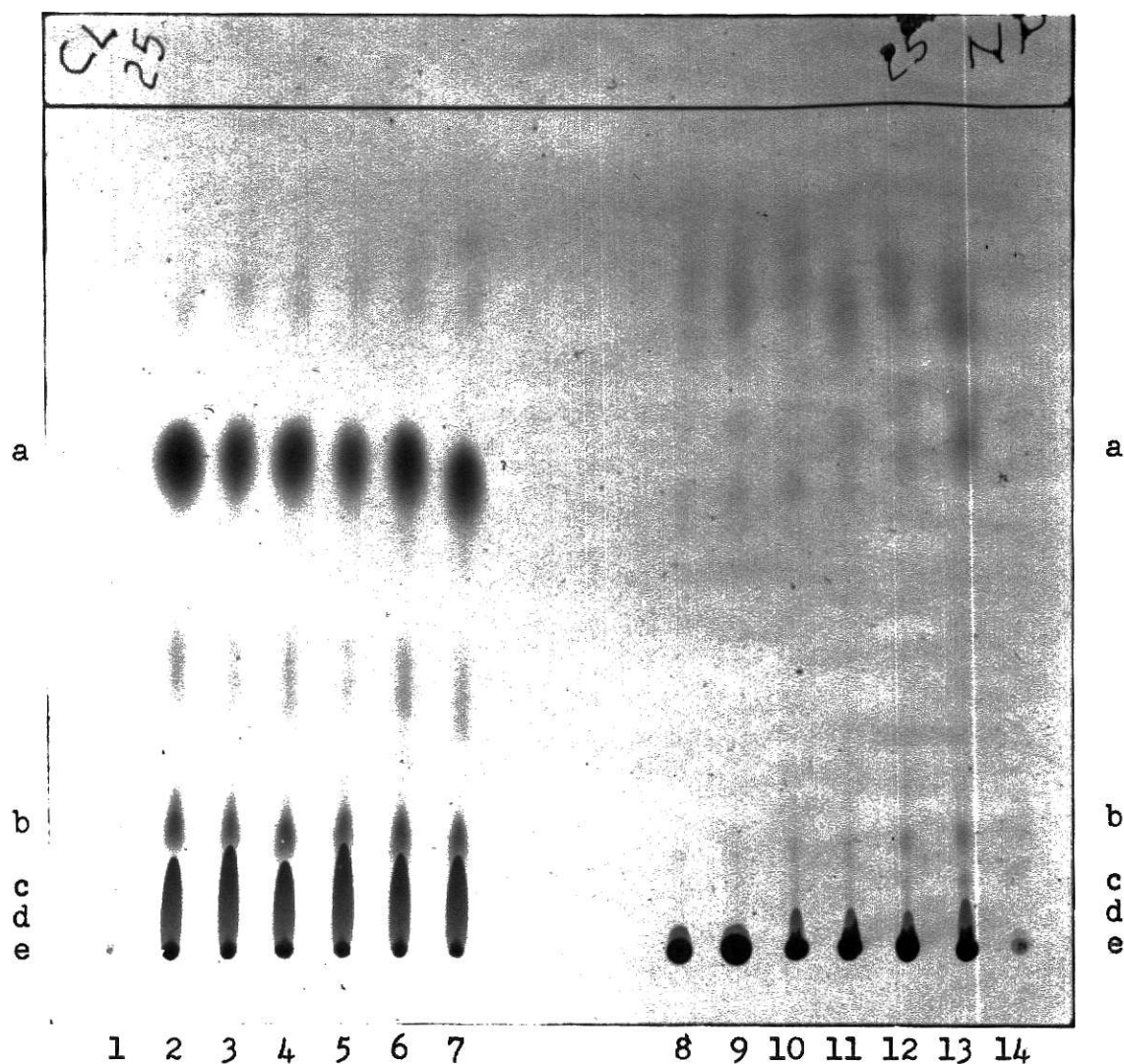


Fig. 19. Comparing non polar lipids of original and air-classified Clintford flours.

From left to right: 1) reference mixture as in 1) Fig. 9, 2-7) original and B, C, D, E, EE flours, extracted with petroleum ether; 8-13) the same samples extracted with water saturated butanol following petroleum ether. 14) as 1). Tentatively identified as in Fig. 9.

