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SOLVENT-EXTRACTED RAPESEED OIL MEAL AS A PROTEIN SOURCE
FOR PIGS AND RATS

by

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A THESIS

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ABSTRACT

Four experiments were designed to study the addition of Polish-type solvent-extracted rapeseed oil meal to diets for pigs and rats. Rapeseed oil meal was incorporated into the rations at three different levels on an equivalent protein basis at the expense of soybean oil meal; one diet high in rapeseed oil meal was supplied with additional supplemental lysine.

Pigs and rats had decreased rates of gain and lowered efficiencies of food utilization in the starter and grower periods as the levels of rapeseed oil meal in the diets increased. During the finisher period no ration effect was evident with either species. Individually-fed pigs were less affected than group-fed animals, probably because of a lower feed intake, and thus a lower goitrogen intake per unit of body weight, by the former animals.

No significant ration effects were observed on carcass characteristics of pigs at any level of rapeseed oil meal. Pigs did tend to have longer carcasses and smaller areas of loin as the percentage of rapeseed oil meal increased.

Female rats conceived normally but fetal resorptions and stillbirths were prevalent on the rapeseed oil meal diets. There was evidence of impaired conception in gilts fed high levels of rapeseed oil meal. Liveability was poor and weaning weights were low with both pigs and rats when rapeseed oil meal was a dietary constituent. These results suggest that rapeseed oil meal should be used judiciously in reproduction rations or in rations for pigs to be kept for breeding purposes.

Digestibility coefficients for nitrogen, energy and dry matter tended to decrease as the levels of rapeseed oil meal increased, although

this effect was significant only with rats. The addition of lysine to the experimental diets may have been responsible for improved digestibility coefficients with pigs but the rat data did not confirm this observation. Nitrogen retention was significantly lowered when high levels of rapeseed oil meal were fed to rats.

Blood serum protein-bound iodine and standard metabolic rates were used as criteria of thyroidal activity. Rat serum protein-bound iodine levels were not affected by the rapeseed oil meal goitrogen although a significant reduction in protein-bound iodine was noted with pigs fed high levels of rapeseed oil meal. Standard metabolic rates in 5 week old rats were reduced by the high levels of rapeseed oil meal feeding.

Thyroid gland hypertrophy and histological disturbances were evident with rats at 5 and 6 weeks of age on the high rapeseed oil meal diets. When rats were 12 weeks of age however, the glands seemed to be functioning normally. Weights and histological sections of thyroid glands of pigs at market weight or older did not indicate serious malfunction. Adrenals and livers from rats at 12 weeks of age and following reproduction showed no gross abnormalities. Livers from swine on high levels of rapeseed oil meal showed a moderate degree of hypertrophy.

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INTRODUCTION

Rapeseed production in Western Canada has enjoyed a dramatic production increase during the past several years and now ranks as an important farm industry. Accelerated foreign demands have been partially responsible for the increased production but large quantities of rapeseed are processed in Canada, primarily as a source of oil.

A byproduct of the extraction process is rapeseed oil meal¹ which according to present specifications of the Canadian Feeding Stuffs Act and Regulations (1952) cannot be incorporated into commercial rations for breeding stock or into starting rations for pigs. R.O.M. may be used as only 25% of the total supplemental protein for other classes of pigs.

Recorded information on the nutritional value of R.O.M. for various species of livestock is not extensive, although the scientific literature is replete with evidence that members of the genus Brassica have a marked goitrogenic effect. Morrison (1959) estimated the protein digestibility coefficient of R.O.M. to be 85% but unpalatability and amino acid deficiencies of the meal have been reported. These factors, coupled with the aforementioned thyrotoxicity, have cast some doubt upon extensive usage of this material as a livestock feed. In view of the economic importance of rapeseed production to growers and oil seed processors, and the possibility of a relatively inexpensive home-grown protein supplement for livestock, considerably

¹In the interests of brevity R.O.M. is subsequently used to designate the byproduct obtained during the extraction of oil from various kinds of rapeseed. Polish-type rapeseed oil meal (Brassica campestris L.) was used in the current study.

more information should be obtained concerning the use and limitations of this byproduct.

In Canadian studies of R.O.M. up to 1961, a meal produced by the expeller process was tested. Now that a solvent-extracted R.O.M. has become available, the need to reevaluate the use of this meal for pigs has arisen. Complementary studies with rats were carried out to substantiate the data obtained from the swine experiments.

REVIEW OF LITERATURE

Bell (1955) in his excellent review article concerning the nutritive value of R.O.M. indicated that the first North American report on the feeding value of this byproduct of the vegetable oil industry appeared in 1944. In Germany, however, Morrison (1959) states that "it has long been used for stock" although it must be "fed carefully to avoid injurious results". Bell (1955) reported that published German experiments on the subject date back to 1872 although the most conclusive studies regarding the chemical composition and nature of the goitrogenic factors present in R.O.M. have appeared in the last two decades.

A. Presence of a Toxic¹ Factor (or Factors)

Bell (1955) referred to the identification by Sjollem in 1901 of crotonylisothiocyanate as a constituent of the essential oil fraction of rapeseed. In 1920 Viehove et al. (cited by Bell 1955) found crotonyl and allylisothiocyanate to be present in rape and mustard seed and estimated their relative toxicities with rabbits. Both Astwood et al. (1949) and Carrol (1949) isolated a goitrogenic substance from Brassica seeds, including rapeseed. This substance was later proved to be L-5-vinyl-2-thiooxazolidone and was synthesized in 1950.

In 1949 Matet et al. (cited by Bell 1955) isolated the glucosides sinigrin and gluconapin from rapeseed; these substances being precursors to allyl and crotonyl isothiocyanates. Allylcarbinyl isothiocyanate (3-butenyl isothiocyanate) is the main mustard oil in

¹The term "toxic" as used in this manuscript means production of varying degrees of abnormality with respect to body growth, thyroid size and/or its histology and several other disturbances of tissues or functions.

R.O.M. and Pitt-Rivers (1950) suggested that this compound might be converted by oxidation, possibly with the aid of an enzyme, to L-5-vinyl-2-thiooxazolidone.

Obvious hypertrophy of thyroid glands of rats fed rapeseed was reported by Kennedy and Purves (1941). Pettit et al. (1944) observed that while 14% R.O.M. in chick starters was satisfactory, a level of 20% resulted in a high mortality rate. Blakely and Anderson (1948) reported hyperplastic goitres in turkey poultts fed 20% R.O.M. and Bell (1955) states that Turner found marked goitre and growth depression in chicks fed diets containing up to 40% R.O.M. Dow and Allen (1952) reported thyroid hyperplasia but this was accompanied by accelerated body weight gain and improved efficiency of feed utilization with young chicks fed R.O.M. at levels as high as 25% of the total diet.

Bell and Williams (1953) working with mice, observed growth depression when various levels of R.O.M. were added to the diet. Hussar and Bowland (1959) fed R.O.M. at 2 and 10% levels of the ration to swine and rats and observed consistent reduction in liveweight gains at the 10% level although the results were quite variable at the 2% level. Histological studies revealed characteristic thyroid abnormalities thereby further confirming the goitrogenic action of the meal with these species.

Renner et al. (1955) observed that Argentine-type rapeseed exhibited a greater goitrogenic effect than the Polish-type when this meal was fed to chicks. Clandinin (1959) confirmed these findings with chicks although Bell (1957) reported no difference between these two

varieties when fed to mice even though the Argentine type contained a higher level of goitrogen.

Clandinin et al. (1959) reported that isothiocyanate content was markedly affected by variety although no consistent effect of location of growth was observed. Thiooxazolidone content was not only affected by variety but also appeared to be influenced by the environmental conditions under which the seed was grown.

Bell et al.¹ (1960) found that as well as having widely divergent absolute quantities of isothiocyanate and thiooxazolidone, the relative proportions of each in rapeseed varied according to variety, season and location. Using purified preparations of these two compounds Bell et al. (1960) found them to be about equal in toxicity in mice. Levels of 0.2% of isothiocyanate or thiooxazolidone caused some mortality while levels of 0.08% resulted in severe growth depression. These results, coupled with the in vitro conversion of thiocyanate to the thiooxazolidone demonstrated by Clandinin et al. (1959), tend to confirm Pitt-Rivers suggestion that this transformation can also occur in vivo.

Evidence has also been accumulated by another route to verify the presence of a toxic factor(s) in R.O.M. Allen and Dow (1952), following the report of Astwood et al. (1949) that the active principle was water soluble, noted a marked decrease in toxicity when the R.O.M. was fed to chicks subsequent to hot water extraction. These workers (1954) found that any residual toxicity could be alleviated by iodine

¹Private communication. A summary of these results are given in the Minutes of the Associate Committee on Animal Nutrition.

supplementation and therefore attributed it to thiocyanate. Clandinin et al. (1959) and Goering et al. (1960) have found enzymatic extraction procedures effective in removing much of the toxic effect of R.O.M. thereby further substantiating the already convincing evidence suggesting the presence of a toxic goitrogenic factor.

B. Mode of Action of the Toxic Factor (or Factors)

Probably the first knowledgable proposal as to the action of the goitrogen of the Brassica genus came from Marine in 1929. Greer (1950) reports that Marine believed that the goitrogen exhausted the thyroxine content of the thyroidal cells which led to hyperplasia; the immediate cause for goitre development was considered to be a relative lack of iodine. Furthermore it was believed that certain oxidation-reduction systems were poisoned and this created a demand for greater hormone production by the gland.

In studies with supplemental iodine, Marine et al. (1930) showed that this element fed to rabbits effectively counteracted the goitrogen from cabbage. Bell (1955) reports that Kennedy et al. and Purves found iodine to be only partially effective against the rapeseed factor in rats as oral iodide partially reduced gland hypertrophy but the hyperplastic condition persisted. Kratzer et al. (1959) reported that Protamone (an iodinated casein derivative containing about 1% thyroxine) reduced thyroid weight but not growth depression in chicks and poults. Bell and Baker (1957) and Bell (1957) using mice, found that Protamone and KI both failed to counteract the growth depressant factor of R.O.M. although Allen and Dow (1954) reported that iodine supplementation ostensibly precluded activity of the goitrogen.

Hussar and Bowland (1959) reported a decrease of over 300% in body weight to thyroid gland weight ratio when R.O.M. was fed at a 10% level to swine; histological studies also showed an increase in cellular components. These workers noted with rats that there was a decreased level of I^{131} per unit thyroid gland as the R.O.M. increased. This was taken to denote either a reduced uptake or more rapid turnover rate of I^{131} . The ratio of radioactive iodine in the thyroid gland to radioactive blood serum protein-bound iodine declined as the level of R.O.M. increased, thereby indicating that abnormalities and biological disturbances of sufficient magnitude to effect hormone biosynthesis had occurred.

Astwood (1949) concluded that the mechanism of goitrogenesis from rapeseed is similar to the sulphonamides and thioureas, that is, the primary effect is one of thyroid inhibition. This interference with thyroxine synthesis results in a concomitant increase in thyrotropin synthesis which in turn leads to hyperplasia.

Greep (1954) proposed that a possible mode of thyroidal interference by antithyroidal drugs is the blockage of protein iodination of amino acid derivatives in the gland. This hypothesis is substantiated by increased iodine content of the gland during such a medication.

Bell (1955) states that Benda showed that respiration in guinea pig liver slices was depressed 50% by 10^{-2} molar concentrations of allylthiocyanate and that Flichenstein and Berg demonstrated inhibition of dehydrogenation reactions involving pyruvic, lactic, citric and succinic acids by allylthiocyanate.

Tissue homogenates from rats injected with thyroxine or triiodothyronine have consistently shown elevated oxygen consumption (Reid and Kossa, 1954; Barker and Klitgaard, 1952; Boyd and Oliver, 1960.) Thiourea, however, included in tissue homogenates decreased oxygen consumption (Reid and Kossa, 1954). It is well known that hyperthyroid subjects have elevated basal metabolism and Pitt-Rivers and Tata (1959) have theorized that this effect may be due to the uncoupling of oxidation from phosphorylation. Thyroxine has been reported to uncouple oxidation from phosphorylation by Tapley et al. (1955), Tapley and Cooper (1956) and others. This being so, one would expect a decrease in the P/O ratio (number $\curvearrowright$ PO₄ bonds: O₂ consumed) and a resultant increased basal metabolic rate attributable to inefficient trapping of energy. The mechanism of this uncoupling has been attributed to inhibition of the transhydrogenase reaction involving diphosphopyridine nucleotide and triphosphopyridine nucleotide by Solomon and Dowling (1960). Tapley (1956) reported liver mitochondrial swelling in hyperthyroid rats and felt that uncoupling of oxidative phosphorylation was due to morphological alterations of mitochondria.

Allen and Dow (1954) reported normal serum protein-bound iodine values of chicks fed R.O.M. Bell¹ (1957) using oxygen consumption as a measure of thyroid activity noted decreased oxygen consumption of mice fed R.O.M.

Although the specific action of the R.O.M. goitrogens is not known, it seems conceivable that their action could be elicited either

¹Private communication. A summary of these results is given in the Minutes of the Associate Committee on Animal Nutrition.

directly at the cellular level as Bell (1955) suggests and/or by acting indirectly through the thyroid gland.

C. Nutritional Value of Rapeseed Oil Meal

Morrison (1959) reported the average analysis of R.O.M. as 33.5% crude protein, 8.1% fat and 10.8% fibre. The crude protein digestibility coefficient of 85% reported by Morrison is within the range of 82-86 percent cited by Bell (1955). Studies to accurately appraise the nutritive value of R.O.M. are confounded to a greater or lesser extent by the presence of the goitrogenic factor(s) which puts an additional stress on the animal over and above any actual nutritional shortcomings of the meal.

Klain et al. (1956) reported that as the percentage of R.O.M. in the diet rose, chick growth decreased. This effect was ascribed to inadequacy in the amino acid, arginine. Bell (1955) compared amino acid analysis of R.O.M. with chick requirements and concluded that R.O.M., when used as the only source of protein would supply adequate levels of all essential amino acids except phenylalanine. Kratzer et al. (1954) and Dow and Allen (1954) noticed a growth response in chicks fed supplemental lysine. This contrasts with Bell's 1957 observation that additional lysine was ineffective for mice. Clandinin (1949) analysed samples of Argentine and Polish-type R.O.M. and reported that the protein of these meals contained 3.6 and 4.2% of lysine respectively. Clandinin et al. (1959) felt that a lysine deficiency probably was a contributing factor to depressed growth which resulted when chicks were fed R.O.M. and cautioned that excessive amounts

of heat employed during the extraction of oil from rapeseed adversely affects the nutritive value of the resulting R.O.M. by reducing the available lysine content.

Hussar and Bowland (1959) reported that a 10% level of R.O.M. depressed rate of gain and in some cases efficiency of feed utilization in swine and rats. Food consumption was not adversely affected in these experiments or in work with mice (Bell, 1953) although Bezeau et al. (1960) reported unpalatability with sheep when R.O.M. was fed at a level of 30% of the ration. Using an Argentine-type R.O.M. as 10% of the total ration for swine and rats Hussar and Bowland (1959) reported depressed apparent digestibility of dry matter, energy and nitrogen, with significant effects occurring only in rats. Retention of digestible nitrogen was unaltered. Bezeau et al. (1960) reported that at 10% of the ration R.O.M. was equal to linseed oil meal for ewes although at 30% R.O.M. there was a decrease in the protein and energy retained.

Bell (1955) reported that Burkitt et al. found that R.O.M. could be fed to beef cattle at levels up to 2 lb. per head daily with no obvious ill effects. Allen and Dow (1952) reported that steers will tolerate it in amounts as high as 25% of the grain component of the ration.

Asplund¹ (1961) found that dairy cows were not affected when R.O.M. was fed as 10% of the total dry matter intake, but at a 20% level, the cows showed a rapid decline in milk production. No deleterious effects on milk composition were noted. These observations tend

¹Private communications. A summary of these results is given in the Minutes of the Associate Committee on Animal Nutrition.

to suggest that ruminants are less susceptible to high levels of R.O.M. than are monogastrics.

Bell (1957) noted species and sex differences in response to R.O.M. Male mice appeared to be more severely affected than females. Hussar and Bowland (1959) noticed the reverse in rats and found that ovariectomy apparently reduced the increased susceptibility of the female rats.

Much of the previous work on the rapeseed oil meal problem has been carried out using an expeller-extracted product. Very limited information is published comparing the recently available solvent-extracted meal with the expeller meal. Goering et al. (1960) compared expeller and solvent-extracted R.O.M. subsequent to an enzymatic treatment designed to remove the toxic factors. Working with rats they showed expeller-extracted meal to be inferior. On comparing a mixture of 50% soybean oil meal and 50% expeller R.O.M. with 50% soybean oil meal and 50% solvent R.O.M., a 15% increase in growth and 10% increase in efficiency of feed utilization was observed in favour of solvent R.O.M. Bell¹ (1959) compared solvent and expeller-processed R.O.M. in the diets of mice and found no significant differences in gain as a result of the different methods of processing.

D. Carcass Characteristics

Bell and Baker (1957) fed a diet consisting of 17% R.O.M. to mice; carcass composition indicated that the males on the R.O.M. diet tended to have more water with less fat and protein than control animals. Females did not follow the same trend.

¹Private communications. A summary of these results is given in the Minutes of the Associate Committee on Animal Nutrition.

Allen and Dow (1954) reported no abnormal effects on carcass quality of chickens fed 17% R.O.M. Blakely et al. (1960) reported superior carcass scores from turkeys fed 23% whole rapeseed.

Thiouracil feeding has been suggested by Pipes et al. (1958) as a method to induce a hypothyroid condition in cattle thereby facilitating the fattening process. Bowland (1951) reported that thiouracil supplemented swine rations decreased backfat and increased area of loin in pig carcasses although these advantages were accompanied by a definite decrease in length of side. Hussar and Bowland (1959) reported a trend toward shorter carcasses from pigs receiving 10% R.O.M. although a reduced area of loin was the only characteristic that reached statistical significance.

E. Gestation and Lactation

Information is very limited in most species on the effect of R.O.M. on reproduction. One of the first reports was by Frolich and Haring (1937) who stated that "suckling sows can be fed daily 400 gm. dry rapeseed oil meal per sow without harm to the sow's health or growth of the young. The mustard oil of the rapeseed oil meal has no effect on breeding sows, but it is advisable to guard against digestive disturbances by supplying powdered charcoal".

Lucas et al. (1958) fed rations containing thiouracil and thyroprotein to gilts and observed a significant increase in embryonic mortality in the thiouracil lot after 25 days gestation. Thiouracil fed animals had an average gestation period 10 days longer than controls and farrowed only 3.3 as compared to 8.7 pigs per litter. Bell and

Weir (1952) reported that R.O.M. and linseed oil meal were equally good in maintaining gestation weights in ewes and improving the weight and vitality of lambs at birth as compared to a ration with no protein supplement. Bezeau et al. (1960) fed levels of R.O.M. up to 30% of the ration. Ewes had subnormal weight gains during the gestation period and lambs were lighter at birth and at 6 weeks of age than control lambs were.

Bell and Baker (1957) fed 30% R.O.M. to mice during gestation and lactation. They found that breeding appeared to be satisfactory and that the number of young per litter was not markedly reduced by the R.O.M. diet. Bell and Baker suggested, however, that at the 30% level of feeding, R.O.M. imposed sufficient additional stress during the lactation period to interfere with normal performance.

Kennedy and Purves (1941) fed whole rapeseed as 45% of the total ration and reported a delay in development of the ovaries of immature female rats to the extent that the animals would not become pregnant until the age of 5 to 7 months rather than the normal 2 to 3 months. Jones et al. (1946) reported that hypothyroidism had no apparent effect on male fertility in rats and that female rats fed thiouracil for more than 100 days were not sterile although they resorbed their litters. Krohn and White (1950, Maqsood (1952) and Parrott et al. (1960) observed that hypothyroid rats ovulated but fewer young were brought to term due to fetal resorption.

The thyroid hormones, although not essential for initiation or maintenance of lactation, may be important in regulating the level of

milk secretion. This conclusion is based primarily on studies showing that thyroidectomy reduces, but does not abolish, milk secretion while under suitable conditions the administration of thyroxine generally induces an increase in milk secretion (Reineke, 1946; Benson and Cowie, 1957; Blaxter, 1952). Grosvenor (1961) has reported that thyroid secretion rate obtained from primiparous lactating rats during the first 14 days of lactation was 130% more than obtained from non-lactating females of the same age. There appeared to be a definite correlation between thyroxine secretion rate and intensity of lactation. Thyroactive compounds have been tested as lactation stimulants for swine by Davis et al. (1959), Dudley et al. (1959), and Johnson et al. (1959). Johnson et al. (1959) reported increased growth rate of litters and attributed it to thyroid hormone. The other workers reported no response although all were unanimous in stating that the treated sows lost more weight during the lactation period.

EXPERIMENTAL

OBJECTIVE

The main objective of these experiments was to evaluate solvent-extracted Polish-type rapeseed oil meal fed at varying levels in the diets of swine and rats. Supplemental lysine was added in one ration to study the effect of additional lysine in diets high in R.O.M.

Specific experiments were designed to study:

- (a) Rate of gain, dry matter, energy and nitrogen digestibility, efficiency of feed utilization and reproductive performance of individually-fed pigs and rats.
- (b) Rate of gain, efficiency of feed utilization and carcass characteristics of group-fed pigs from weaning to market weight.
- (c) Serum protein-bound iodine levels of swine at market weight and of rats at 6 and 12 weeks of age.

METHODS AND PROCEDURES

A. Formulation of Experimental Rations

Since the primary objective of these experiments was to study the effect of varying levels of R.O.M. in the rations of swine and rats, the five rations used were kept as nearly identical as possible in overall composition particularly with respect to protein levels. Rapeseed oil meal was substituted at the expense of soybean oil meal (S.O.M.) and wheat. The substitution levels of R.O.M. are outlined below:

Ration No.

1	Basal, no R.O.M.
2	25% of S.O.M. protein equivalent replaced by R.O.M.
3	50% " " " " " " "
4	100% " " " " " " "
5	100% " " " " " " "

with 0.2% supplemental lysine.

The established procedure of changing the composition of the ration to meet the nutritional requirements of the pig at different physiological stages was adhered to; these periods being starter, grower, finisher and reproduction (Table 1).