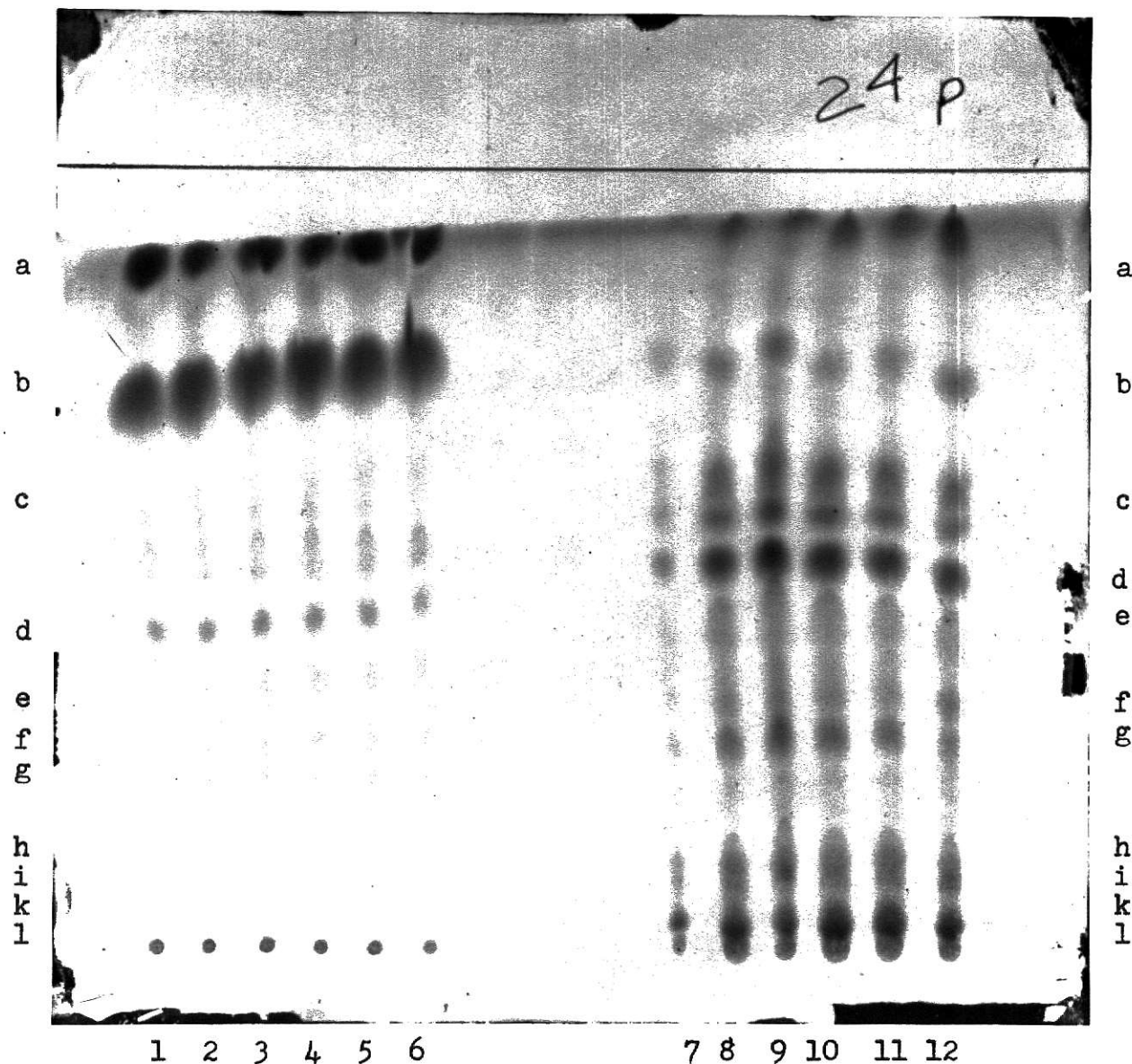


Fig. 20. Comparing polar lipids of Clintford original and air-classified flours.

From left to right: 1-6) original and air-classified fractions in the usual sequence, extracted with petroleum ether; 7-12) the same samples in the same sequence extracted with water saturated butanol as in Fig. 19. Tentatively identified as in Fig. 12.

SUMMARY AND CONCLUSIONS

Four oat varieties from the 1968 crop were investigated for their response to air classification and the study of their lipids.

The gross composition of oats, groats, and their air-classified fractions was determined by the usual AOAC methods. The protein content was determined also by a dye-binding technique. The correlation between the data obtained by this technique and the Kjeldahl values was highly significant when the determinations were performed on samples ground according to the equipment manufacturers' prescriptions ($r = 0.96$).

The method turned out to be unsuitable for air-classified flours (mainly the fine fractions).

Lipid extraction was performed in two steps with solvents of extreme polarities.

The first extraction with petroleum ether as the non polar solvent (Skelly F, b.p. 35-60°) removed the free lipids. The subsequent extraction was performed with water-saturated butanol - an extremely polar solvent system - to remove the bound lipids. The lipids both "free" and "Bound" were characterized by thin-layer adsorption chromatography.