Table 1 shows the composition of the experimental rations. Table 2 shows the percentage of R.O.M. in the total ration. Table 3 gives a comparison of the two batches of R.O.M. used in so far as isothiocyanate, thiooxazolidone, total protein and lysine content are concerned. Batch 2 was much lower in thiooxazolidone and lower in isothiocyanate than batch 1.

The rats used in these studies were also fed the rations shown in Table 1. The starter period was 2 weeks in length, the grower period 3 weeks and the finisher period 4 weeks. The reproduction ration was fed from the time of breeding (12 weeks of age) until the end of lactation.

The feeds were ground and mixed at the University Livestock Farm, bagged in approximately 100 lb. lots and stored in a dry unheated shed. To prevent the rats from sorting their food, diets for this species were further ground through a Number 1B screen in a Model D Comminuting Machine¹. The rat diets were kept in sealed glass jars in the rat room of the Animal Science Laboratory.

B. Methods and Procedures in Swine Experiments

The swine experiments were divided into two separate trials. The first batch of R.O.M. (Table 3) was fed throughout Experiment 356A and until mid-to-late gestation in Experiment 356, depending on how early the gilts were bred. During the remaining portion of the reproduction

¹ Model D. Comminuting Machine manufactured by the W.J. Fitzpatrick Company, Chicago, U.S.A.

Table 1.

Formulation and Composition of Basal Rations for Lot 1 in Rapeseed Oil Meal Experiments

Ration - Period Fed -	Starter 15-50 lb.	Grower 50-110 lb.	Finisher 110-200 lb. (210 for gilts)	Reproduction 210 lb. +
<u>Ingredients</u>				
Wheat	72.45	25.0	25.0	15.0
Barley	--	46.7	36.9	29.9
Oats	--	10.0	25.0	40.0
Sucrose	10.0	--	--	--
Soybean oil meal ¹ (44%)	13.0	13.0	8.0	10.0
Fishmeal	3.0	2.0	2.0	2.0
Alfalfa meal (dehy.)	--	2.0	2.0	2.0
Iodized salt	0.5	0.5	0.5	0.5
Ground limestone	0.5	0.5	0.5	0.5
Zinc sulfate	0.05	0.05	0.05	--
Vitamins A and D ₂	+	+	+	+ 2.5 times
Vitamin B-complex ² supplement	0.3	0.15	--	--
Antibiotic supplement ³	0.2	0.1	0.05	--

Ration Composition by Analysis

<u>Ration</u>				
1 - Protein %	17.0	15.4	16.1	17.4
Combustion energy-kcal./gm.	4.06	3.99	3.97	4.07
2 - Protein %	17.2	16.6	17.2	16.6
Combustion energy-kcal./gm.	4.05	4.11	3.94	4.02
3 - Protein %	17.3	16.6	18.0	16.5
Combustion energy-kcal./gm.	4.03	4.04	3.96	4.03
4 - Protein %	17.2	16.6	16.8	16.7
Combustion energy-kcal./gm.	3.97	4.12	3.69	4.06
5 - Protein %	17.0	16.6	16.4	16.3
Combustion energy-kcal./gm.	3.98	4.01	3.88	4.05

¹ Rapeseed oil meal substituted on an equivalent protein basis at the expense of soybean oil meal and wheat. Soybean oil meal and/or R.O.M. represented 73% of the total supplemental protein in the starter ration, 76% in the grower ration and 66% in the finisher ration.

² Each pound contains: riboflavin, 1 gm.; calcium pantothenate, 2 gm.; niacin, 4.5 gm.; choline chloride, 5 gm.; folic acid, 30 mg.; B₁₂, 4.5 mg.

³ Equal parts TM-10 and Aurofac-10.

Table 2.

Rapeseed Oil Meal as a Percent of the Total Ration

	Ration				
	1	2	3	4	5
Starter	0.0	3.9	7.8	15.6	15.6
Grower	0.0	3.9	7.8	15.6	15.6
Finisher	0.0	2.4	4.8	9.6	9.6
Reproduction	0.0	3.0	6.0	12.0	12.0

Table 3.

Comparison of the Two Batches¹ of Rapeseed Oil Meal Used in Experiments

	Isothiocyanate Content mg./gm.	Thiooxazolidone Content mg./gm.	Protein %	Lysine as % of Protein in the Meal
Batch 1	3.51	6.33	37.9	5.15
Batch 2	2.45	1.95	35.7	5.46

¹ Isothiocyanate and thiooxazolidone contents supplied through the courtesy of Dr. J. M. Bell, Department of Animal Science, University of Saskatchewan, Saskatoon.

period in Experiment 356 the second batch of R.O.M. was used.

Experiment 356. Twenty gilts and 10 boars were individually-fed and continued on experiment until termination of the reproduction period. Information pertaining to rate of gain, efficiency of feed utilization, energy, dry matter and nitrogen digestibility, litter sizes and weights and other reproduction data were obtained.

Experiment 356A. Forty pigs were fed ad libitum in groups of 4 pigs per lot replicated so that 8 pigs were on each ration treatment. Rate of gain, efficiency of feed utilization and carcass characteristics

were studied. Thyroid glands and livers were taken for morphological studies and serum protein-bound iodine levels were determined.

(a) Growth Experiments

1. Individual-feeding trials

Experiment 356 was initiated on April 19, 1961 and terminated on June 16, 1962. Thirty pigs, 5 gilts each from 4 litters and 5 boars each from 2 litters averaging 5 weeks of age and 16 pounds in weight were balanced with respect to littermates and assigned to the 5 rations.

All pigs were individually-fed for a period of 1 hour twice daily beginning at 8 a.m. and 4 p.m. and ran together in groups when not being fed. Water was supplied in automatic watering bowls and was available except during the feeding periods. At about 120 days of age the boars were separated from the gilts and maintained in separate pens. The animals were weighed weekly and individual feed consumption data recorded.

2. Group-feeding trials

Experiment 356A was initiated on April 28, 1961, and terminated on October 4, 1961. Twenty castrated males and 15 females of Landrace-Yorkshire breeding and 5 gilts of Yorkshire breeding averaging 20 pounds and 6 weeks of age were allotted to the 5 ration treatments. Uniform distribution on the basis of littermates was made in so far as this was possible. The animals were housed in groups of 4 and were fed ad libitum.

Pig weights were recorded at weekly intervals and at the first weekly weighing after a pig reached 195 lb. liveweight it was shipped to market. Following slaughter, Canadian Government carcass grades and Record of Performance (National Bacon Hog Policy, 1959) measurements and scores were obtained.

In both experiments pigs were ear notched at birth for identification

purposes and were routinely treated for anemia. Ascarids were controlled by treatment with a cadmium oxide compound prior to placing the pigs on test.

(b) Reproduction

Upon reaching 210 lb. liveweight the animals in Experiment 356 were transferred to the reproduction ration at a restricted intake of 6 lb. per day. A week prior to being bred at 240 days of age the females were flushed by being offered 7.2 lb. of feed per day although often the extra feed was refused.

Breeding was carried out according to the schedule shown in Table 4.

Table 4.

Breeding Schedule for Gilts and Boars, Experiment 356

<u>Lot</u>		<u>Boars Used For Breeding</u>	
		<u>1st and 2nd</u> Mating	<u>3rd and 4th</u> Mating
1	2 gilts mated to:	lot 1	5
	2 gilts mated to:	lot 5	1
2	2 gilts mated to:	lot 2	4
	2 gilts mated to:	lot 4	2
3	2 gilts mated to:	lot 3	2
	2 gilts mated to:	lot 2	3
4	2 gilts mated to:	lot 4	3
	2 gilts mated to:	lot 3	4
5	2 gilts mated to:	lot 5	1
	2 gilts mated to:	lot 1	5

In an instance where a gilt did not settle on first service, she was then bred to the other boar of the same lot. Any further matings were made to boars of the opposite lot. If after the fourth mating the

gilt had not conceived she was marketed and the reproductive tract examined. Approximately 1 week prior to farrowing the sows were placed in farrowing pens and 1 lb. of ration was replaced by 1 lb. of bran. Normal precautionary measures were followed during the farrowing period. During the lactation period of 5 weeks, sows received a basic allowance of 6 lb. per day and an additional 0.75 lb. for each suckling pig. Litter sizes, birth weights and livability records were taken.

(c) Digestibility and Retention Studies and Methods of Analysis

Two gilts and 1 boar from each of the 5 lots were used for digestibility studies which were done by the chromic oxide indicator method. Moore (1958) used chromic oxide for determining digestibility coefficients for swine and found it to be comparable to a total collection technique. A similar technique to the one used in the current study has previously been employed in this Department by Plank (1961) and gave satisfactory results while being considerably less work than the total collection method.

The normal ration with 1% Cr_2O_3 added was fed for a 4 day acclimatization period after which time fecal collections of 100 grams were made at 8 a.m. and 4 p.m. each day for 3 days. The 4 periods of collection were at approximately 75 and 150 lb. during the grower and finisher periods respectively, and at 90 and 21 days of gestation and lactation respectively. In order to have a comparison between sexes, digestibility data was collected from the boars at 340 days of age, this time approximating the 90th day of gestation with the gilts. The fecal samples were dried for 12 hours at 105°C . and ground to pass through a No. 20 mesh screen in a small Wiley Mill¹. A suitable

¹ Wiley Mill No. 4276 sold by A.H. Thomas Company, Philadelphia, Pa, U.S.A.

aliquot of the resulting sample was sealed in a glass jar at laboratory temperatures until analysed for nitrogen, energy and Cr_2O_3 .

The Kjeldahl-Gunning method of analysis using boric acid to retain the ammonia was used for nitrogen determination of feeds, feces and urine in both the swine and rat studies. Fecal and feed samples were analyzed in the Parr Oxygen Bomb Calorimeter¹ to obtain caloric values.

Chromic oxide determinations were carried out by the method of Hill and Anderson (1958) as modified by Renner² (1962). Absorbence was read at 450 mu using the Beckman D.U. Spectrophotometer³.

(d) Blood Serum Protein-Bound Iodine⁴

On the week when the pigs in Experiment 356A reached 195 pounds a sample of blood was withdrawn from the anterior vena cava (Carle and Dewhirst, 1942) using a 50 ml. syringe and a 14 gauge, 2 inch needle. The blood sample was placed in a test tube and allowed to stand at 10° C. until the clot had retracted. After centrifuging at a speed of 1500 R.P.M., 15 ml. of serum was withdrawn and stored at 10° C. until analysis. The method of analysis was similar to the one used by Brown (1953) with the following modifications. Since it was often noticed that complete combustion of carbon was not being achieved in the ashing step, the ashed samples were taken up in 1 ml. of distilled water, dried and re-ashed for 10 minutes at 600° C. A similar procedure has been reported by Foss et al. (1960) except that these workers used a second ashing period of 30 minutes. The possibility of removing the carbon by filtration through a fine glass wool pad was investigated and was found to be

¹ Parr Instrument Company, Moline, Ind., temperature changes recorded on a Brown Elektronik Recorder manufactured by Minneapolis Honeywell Regulator Company, Philadelphia, Pa.

² Private communication. Twenty ml. conc. H_2SO_4 following digestion.

³ Beckman, Model D.U. Spectrophotometer manufactured by Beckman Instruments Incorporated, Fullerton, California.

⁴ Subsequent references to protein-bound iodine will be abbreviated to P.B.I.

advantageous because of increased speed. However, because filtration represented a possible source of iodine contamination, the second ashing was ultimately chosen as a means of eliminating residual carbon, thereby facilitating subsequent steps of the analysis.

Following the addition of ceric ammonium sulfate to arsenious acid a fifteen minute reaction period was employed. Samples were read in an Evelyn Photoelectric Colorimeter¹ at half minute intervals. A 420 millimicron blue filter was used for all readings. Each sample was done in duplicate, each duplicate being split into 2 equal aliquots for the final redox reaction. This gave a total of 4 readings per sample. If satisfactory checks were not obtained additional duplicate samples were analysed.

(e) Organ and Tissue Studies

Thyroid glands, livers, adrenals and ovaries were collected at the Swift Canadian Packing Plant shortly after the animals from Experiment 356 and 356A had been killed by severing the jugular vein. The glands with the exception of livers were sealed in small glass jars and were taken immediately to the Animal Science Research Laboratory. Thyroid glands and livers were weighed and random portions of ovaries, adrenals and thyroids were placed in Bouins² Solution. Treatment of the tissues and preparation of histological sections was similar to the methods used by Hussar (1958).

C. Methods and Procedures in Rat Experiments

The rat trials were set up as replications of the swine studies

¹ Made by Rubicon Company, Philadelphia, Pa.

² Bouins Solution: 75 parts saturated solution of picric acid, 25 parts formalin, 5 parts acetic acid.

with the hope that any information so gained would be of value in interpreting the swine data, as well as in showing species differences.

For these studies, 21 day old weanling rats of the Sprague-Dawley strain were used. The R.O.M. was obtained in 2 batches and since variability in quality of the meal seemed possible 1 batch was used in Experiment 1A, the second in Experiment 1B. The pertinent data on composition of the 2 meals appears in Table 3.

(a) Growth Experiments

1. Experiment 1A

Since 80 weanling rats were required for this experiment and this number could not be obtained at any one time, the rats were placed on trial in smaller balanced groups as they became available between June 9 and August 8, 1961. Each of the 8 groups of 10 rats was equalized for sex, the groups being designated by the letters A to H assigned in chronological order. Groups A and B and groups F and G remained intact and were used for reproduction studies. Five rats of each of the other groups were killed at the end of 2 weeks on trial; the remaining males and females were killed at the end of the finisher period.

Following the method described by Sibbald (1957), the weanling rats were fed ad libitum the experimental diets to which they had been allotted. Food consumption and rat weights were recorded at the time of weaning and at 7 day intervals until the end of the trial.

2. Experiment 1B

Forty rats were placed on experiment in April 1962. Rats were allotted to the experimental rations in groups of 10 as in Experiment 1A. The animals were fed ad libitum, 2 rats of the same sex per cage. Individual weights were recorded at time of weaning and at 7 day

intervals until the end of the experiment. Ten rats were killed at 6 weeks of age and another 10 at 7 weeks of age; the remaining 20 were maintained on experiment until killed at the end of the finisher period.

(b) Reproduction

Forty rats, 20 of each sex were transferred to the reproduction ration at the end of their ninth week on trial in Experiment 1A and were kept for breeding purposes. The breeding plan was similar to the one used with pigs and outlined in Table 4 except, rather than having 1 male and 2 females per lot, there were equal numbers of each sex.

(c) Digestibility and Retention Studies

After a ration acclimatization period of 7 days all 80 rats in Experiment 1A were placed in metabolism cages. Feed consumption and weight gains were recorded at the beginning and end of the 1 week period.

On approximately the eighteenth day of gestation 2 females from each lot were placed in the metabolism cages for 24 hours. A similar 24 hour metabolism period was planned originally for the fourteenth day of lactation but because of lactation failure in the rats this period was deleted.

Prior to the start of the metabolism trials the cages were washed and sprayed with a saturated solution of boric acid in 95% ethanol. The methods of collection of feces and urine were similar to those described by Likuski (1959). Methods of analysis for nitrogen and energy have previously been dealt with (Swine Experiments, Section (c)).

(d) Standard¹ Metabolic Rates

After 17 to 20 days on trial standard metabolic rates were measured with 40 rats from Experiment 1A, thereby giving a total of 8 rats per ration treatment. Equal numbers from each sex were used. The apparatus (Figure 1) was similar to that used by Bailey et al. (1957). Prior to being placed in the air tight chamber in the water bath the animals, weighing 100 ± 25 grams were fasted for approximately 12 hours. At the beginning of the experiment the system was flushed out with oxygen to provide an enriched atmosphere. An acclimatization period of 30 minutes was allowed in the water bath at 28 to 32° C. A desk lamp with a 30 watt bulb about 2 feet from the water bath was found to be helpful in quieting the animals and this was adopted as standard procedure. Twenty-five ml. of 10% NaOH was used to absorb exhaled carbon dioxide. Water was used in the manometer and the amount of distilled water required to equilibrate the arms in a given time period was taken to be the oxygen consumed by the rat. Final calculations were based on three 15 minute periods. The weight was adjusted to the 3/4 power of body weight as Brody (1945) and others have suggested.

Twenty rats from Experiment 1B were used for standard metabolic rate studies. The first group of 10 was treated as in Experiment 1A. The only difference with the second group was that instead of being 38 to 41 days of age they were 45 to 48 days old. It was hoped that by extending the period 1 week with the second group that information could

¹ The term is taken from Maynard and Loosli, 1962, Animal Nutrition, 5th Edition, McGraw-Hill Book Company, Inc., Toronto. In this study, standard metabolic rate means "The heat produced in a thermoneutral environment (28 to 32° C.) after a 12 hour fasting period."

be obtained suggesting when the most drastic goitrogenic effect occurred.

Figure 1. Apparatus for Standard Metabolic Rate Determinations

(e) Blood Serum Protein-Bound Iodine

Twenty of the 40 rats used in the standard metabolic rate determinations in Experiment 1A were allowed access to feed for 24 to 36 hours. It was felt that a recuperation period was necessary to allow the animals to readjust after the fasting period. The animals were then decapitated and blood taken for serum P.B.I. determinations. Another 20 rats were killed upon completion of the finisher period and blood was again collected for serum P.B.I.

Serum separation and P.B.I. determinations were done according to the previously outlined method (Swine Experiments, Section (d)).

(f) Organ and Tissue Studies

Certain organs of the rats from Experiment 1A were excised and used for glandular studies. Ovaries, testes and adrenal glands of the 6 week old rats were taken and preserved in Bouins solution. Ovaries, testes, adrenal glands and livers of the 12 week old rats were obtained.

Upon completion of the reproductive period the 20 males and 20 females used for this portion of the study were killed and adrenal glands and livers excised and weighed. A visual examination was made of the female reproductive tracts to detect gross abnormalities.

Subsequent to the standard metabolic rate determinations, the rats in Experiment 1B were killed and the thyroid glands removed, weighed and placed in Bouins solution. The remaining 20 rats, not involved in the standard metabolic rate studies, were killed at the age of 12 weeks.

All tissues except livers were examined histologically by a method similar to that reported by Hussar (1958).

D. Statistical Analysis

The statistical evaluation of the data was carried out using analysis of variance as described by Goulden (1960). Duncan's Multiple Range Test (1956) was used to evaluate differences between means.

In the discussion of the data in this manuscript non-significant means will be designated as such and other differences mentioned will be understood to be statistically significant at either the 5 or 1% level. No interactions occurred between sex and ration except those that are specifically referred to in the text.

RESULTS AND DISCUSSION

In the interests of brevity the dietary levels of R.O.M. in the current study will be referred to as 0, 25, 50, 100 and 100% + lysine. The reader is reminded that these are relative, not absolute values and denote only the per cent of soybean oil meal being replaced by R.O.M. in the ration (see Table 2). Levels of R.O.M. used in experiments referred to will be given as per cent of the total ration in so far as this is possible.

A. Rate of Gain, Food Consumption and Efficiency of Food Utilization

Because of overall similarity in design of experiments, the data from both species will be discussed simultaneously thereby facilitating between species comparisons. The results are presented in Tables 5 to 8 and Figures 2 and 3.

(a) Starter Period. - 15 - 55 lb. liveweight for pigs in Experiment 356 and 20 - 50 lb. in Experiment 356A; 3 to 5 weeks of age for rats.

Both species showed a reduction in rate of gain and, to a lesser degree, a depression in efficiency of food utilization at the 100% levels of R.O.M. supplementation (Tables 5 to 8). When one compares the 100% level of R.O.M. to the 0 and 25% levels, rate of gain was reduced 35% and 15% more food was required per unit of weight gain in rats while corresponding figures for the pigs were 20 and 8% respectively. This species difference could be explained on the basis of an extremely rapid rate of gain and synthesis of body protein in the rat at this early age. A malfunctioning thyroid gland would be expected to exert a maximum effect under such conditions. At the 50% level of R.O.M. supplementation in the ration, the results were quite variable but, in general, the performance of this group was inferior to the groups receiving 0 and 25%

R.O.M. when daily gain and efficiency of food utilization are the criteria of measurement.

The addition of supplemental lysine in lot 5 to the animals receiving 100% R.O.M. had no influence on the rate of gain or efficiency of food utilization. The lysine requirement of growing pigs has been estimated to be 0.65% of the total diet for 20 to 70 lb. pigs receiving 16% protein rations, (N.R.C. Nutrient Requirements, 1959). The basal starter ration contained 0.82% lysine while the 100% and 100% + lysine diets contained 0.78 and 0.86% lysine respectively. On the basis of the aforementioned requirements the ineffectiveness of the supplemental lysine is not unexpected, although Kratzer et al. (1954) and Clandinin et al. (1959) have reported R.O.M. to be low in lysine for growing chicks. Bell's (1957) results with mice are in agreement with the present findings.

Low food intake, presumably associated with lack of palatability of the diets occasionally occurred with rats fed on diets high in R.O.M. If the animals were not placed on the experimental diets immediately after weaning, excessive food wastage resulted although within a few days they accepted their food readily. Bowland (1957) noted that pigs prior to weaning found R.O.M. quite unpalatable even at low levels although Hussar and Bowland (1959) found that animals ate normal levels of rations containing R.O.M. after weaning provided no alternative feed was available. Average daily feed consumption of pigs (Tables 5 and 6) was not affected by R.O.M. in the present study.

Male rats grew faster than females and required less food per gram of gain. Boars and barrows grew slightly faster than gilts. These results with pigs are in agreement with Fredeen (1953), Charette (1961)

and others.

(b) Grower Period. - 55 - 110 lb. liveweight for pigs in Experiment 356 and 50 - 110 lb. in Experiment 356A; 5 to 8 weeks of age for rats.

Pigs fed ad libitum in Experiment 356A showed significant reductions in rate of gain on rations 3, 4 and 5 containing 50 or 100% R.O.M. although the individually-fed pigs were not similarly affected. The individually-fed animals were restricted in feed intake since they ate only 73% of the feed consumed per day by the ad libitum fed pigs. Despite this, they made 90% of the weight gain and were therefore 20% more efficient feed converters. Bowland (1957) and others have reported a similar increase in feed utilization when feed intake of pigs was restricted. It is possible that the greater intake of goitrogenic factor per unit body weight with the group-fed animals resulted in a more noticeable ration effect.