No significant correlation between free lipids and protein content was found; however, a rather high correlation ($r = 0.90$) between bound lipids and protein level was observed with the regression line passing through the origin.

No correlation between free and bound lipids was found.

TLC showed triglycerides as the predominate compound in the petroleum ether extract, accompanied by small amounts of 1,3- and 1,2-diglycerides. Traces of free fatty acids and some plant sterols also were detected. Polar compounds were present in both the free-lipid extract and the W.S.B. (Water saturated butanol) following petroleum ether extracts where their amount was substantially higher.

Tentatively identified glycolipids were traces of monogalactogyl glycerides; furthermore, a type of plant cerebro-sides and considerable amounts of digalactogyl glycerides were detected in the water saturated butanol extract. The fraction corresponding to the monogalactogyl glycerides in the petroleum ether extract was -naphtol negative which seems to indicate that triglycerides were the main compound at this R_f and amount.

Phosphatidyl ethanol amines, lecithins, phosphatidyl serines, and lysolecithins were tentatively identified among the phospholipids. The Clintford variety seemed to reveal more additional phosphor-containing lipid compounds than the other varieties. No substantial qualitative differences were observed among lipid classes obtained from the oats, oat groats, or their air-classified flours.

During air classification, the protein shift towards the fine fractions was much lower than in other cereal grains. The lipids (mainly the bound fraction) seemed to be shifted along with the proteins (total lipids vs. protein $r = 0.5^{xx}$).

The results seem to indicate that the response of oat flours to air fractionation is not particularly favorable. This phenomenon seems to be due to the high lipid content of the oats endosperm, which leads to a certain degree of stickiness of these flours, and hence to difficulties in their air handling.

SUGGESTIONS FOR FUTURE WORK

The lipids in the oat grain constitute one of their main attributes through their high content in linoleic acid which is essential for the human diet. Their content in a high number of polar compounds, as revealed by this first attempt of adsorption TLC, presents a wide field for future investigation. More details about the composition of these lipids can be obtained by a reverse phase partition TLC and finally by two-dimensional chromatography. Such informations may lead to the eventual manufacturing of new food products hence increase the usefulness of this cereal.

Oats have the best balanced protein among all cereal grains. The 25% protein containing species should induce plant breeders to work on the transfer of the high protein from the wild oats to the cultivated types. Such a jump in protein level would make oats an even better food for children and adults - which might be important to a protein demanding world.

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A STUDY OF LIPIDS OF OAT VARIETIES

by

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This study deals with the determination of free and bound lipids of four oat varieties, of their groats and their air-classified flours, and with the characterization of the lipids by thin-layer adsorption chromatography. Furthermore, it was an attempt to examine the response of oat flours to air-classification by following the eventual shifts occurring in the gross composition of the flour fractions compared to the original flours.

Lipid extraction was performed in two steps with solvents of extreme polarities. The first extraction with petroleum ether (a non polar solvent) (Skelly F, b.p. 35-60°) removed the free lipids. The subsequent extraction was made with water-saturated butanol (a highly polar solvent system) to remove the bound lipids. Both free and bound lipids were characterized by TLC. Triglycerides were found to be the predominate compound in the free lipids fraction, accompanied by small amounts of 1,3- and 1,2-diglycerides. Traces of free fatty acids and the presence of some free plant sterols also were detected among the non polar compounds.

Polar compounds were present in the free lipid fraction; however, their amount was substantially higher in the bound lipids extract. Tentatively identified glycolipids were monogalactosyl glycerides; furthermore, a type of plant cerebro-sides and considerable amounts of digalactosyl diglycerides were detected. The presence of phosphatidyl ethanolamines, lecithins, phosphatidyl serines, and lysolecithins was

tentatively identified among the phospholipids. No qualitative differences were observed between lipid classes obtained from the oats, groats, or air-classified flours.

Ash, moisture, crude fiber, and proteins were determined by the usual AOAC methods. Proteins also were determined by a dye-binding method. The results of this technique were highly correlated ($r = 0.96$) with the Kjeldahl data, when performed on samples prepared according to manufacturers' prescriptions. It turned out to be unsuitable for air-classified flours.

The protein shift towards the five fractions of the air-classified flours was much lower when compared to other cereal grains. No significant correlation was found between ash and protein, nor between ash and particle size. The protein shift seemed to be accompanied by a lipid shift. A highly positive correlation ($r = 0.90$) between bound lipids and protein level was observed with the regression line passing through the origin. No correlation between free and bound lipids was found. The protein and lipid levels of the "as milled" and residual flours were comparable except in the Jaycee variety which was a particularly high lipid-containing species. This variety had lipids and proteins concentrated in the residual fraction. The starchy fraction was lowest in protein and lipid content.