In the rat trials, rate of gain and efficiency of food utilization were reduced by the high levels of R.O.M. although some degree of tolerance appeared to be developing during the latter part of the grower period.

Lysine had no significant effect on either rate of gain or efficiency of food utilization in either species.

Barrows gained 0.07 lb. per day faster than gilts in Experiment 356A while boars and gilts in Experiment 356 had almost identical gains of 1.28 and 1.30 lb. per day respectively. On the basis of published work in which boars have gained faster than either gilts or barrows (Fredeen, 1953; Charette, 1961) it seems possible that at this particular stage of growth boars in Experiment 356 were affected more by the R.O.M. diets than gilts were. Barrows in lot 1 in the group-fed trials gained

0.14 lb. per day faster than gilts. This suggests a greater tolerance to the R.O.M. goitrogen by the females. Male rats grew 40% faster and required only 60% of the food to make body weight gains equal to the females.

Feed intake with the individually-fed pigs declined on the 100% levels of R.O.M.; however, group-fed animals ate normal levels of all rations. Food consumption was not affected by levels of R.O.M. in the rat experiments.

(c) Finisher Period. - 110 - 210 lb. liveweight for pigs in Experiment 356 and 110 - 195 lb. in Experiment 356A; 8 to 12 weeks of age for rats.

By the beginning of the finisher period it appeared that the pigs had become tolerant to the rations high in R.O.M. and body weight gains in all 5 ration groups were similar. The same tendency was evident with the rats and ration effects were not significant in either average daily gain or efficiency of food utilization.

It should be pointed out that the finisher ration contained only 9.6 as compared to 15.6% R.O.M. in the grower ration. This lowered level of R.O.M. intake may at least partially account for the improved performance of both species of animals.

Barrows fed ad libitum and individually-fed boars gained 0.13 and 0.36 lb. per day more than gilts in their respective experimental groups. Boars were 20% more efficient feed converters than gilts. Barrows and gilts did not differ in efficiency of feed utilization although barrows ate 0.37 lb. more feed per day thus accounting for the increased daily gain. These sex differences are in agreement with previously mentioned workers.

(d) Summary of the Overall Period of Growth.

With both swine and rats, during the starter and grower periods, rate of gain and efficiency of food utilization decreased with increasing levels of R.O.M. above 25% of the supplemental protein. This effect was most pronounced at the 100% level of supplementation; these results are in agreement with Hussar and Bowland (1959). The 25% level of R.O.M. supplementation caused no deleterious effects on rate of gain or efficiency of food utilization. The 50% level gave inconsistent results indicating that this regimen was borderline; some animals tolerated it extremely well while others were moderately to severely affected.

Supplemental lysine added to the 100% R.O.M. ration did not have any consistent effect on either species. McWard et al. (1959) found that with levels of 12.8 and 21.7% protein in grower rations for pigs that 5.55 and 4.38% lysine (expressed as % of the total protein in the ration) respectively gave optimum growth. A similar report by Magruder et al. (1962) indicated that the lower the level of protein in a ration the greater the proportion of lysine must be. On the basis of the N.R.C. Nutrient Requirements for Swine (1959) one would not expect to get any response from lysine in Experiments 356 and 356A. The levels of 0.78 and 0.86 in the starter and grower rations should be adequate although the 0.68% level of lysine in ration 4 of the finisher period may be borderline. In any case, the growth data in Tables 5 to 8 indicate that lysine levels in the diets were not a limiting factor.

There was definite evidence of increased feed efficiency with pigs when feed intake was restricted and it is suggested that this point may be of practical significance as a means of reducing the toxicity of R.O.M. when fed at high levels. This increased efficiency of feed utilization has been previously reported by Barber et al. (1957) and Berg and Bowland (1958).

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Barrows grew 0.06 pounds per day faster than gilts. Boars grew 0.14 pounds per day faster and were 13% more efficient feed converters than gilts in Experiment 356. Crampton and Ashton (1945), Bennett and Coles (1946) and Fredeen (1953) have reported similar sex effects. Increased efficiency of feed utilization by boars has been noted by Charette (1961).

Male rats gained 30% faster and were 18% more efficient food converters than female rats. The growth data (Tables 7 and 8 and Figures 2 and 3) do not show any clear evidence that one sex was more severely affected than the other. No statistically significant sex x ration interactions were present.

Palatability was no problem with levels of R.O.M. as high as 15.6% of the total ration.

Growth data and food efficiency figures indicate that despite the rather marked difference in thiooxazolidone and isothiocyanate contents of the two meals used in rat trials 1A and 1B (Table 3) the value as a protein supplement has not been changed appreciably. Clandinin¹ (1962) fed these same meals to chicks and found that thyroid to body weight ratios were increased 320% and 290% by batches 1 and 2 respectively. Body weight gains were 8% lower when chicks received 15% R.O.M. from batch 1 as compared to batch 2. This reduction in rate of gain was not so great as the thiooxazolidone and isothiocyanate contents of the meals would indicate (Table 3) and for this reason it may be that thiooxazolidone and isothiocyanate are not the only toxic factors in R.O.M. as Bell and Williams (1953) have suggested.

¹Private communication.

Figure 2. Growth Curves for Male Weanling Rats Fed Various Levels of R.O.M. for a Period of 9 Weeks.

Figure 3. Growth Curves for Female Weanling Rats Fed Various Levels of R.O.M. for a Period of 9 Weeks.

Table 5.

Rate of Gain, Feed Consumption and Efficiency of Feed Utilization
Individually-Fed Pigs, Experiment 356¹

R.O.M. %	0	25	50	100	100 + L ²	Boars	Gilts ³
Number of pigs	6	6	6	6	6	10	20
Av. initial wt. lb.	15.7	15.6	16.2	15.8	15.6	16.5	15.4
Av. age on test days	36	36	36	36	36	38	36
Av. final wt. lb.	217	213	215	214	216	216	215
Av. ⁴ age at 210 lb. days	207	211	210	212	215	196	218
<u>Starter Period (15 - 55 lb.)</u>							
Av. daily gain lb.*	0.79 ^A	0.78 ^{AB}	0.73 ^{ABC}	0.60 ^C	0.65 ^{BC}	0.74	0.69
Av. daily feed lb.	1.72	1.64	1.63	1.44	1.52	1.65	1.56
Feed per 100 lb. gain lb.	220	212	227	241	238	229	227
<u>Grower Period (55 - 110 lb.)</u>							
Av. daily gain lb.	1.34	1.32	1.29	1.32	1.19	1.28	1.30
Av. daily feed lb.*	3.59 ^A	3.47 ^{AB}	3.48 ^A	3.30 ^{AB}	3.18 ^B	3.37	3.42
Feed per 100 lb. gain lb.	262	268	274	252	268	260	267
<u>Finisher Period (110 - 210 lb.)</u>							
Av. daily gain lb.	1.35	1.25	1.36	1.52	1.37	1.61	1.25**
Av. daily feed lb.	5.28	5.02	5.43	5.53	5.62	5.55	5.29
Feed per 100 lb. gain lb.	400	430	419	372	411	348	435
<u>Overall Period (Start to 210 lb.)</u>							
Av. daily gain lb.	1.18	1.13	1.14	1.13	1.10	1.23	1.09**
Av. daily feed lb.	3.37	3.75	3.74	3.48	3.66	3.66	3.69
Feed per 100 lb. gain lb.	324	336	337	312	336	299	344**

* Significant at P<5%. ** Significant at P<1%.

¹ Means in each row with the same superscript did not vary significantly from each other.

² Lysine abbreviated to L.

³ Asterisks in this column in this and subsequent tables denote significant differences between sexes.

⁴ Adjusted to 210 lb.

Table 6.

Rate of Gain, Feed Consumption and Efficiency of Feed Utilization
Group-fed Pigs, Experiment 356A¹

R.O.M. %	0	25	50	100	100 + L	Barrows	Gilts
Number of pigs	8	8	8	8	8	20	20
Av. initial wt.	20.0	20.0	20.4	20.3	19.5	20.5	19.6
Av. age on test	43	42	40	41	40	42	40
Av. final wt.	199	199	203	201	199	200	200
Av. ² age at market	164	169	176	175	181	172	176
<u>Starter Period (20 - 50 lb.)</u>							
Av. daily gain	1.11 ^A	1.10 ^A	0.96 ^B	0.89 ^B	0.88 ^B	1.01	0.96
Av. daily feed	2.66	2.60	2.44	2.34	2.26	2.45	2.47
Feed per 100 lb. gain	240	238	254	262	256	244	256
<u>Grower Period (50 - 110 lb.)</u>							
Av. daily gain	1.54 ^A	1.48 ^{AB}	1.37 ^C	1.40 ^{BC}	1.36 ^C	1.47	1.40
Av. daily feed	4.90	5.04	4.06	4.14	3.21	4.92	4.43
Feed per 100 lb. gain	319	342	296	294	380	336	316
<u>Finisher Period (110 - Market Weight)</u>							
Av. daily gain	1.70	1.58	1.56	1.64	1.54	1.67	1.54*
Av. daily feed	6.94 ^{BC}	7.16 ^{AB}	6.80 ^{BC}	6.50 ^C	7.63 ^A	7.19	6.82*
Feed per 100 lb. gain	412 ^{BC}	456 ^{AB}	436 ^{BC}	398 ^C	496 ^A	432	446
<u>Overall Period (Start to Market Weight)</u>							
Av. daily gain	1.48	1.42	1.34	1.34	1.28	1.41	1.35
Av. daily feed	5.12	5.27	4.80	4.52	5.18	5.08	4.87
Feed per 100 lb. gain	346	372	358	338	405	361	366

* Significant at P < 5%

¹ Means in each row with the same superscript did not vary significantly from each other.² Adjusted to 200 lb.

Table 7.

Rate of Gain, Food Consumption and Efficiency of Food Utilization
Rat Experiment 1A¹

R.O.M. %	0	25	50	100	100 + L	Males	Females
Number of rats ²	12	12	12	12	12	30	30
Av. initial wt.	39	39	42	41	40	40	40
<u>Starter Period (3 to 5 weeks of age)</u>							
Av. daily gain	gm.**	5.06 ^A	4.74 ^A	3.94 ^B	3.11 ^C	4.28	3.83*
Food per 100 gm. gain	gm**	230 ^B	230 ^B	263 ^{AB}	296 ^A	248	269**
<u>Grower Period (5 to 8 weeks of age)</u>							
Av. daily gain	gm.**	4.03 ^A	4.27 ^A	3.58 ^A	2.70 ^B	4.36	2.52**
Food per 100 gm. gain	gm.**	458 ^B	428 ^B	451 ^B	549 ^{AB}	393	608**
<u>Finisher Period (8 to 12 weeks of age)</u>							
Av. daily gain	gm.	2.16	2.16	2.43	2.21	2.96	1.59**
Food per 100 gm. gain	gm.	954	967	928	973	736	1169**
<u>Overall Period (3 to 12 weeks of age)</u>							
Av. daily gain	gm.**	3.42 ^A	3.42 ^A	3.07 ^A	2.54 ^B	3.68	2.37**
Food per 100 gm. gain	gm.**	497 ^B	512 ^B	518 ^B	610 ^A	472	627**
Av. total gain (males)	gm.	263	260	235	198		
Av. total gain (females)	gm.	169	170	151	122		
Av. total food consumed (males)	gm.	1142	1162	1057	990		
Av. total food consumed (females)	gm.	947	976	882	869		

* Significant at P<5%.

** Significant at P<1%.

¹Means in each row with the same superscript did not vary significantly from each other.
²16 rats per lot in the starter period.

Table 8.

Rate of Gain, Food Consumption and Efficiency of Food Utilization
Rat Experiment 1B¹

R.O.M. %	0	25	50	100	100 + L	Males	Females
Number of rats ²	4	4	4	4	4	10	10
Av. initial wt.	36 gm.	38	35	39	38	38	37
<u>Starter Period (3 to 5 weeks of age)</u>							
Av. daily gain	gm.** 4.72 ^A	4.27 ^{AB}	3.76 ^B	2.61 ^C	2.41 ^C	3.68	3.43
Food per 100 gm. gain	gm.* 258 ^C	287 ^C	343 ^{BC}	502 ^A	476 ^{AB}	367	379
<u>Grower Period (5 to 8 weeks of age)</u>							
Av. daily gain	gm.* 4.00 ^A	3.92 ^A	3.88 ^A	3.16 ^{AB}	2.64 ^B	4.41	2.63**
Food per 100 gm. gain	gm. 448	472	428	506	478	385	548
<u>Finisher Period (8 to 12 weeks of age)</u>							
Av. daily gain	gm. 2.02	1.79	1.93	2.01	1.76	2.34	1.46**
Food per 100 gm. gain	gm. 970	940	844	760	787	735	985
<u>Overall Period (3 to 12 weeks of age)</u>							
Av. daily gain	gm.** 3.32 ^A	3.03 ^{AB}	2.92 ^{AB}	2.62 ^{BC}	2.21 ^C	3.33	2.31**
Food per 100 gm. gain	gm. 514	530	547	566	610	500	607**
Av. total gain (males)	gm. 253	232	227	191	145		
Av. total gain (females)	gm. 166	150	141	138	133		
Av. total food consumed (males)	gm. 1118	1088	1105	984	857		
Av. total food consumed (females)	gm. 974	888	858	857	836		

*Significant at P < 5%. **Significant at P < 1%.

¹Means in each row with the same superscript did not vary significantly from each other.²8 rats per lot in starter period.

B. Carcass Characteristics

The criteria for carcass quality evaluation are presented in Table 9. Although no statistically significant ration effects resulted from these trials, some trends are worthy of comment.

Animals on the 50 and 100% R.O.M. rations had longer carcasses. This could probably be expected as the animals were older at market weight. Hussar and Bowland (1959) fed R.O.M. as 10% of the total ration and reported that pigs tended to have shorter carcasses than control animals. Similar results were reported with thiouracil feeding (Bowland, 1957). No explanation of this apparent contradiction can be offered.

Pigs on the 50 and 100% levels of R.O.M. had a decreased area of loin. This confirms a similar report by Hussar and Bowland (1959).

Backfat measurements were essentially unchanged by R.O.M. feeding. Group-fed animals had 1.42 as compared to 1.32 inches of backfat for individually-fed animals. Zessin *et al.* (1961) and others have reported a similar backfat reduction when feed intake was restricted.

The levels of R.O.M. used did not influence dressing percentage. Pigs on the 50 and 100% R.O.M. rations had higher R.O.P. scores and superior carcass grades to the 0 and 25% R.O.M. groups.

Gilts showed overall superiority in carcass quality as compared to barrows but there was no ration x sex interaction in carcass characteristics. Bennett and Coles (1946), Charette (1961) and others have reported similar differences between gilts and barrows on non-goitrogenic rations.

In general one might conclude that the levels of R.O.M. used in this study have had no marked effect on carcass quality. Any possible advantages derived at the 100% level of R.O.M. feeding would certainly be offset by decreased rate of gain and reduced efficiency of feed utilization.

Table 9.

Carcass Characteristics of Group-Fed Pigs Experiment 356A, and Probed Backfat Thickness
of Individually-Fed Pigs, Experiment 356

R.O.M. %	0	25	50	100	100 + L	Barrows	Gilts
Number of pigs	8	8	8	8	8	20	20
Av. hot carcass weight lb.	159	159	162	157	158	158	159
Dressing %	80.2	79.8	79.8	78.4	78.9	79.1	79.7
Length of side in.	31.0	31.2	31.4	31.6	31.5	31.0	31.6*
Av. backfat thickness in.	1.49	1.48	1.37	1.36	1.42	1.48	1.37*
Area of loin sq. in.	3.93	3.64	3.83	3.52	3.43	3.51	3.83
Government Grades							
A	2	4	3	4	6	8	11
B	5	3	5	4	1	9	9
C	1	1	0	0	1	3	0
R.O.P. score	59	56	65	68	64	56	69*
Av. backfat thickness Individually-fed pigs ¹	1.40	1.27	1.30	1.30	1.37	1.32 ²	1.34

*Significant at $P < 5\%$.

¹Average of 3 probes, shoulder, back and loin, 2 boars and 4 gilts per lot.

²Average of 10 boars.

C. Reproduction

Table 10 outlines the data from the reproduction studies with pigs and rats. These results show considerable variation both within and between groups. While such variation is normal in reproduction studies it creates problems of interpretation. This consideration, coupled with limited numbers of animals, prompted the author to dispense with any statistical analysis of the data.

Only 15 of the original 20 gilts farrowed. One gilt was lost from each of lots 2, 3 and 4 because these animals did not conceive in the prescribed 4 oestrus periods. One gilt in lot 3 was injured at the time of breeding and the other gilt from lot 4 had not conceived when she died of infectious nephritis 112 days after reaching 210 lb. in weight.

The results are confounded by the fact that 5 littermate gilts, 1 in each group, were either late in reaching puberty or were much more severely affected by R.O.M. than other gilts. Two of these animals (lots 2 and 3) were marketed because they did not become pregnant; the third (lot 4) died of infectious nephritis. The gilts from this litter in lots 1 and 5 conceived normally on the first breeding period.

The most striking effect of the 50 and 100% levels of R.O.M. was the delay in onset of puberty. All gilts that farrowed on the 0 and 25% levels of R.O.M. conceived on first service after they reached the required breeding age; oestrus periods had been noted in these groups previous to this time. Gilts on the 50 and 100% levels of R.O.M. did not conceive until an average of 62 days after reaching 210 lb. while the basal and 25% R.O.M. groups were pregnant 21 days earlier. Kennedy

and Purves (1941) noted a delay in development of the ovaries of immature rats fed rapeseed. Gilts on the high R.O.M. diets may have experienced a delay in onset of puberty because of a delay in ovarian development as Kennedy and Purves suggested. Boars in all 5 lots had reached sexual maturity at the age of 240 days and breeding performance, as judged by their ability to sire litters, seemed completely normal in every instance.

Bezeau et al. (1960) reported that ewes fed R.O.M. as 30% of the total ration lost weight during the gestation period. The maximum level of R.O.M. in the reproduction period in the current study was 12% of the total ration and gestation gains were normal. Lucas et al. (1958) noted that thiouracil-fed pigs had longer than normal gestation periods; gilts in these experiments were not similarly affected as length of the gestation period was uniform at 112 to 114 days.

Number of young born alive per sow and birth weights were normal in all groups. The weight of the piglets at 5 weeks of age was reduced from 16.6 lb. per animal in the basal group to 13.1 lb. in the 100% R.O.M. groups. Presumably this could be a result of lactation inadequacy such as Bell (1957) has reported with mice.

Rats used in the reproduction study all seemed normal in so far as age of sexual maturity and conception rate were concerned. Maqsood (1952) reported that Rowlands observed that male rats rendered hypothyroid by thyroidectomy did not experience normal testicular maturation. Krohn and White (1950) and Jones et al. (1946) have reported normal conception rate in thyroidectomized and thiouracil treated rats but both noted that fewer young were brought to term due to foetal resorption.

The number of rats born per litter was reduced from 11.8 at the 0 level of R.O.M. to 9.0 at the 100% level of R.O.M. feeding. The main difference, however, was that females on the 100% R.O.M. averaged only 6.5 living young at birth as compared to 11.8 for the basal group. It is of interest to note that 2 of the 4 females on the 100% R.O.M. + lysine diet completely resorbed their litters. Jones et al. (1946) has reported 100% resorption of litters in rats when thiouracil was fed for over 100 days. If this phenomenon is due entirely to a thyroid condition then one wonders why some resorption did not occur in group 4 which should have been equally affected to group 5.

Average birth weights of rats were quite normal in all ration groups but since serious lactation difficulties arose in many animals regardless of ration no further litter data are available. Only 4 of the 18 litters were weaned, 1 each in groups 1 and 2 and 2 litters in group 3. Post mortems on several females revealed little or no active mammary tissue, the reason for this lactational disturbance is not known but it certainly could not be attributed entirely to R.O.M. as females on the basal diet were similarly affected.

Histological sections of male and female gonads at 12 weeks of age revealed no distinct abnormalities in any of the rats.

Levels of R.O.M. up to 6% of the total diet (lot 3) can be used in rat and swine diets with no consistent deleterious effects. The 12% level (lots 4 and 5) promoted foetal resorption and stillbirths in rats. Lactation results were not encouraging with either species.

Table 10.

Reproductive Performance of Pigs in Experiment 356 and of Rats in Experiment 1A

<u>Pigs</u>					
R.O.M. %	0	25	50	100	100 + L
Sows farrowing per lot	4	3	2	2	4
Days from 210 lb. to breeding	39	43	68	54	63
Sow wt. at breeding	lb. 257	258	264	266	274
Total gain gestation period	lb. 131	142	140	134	121
No. born alive per litter	9.0	8.7	9.5	8.0	9.0
No. weaned per litter	7.5	7.3	9.0	5.0	6.8
Av. birth wt. per pig	lb. 2.6	2.1	2.7	2.1	2.4
Av. wt. per pig (5 weeks of age)	lb. 16.6	15.5	15.4	12.6	13.5
Loss in wt. of sow (farrowing to weaning)	lb. 63	65	89	60	52
Length of gestation period	113	114	113	112	113
<u>Rats</u>					
No. of litters per lot ¹	4	4	4	4	2
Number born per litter	11.8	12.2	11.5	9.0	9.0
Number born alive per litter	11.8	11.5	10.3	6.3	7.0
Av. birth wt.	gm. 5.9	6.0	6.4	5.7	6.1

¹Two resorptions from lot 5. Averages do not include these 2 females.

D. Digestibility and Retention Experiments

(a) Swine Studies

In order to simplify comparisons between pig and rat digestibility data, the results for apparent digestibility of nitrogen, energy and dry matter¹ are given as digestion coefficients. Digestibility figures are based on 10% moisture in both feeds and feces since it was found that random samples of feed and feces showed dry matter contents of $90 \pm 1\%$.

The data from the swine trials (Table 11) show considerable variability. In general lot 4 animals receiving 100% R.O.M. exhibited the lowest digestibility coefficients of any group. Lot 5 animals, receiving 100% R.O.M. plus lysine, were higher than lot 4 except in the finisher period. This increase reached statistical significance ($P < 5\%$) for A.D.E. and D.M. digestibility in the grower period and for A.D.N. in the reproduction period.

A.D.N., A.D.E. and D.M. digestibility tended to decrease from lot 1 to lot 4. Ration 3 (50% R.O.M.) pigs did not follow any particular trend and it seems reasonable to conclude that the 50% level of R.O.M. supplementation did not, on the average, affect digestibility coefficients. Hussar and Bowland (1959) got more consistent results in their work and reported decreased energy and dry matter digestibility at a 10% level of R.O.M. supplementation.

It will be recalled from the previous discussion of rate of gain and efficiency of feed utilization (Section A, Tables 5 and 7) that during the grower and particularly the finisher periods, pigs on the high R.O.M. diets became more tolerant of the R.O.M. goitrogen. This could explain why A.D.N., A.D.E. and D.M. digestibility coefficients obtained¹A.D.E., A.D.N. and D.M. digestibility refer to apparent digestibility of energy, nitrogen and dry matter.

from pigs which had been on experiment for at least 75 days did not show marked effects of the R.O.M. rations. Possibly if a digestibility study had been conducted 20 to 30 days after the initiation of the experiment when the animals weighed 30 to 40 pounds and were experiencing the most serious growth depression on the high R.O.M. diets, the digestibility results would have been more conclusive.

In the finisher period boars showed significantly higher energy and dry matter digestibility than was shown by gilts. Since there was no sex x ration interaction this sex difference is not attributable to R.O.M.

During the gestation period there was a significant increase of A.D.N. from 75% in lots 1, 2 and 4 to 81% in lots 3 and 5. This effect was particularly evident with the 100% R.O.M. + lysine group which was 6% above the unsupplemented 100% R.O.M. group. Similar, though non-significant, trends were present with A.D.E. and D.M. digestibility in the gestation period. No significant digestibility differences were present in the lactation period although lot 4 animals consistently had lower digestibility coefficients than other groups. The lysine supplemented lot had A.D.N., A.D.E. and D.M. digestibility coefficients 6 to 10% higher than lot 4 animals during gestation and lactation. In view of results obtained with rats, to be reported later, it seems unlikely that the additional lysine was entirely responsible for this noticeable increase in digestibility although since the trend occurred in the grower, gestation and lactation periods one cannot preclude the possibility that improved protein quality was involved. Further experimentation is required to clarify this point.

(b) Rat Studies

The results of the rat digestibility and retention studies are presented in Table 12. The digestibility trial with rats was conducted in the second week of the starter period; in this way 1 week was allowed for ration acclimatization. Contrary to the data from the pig digestibility studies the results were uniform and more amenable to satisfactory interpretation.

Rats on the 0 and 25% R.O.M. diets had higher A.D.N. than rats on the 100% R.O.M. diets. The 50% level of R.O.M. consistently produced intermediate values. At the 0 and 25% level of R.O.M., A.D.N. was only 3.3% greater than the 100% levels. However, a similar comparison of percent A.D.N. retained shows that at the 100% R.O.M. levels 9.5% less A.D.N. was actually utilized as tissue protein. Hussar and Bowland (1959) reported that R.O.M. as 10% of the total ration did not affect the retention of nitrogen although they noted a reduction in A.D.N. at this level of R.O.M. feeding. These results suggest that besides being less readily digested by the animals, the protein of R.O.M. was not used as efficiently as the protein of soybean oil meal. Presumably this difference could be associated with the inferior protein quality of R.O.M. or more probably to a combination of inferior protein quality and an overall metabolic disturbance arising from thyroid gland malfunction.

Thyroxine at normal physiological levels stimulates protein anabolism (Pitt-Rivers and Tata, 1959) and hyperthyroidism increases protein catabolism. Konigsberg (1958) treated developing chicks embryos with thiourea and got inhibition of growth of skeletal muscle cells by

reduced protein accumulation in the cells. Normal protein levels could be restored by thyroxine injection. He concluded that the protein synthesizing mechanism was interfered with when thyroxine was not adequate possibly because of disruption of normal energy pathways. Such an effect could be associated with the decreased nitrogen retention in the present study.

The growth data discussed previously and the digestibility data of this section imply that additional lysine was of no benefit to the rat and that protein quality in so far as lysine is concerned was not limiting.

Apparent digestibility figures for energy and dry matter decreased as the levels of R.O.M. increased. The 50% level of R.O.M. supplementation resulted in a non-significant decrease of 1% while at the 100% level significant reductions of 4% occurred in energy and dry matter digestibility.

There were no differences between males and females in digestibility of A.D.N., A.D.E. and D.M. although there was a tendency for females to retain less of the nitrogen digested.

The data from the digestibility study done on approximately the 18th day of pregnancy followed a pattern very similar to the one previously described. A.D.N., A.D.E. and D.M. digestibilities all decreased with the higher levels of R.O.M. A similar trend was evident with retention of gross nitrogen.

An overall summary of the data indicates that as the level of R.O.M. in the diets increased apparent digestibility of nitrogen, energy

Table 11.

Nitrogen, Energy and Dry Matter Digestibility
Individually-Fed Pigs, Experiment 356¹

R.O.M. %	0	25	50	100	100 + L	Boars	Gilts
Number of pigs	3	3	3	3	3	5	10
<u>Grower period</u>							
A.D.N.							
	65.0 ^B	65.7 ^B	67.1 ^B	68.4 ^B	74.1 ^A	69.4	67.3
A.D.E.	68.7 ^B	68.8 ^B	67.7 ^B	66.0 ^B	73.1 ^A	69.9	68.3
D.M. digestibility	71.9 ^{AB}	70.9 ^{BC}	70.4 ^{BC}	67.3 ^C	74.7 ^A	71.8	70.7
<u>Finisher period</u>							
A.D.N.							
	76.6	72.0	77.6	71.4	68.9	74.9	72.5
A.D.E.	69.0	65.6	67.2	63.0	59.2	68.4	63.0*
D.M. digestibility	71.5	68.8	70.1	68.2	63.1	71.4	66.8*
<u>Reproduction (gilts 90th day gestation, boars 340 days of age)</u>							
A.D.N.							
	76.0 ^{BC}	72.8 ^C	80.3 ^{AB}	76.3 ^{BC}	82.4 ^A	79.5	76.6
A.D.E.	66.0	61.3	70.8	63.0	74.9	71.9	64.8
D.M. digestibility	67.6	63.9	72.3	65.0	75.3	73.3	66.6
<u>Twenty-first day of Lactation</u>							
Number of pigs	2	2	2	2	2		
A.D.N.							
	78.8	76.1	76.5	73.9	79.4		
A.D.E.	70.7	65.5	66.0	64.4	71.8		
D.M. digestibility	72.4	67.8	67.8	65.7	73.6		

*Significant at $P < 5\%$.¹Means in each row with the same superscript did not vary significantly from each other.

Table 12.

Nitrogen, Energy and Dry Matter Digestibility and Nitrogen Retention
Rat Experiment 1A¹

R.O.M. %	0	25	50	100	100 + L	Males	Females
Number of rats	16	16	16	16	16	40	40
Av. initial weight gm.	70.2	67.7	66.6	61.4	64.0	67.4	64.5
Av. food consumed gm.	92.8	87.8	75.5	66.8	68.9	80.8	75.9
Av. daily gain gm.	5.7	5.5	4.3	3.3	3.4	4.8	4.1
Food per 100 gm. gain	234	228	255	349	301	250	297
N ingested mg.	2523	2408	2089	1841	1874	2214	2080
A.D.N. %**	84.5 ^{AB}	85.8 ^A	83.9 ^{AB}	81.7 ^C	82.0 ^{BC}	83.4	83.7
Gross N retained %**	52.5 ^A	52.7 ^A	47.2 ^{AB}	41.8 ^C	43.4 ^{BC}	48.5	46.5
A.D.N. retained %**	61.5 ^A	61.3 ^A	56.3 ^B	50.9 ^B	52.9 ^B	57.8	55.4
A.D.E. %**	86.5 ^{AB}	87.5 ^A	85.7 ^B	82.2 ^C	82.7 ^C	84.7	85.2
Digestible kcal/100 gm. food	352	355	346	327	331	341	342
Kcal. per 100 gm. gain	821	808	880	1136	990	848	1005
D.M. digestibility %**	87.6 ^A	88.4 ^A	86.9 ^A	83.5 ^B	83.8 ^B	85.8	86.3
<u>Eighteenth day of gestation</u>							
Number of rats	2	2	2	2	2		
A.D.N. %	78.7	71.4	59.3	69.2	61.2		
Gross N retained %	42.0	32.0	18.5	43.5	29.5		
A.D.E. %	76.3	66.8	69.8	71.6	69.6		
D.M. digestibility %	77.6	68.8	72.2	73.1	70.2		

*Significant at P < 5%.

**Significant at P < 1%.

¹Means in each row with the same superscript did not vary significantly from each other.

and dry matter decreased although this effect was significant only in the rat experiments. Supplemental lysine in rations containing high levels of R.O.M. may have increased swine digestibility coefficients for nitrogen, energy and dry matter but it was ineffective for rats.

E. Blood Serum Protein-Bound Iodine

Rat blood serum P.B.I. values were determined at 5 and 12 weeks of age. The data are reported in Table 13. There was some difficulty in obtaining enough serum from the 5-weeks-old rats. Making a pooled serum sample from 2 or more rats on the same treatment might be considered as a way of alleviating this problem in the future. The P.B.I. values for the five 12-week-old male rats in lot H were not included in the averages in Table 13 since these determinations were all extremely high. Presumably the samples had been contaminated either from the atmosphere or the reagents. P.B.I. levels fluctuated considerably within, as well as between, groups. R.O.M. did not have any definite effect on P.B.I. values with rats at either 5 or 12 weeks of age.

Swine blood serum P.B.I. values from the group-fed pigs were reduced from 5.32 in the basal lot to 3.13 and 3.56 ug at the 50 and 100% levels of R.O.M. feeding (Table 15) respectively. As will be discussed in a subsequent section swine thyroid glands showed moderate hypertrophy as the levels of R.O.M. increased. The coefficient of correlation between P.B.I. and thyroid gland weight was $-.538$ (38D.F. $P < 1\%$).

P.B.I. values of the magnitude obtained are well within the normal range for humans of 3.4 to 8.0 ug per 100 ml. of serum cited by

Barker (1951). Gawienskie et al. (1955) determined P.B.I. values for swine, presumably normal, and found a range of 1.2 to 5.4 ug per 100 ml. of serum. This means that the P.B.I. values in the current study obtained from animals fed R.O.M. diets, while unquestionably reduced, may still be normal.

The growth and food efficiency data from the finisher period indicate that the animals were not experiencing any particular stress from the 50 and 100% diets. In view of the above observations coupled with conflicting reports in the literature as to the validity of P.B.I. as an indication of thyroidal activity, one hesitates to state unequivocally that a decrease from 5.32 to 3.13 ug iodine per 100 ml. of serum represents a definite deficiency of thyroid hormone secretion. It seems certain, however, that the animals in the 50 and 100% R.O.M. groups showed hypothyroid tendencies.

F. Thyroid Gland Morphology

Group-fed pigs in Experiment 356A did not show marked glandular hypertrophy, although animals on the 50 and 100% R.O.M. diets had thyroid glands 25% heavier than control animals. Histological sections of these glands did not indicate marked hyperplasia or increased cellular components. Hussar and Bowland (1959) found definite evidence of thyroid hypertrophy and hyperplasia when Argentine-type, expeller-extracted R.O.M. made up 10% of the total ration. In the current study the finisher ration contained 9.6% solvent-extracted Polish-type R.O.M. The results suggest that this meal may have been less goitrogenic than the one used by Hussar and Bowland. This could be related to type of meal or method of processing.

Van Middlesworth (1959) reported that propyl thiouracil fed rats could be injected with sodium-l-thyroxine daily until P.B.I. values were 2 to 3 times normal. These animals still showed signs of goitre despite the above normal blood thyroxine level. This phenomenon was attributed to an increased sensitivity of the thyroid gland to pituitary thyrotropin. If other goitrogens (viz. thiooxazolidone) could act similarly, then thyroid gland hypertrophy, unless very drastic should not be considered irrefutable evidence of thyroid malfunction.

Thyroids from rats fed R.O.M. in Experiment 1B showed definite hypertrophy as gland weights increased with increasing levels of R.O.M. By the end of the second week on trial a moderate degree of hypertrophy had begun; rats on the 50 and 100% R.O.M. diets had glands 40% heavier than those on the 0 and 25% R.O.M. groups. After 3 weeks of feeding, hypertrophy was more marked as evidenced by an 80% increase in weight. Thyroid gland weights from animals fed the R.O.M. diets for 9 weeks were still 35% heavier than the 0 and 25% R.O.M. groups. These results are illustrated by Table 14 and Figures 4, 5 and 6.

It has been noted previously that thyroid glands from pigs at market weight on the high R.O.M. rations were only 25% heavier than the basal lot and on histological examination did not show abnormalities. Thyroid studies of rats on trial for 9 weeks tend to confirm these findings. Pipes et al. (1958) and Hussar and Bowland (1959) have reported that a time lapse is necessary for the goitrogen to exert its maximum effect. The present study indicates that with rats this maximum effect comes at about 2 to 4 weeks after the initiation of feeding

R.O.M. Following this time the animals apparently adapted themselves to the goitrogenic diets as is demonstrated by the decreased thyroid hypertrophy and increased body weight gains in the finisher period in comparison to control rats.

G. Standard Metabolic Rates

Basal metabolic rate determinations have been widely used as an indication of thyroidal activity and despite their non-specific nature they must be regarded as one of the most useful techniques for the estimation of levels of thyroid secretion (Pitt-Rivers and Tata, 1959). Thyroxine administration has been demonstrated to accelerate oxygen consumption by most body tissues both in vivo and in vitro (Barker and Klitgaard, 1952; Reid and Kossa, 1955; Boyd and Oliver, 1960).

Elevated basal metabolic rates are accepted as being indicative of a hyperthyroid state and Pitt-Rivers and Tata (1959) have theorized that this may be the result of uncoupling of oxidative phosphorylation. This would result in inefficient energy trapping as high energy phosphate bonds by hyperthyroid animals. One would expect the converse effect in a hypothyroid state such as the R.O.M. goitrogen might induce or, if the R.O.M. isothiocyanates acted directly at the tissue level as Bach and Benda (cited by Bell, 1955) have reported.

Since it was considered unrealistic to use the term basal metabolic rate with rats (as opposed to humans) the conditions were kept as standard as possible and the term standard metabolic rate has been employed (see page 26 for a definition of this term).

As the levels of R.O.M. in the diet increased the standard metabolic rate decreased; this tendency was statistically significant ($P < 1\%$) in Experiment 1A and a similar trend was evident in Experiment 1B.

Figure 4 Rat Experiment 1B

Thyroid Photomicrographs, 2 Weeks on Trial

25% R.O.M. (Rat 2 C, Male).
Thyroid weight 12.5 mg./100 gm. body weight.
Normal, actively secreting gland.

100% R.O.M. + Lysine (Rat 5 C, Female).
Thyroid weight 17.8 mg./100 gm. body weight.
Glandular hypertrophy and hyperplasia are evident.

Magnification = 184 X

Figure 5 Rat Experiment 1 B
Thyroid Photomicrographs, 3 Weeks on Trial

0% R.O.M. (Rat 1 D, Male).
Thyroid weight 7.4 mg./100 gm. body weight.
Normal, actively secreting gland.

100% R.O.M. + Lysine (Rat 5 D, Male).
Thyroid weight 15.2 mg./100 gm. body weight.
Glandular hypertrophy and extreme hyperplasia.
Most follicles devoid of colloid.

Magnification = 184 X

Figure 6 Rat Experiment 1 B

Thyroid Photomicrographs, 9 Weeks on Trial

25% R.O.M. (Rat 2 A, Female).
Thyroid weight 12.3 mg./100 gm. body weight.
Normal, actively secreting gland.

100% R.O.M. + Lysine (Rat 5 A, Male).
Thyroid weight 8.3 mg./100 gm. body weight.
Follicles well filled with colloid indicating normal
activity. Gland shows no hypertrophy although it
may be slightly hyperplastic.

Magnification = 184 X

Rats on the 100% level of R.O.M. showed the most uniform decrease in oxygen consumption while animals on the 50% level were quite variable although the average value shown in Table 13 indicates a general reduction similar to the lot 4 and 5 animals. This variability in performance at the 50% level of R.O.M. supplementation has been noted previously and is ascribed to the greater ability of some individuals to tolerate the goitrogen.

Correlations of $-.385$ and $-.121$ were found between thyroid weight (mg. per 100 gm. body weight) and standard metabolic rate at 17 to 20 days and 24 to 27 days on trial respectively in Experiment 1B. The required coefficient of correlation to reach statistical significance with 8 degrees of freedom at the 5% level is $\pm .632$. Therefore the degree of correlation is rather low and one must conclude that if rate of oxygen consumption can be regarded as being indicative of levels of hormonal secretion as suggested by Pitt-Rivers and Tata (1959) then thyroïdal hypertrophy should be considered supplementary rather than concrete evidence of thyroid gland malfunction.

H. Liver and Adrenal Weights

Liver and adrenal weights showed no significant ration effects with the varying levels of R.O.M. in the rat experiments (Tables 13 and 14). Bell (1955) reported evidence of liver enlargement in R.O.M. fed animals and Money (1955) and Nichols and Rossiter (1955) noted decreased adrenal weights with rats in a hypothyroid condition. Brown-Grant (1958) reported that thyroidectomy and methyl thiouracil treated rabbits showed no definite adrenal changes although he considered it conceivable that hypothyroid animals might secrete less adrenocorticotropic hormone than normal.

Female adrenals, but not livers, were heavier than male glands after 9 weeks on trial. After reproduction females had heavier livers as well as heavier adrenals than males. Once again no ration effect was evident and presumably this is nothing more than a normal reaction to the extra stresses imposed during gestation and lactation.

The market pigs showed a definite but statistically nonsignificant trend to heavier livers with the increased levels of R.O.M. Barber et al. (1960) found that supplemental copper decreased liver weights as compared to control animals that had sub-optimal levels of dietary copper. This work suggests, as the current study does, that nutritional stresses often cause increased demands on the liver and as a result this organ undergoes variable degrees of compensatory hypertrophy depending on the gravity of the situation. Therefore it seems that even though the extent of liver enlargement with the group-fed pigs was moderate, one should not preclude the possibility that the liver and/or other organs were not functioning normally.

Histological sections of adrenal glands from both species did not exhibit abnormalities and photomicrographs have not been included in the manuscript.

Table 13.

Glandular Weights, Blood Serum Protein-Bound Iodine and Standard Metabolic Rates
Rat Experiment 1A¹

R.O.M. %	0	25	50	100	100 + L	Males	Females
<u>After 2 weeks on trial</u>							
Number of rats	4	4	4	4	4	10	10
Serum P.B.I. ²	6.28	5.92	7.10	5.97	5.27	5.84	6.37
Standard ³ metabolic rate**	.484 ^A	.496 ^A	.376 ^B	.404 ^{AB}	.410 ^{AB}	.441	.426
<u>End of 9 weeks on trial</u>							
Adrenal weight (mg./100 gm. body wt.)	6.91	8.32	8.32	10.37	9.98	6.66	10.89**
Liver weight (gm./100 gm. body wt.)	4.02	4.18	4.20	4.48	4.55	4.37	4.20
Serum P.B.I. (av. of 1 male and 2 females)	4.92	4.46	4.29	4.38	4.12		
<u>After reproduction</u>							
Number of rats	8	8	8	8	8	20	20
Adrenal weight (mg./100 gm. body wt.)	8.68	8.98	9.78	8.73	7.72	6.12	11.43**
Liver weight (gm./100 gm. body wt.)	4.16	4.36	4.28	4.63	4.15	3.89	4.74**

* Significant at $P < 5\%$.** Significant at $P < 1\%$.¹Means in each row with the same superscript did not vary significantly from each other.²ug. per 100 ml. blood serum.³Expressed as kcal. per 100 gm. body weight per hour. Body weight is raised to the 3/4 power. Eight rats per lot.

Table 14.

Thyroid Weights and Standard Metabolic Rates
Rat Experiment 1B¹

R.O.M. %	0	25	50	100	100 + L	Males	Females
Number of rats ²	2	2	2	2	2	5	5
Thyroid wt. 20 days on trial (mg./100 gm. body wt.)	11.4	11.6	13.4	19.1	16.5	13.0	15.8
Thyroid wt. 27 days on trial* (mg./100 gm. body wt.)	10.0 ^B	8.9 ^B	19.6 ^A	14.5 ^{AB}	19.4 ^B	12.3	16.6*
Standard metabolic rate (20 days on trial)	.405	.461	.497	.401	.326	.413	.423
Standard metabolic rate (27 days on trial)	.451	.431	.405	.386	.422	.390	.448
Thyroid wt. ³ 9 weeks on trial** (mg./100 gm. body wt.)	7.4 ^B	8.2 ^B	9.2 ^{AB}	12.7 ^A	9.6 ^{AB}	8.6	10.2*

* Significant at P < 5%.

** Significant at P < 1%.

¹Means in each row with the same superscript did not vary significantly from each other.²One male and 1 female per lot.³Two males and 2 females per lot.

Table 15

Blood Serum Protein-Bound Iodine; Liver and Thyroid Weights of Group-Fed Pigs
Experiment 356A¹

R.O.M. %	0	25	50	100	100 + L	Barrows	Gilts
Number of pigs	8	8	8	8	8	8	8
Liver weight gm.	1356	1557	1591	1806	1700	1554	1650
Thyroid weight gm.*	25.5 ^B	28.5 ^{AB}	32.9 ^A	32.3 ^A	32.2 ^A	32.3	28.3 ^{**}
Serum P.B.I.** (ug./100 ml. serum)	5.32 ^A	4.76 ^{AB}	3.13 ^C	3.68 ^{BC}	3.44 ^{BC}	3.85	4.29

* Significant at P<5%.

** Significant at P<1%.

¹Means in each row with the same superscript did not vary significantly from each other.

SUMMARY AND CONCLUSIONS

Solvent-extracted rapeseed oil meal was compared to soybean oil meal as a dietary constituent in rations for pigs and rats. The rations were formulated so that 0, 25, 50 and 100% of the soybean oil meal in the basal ration was replaced by R.O.M. on an equivalent protein basis. The percentages of R.O.M. in the entire rations at the 25, 50 and 100% levels were: starter and grower periods 3.9, 7.8 and 15.6%; finisher period 2.4, 4.8 and 9.6% and reproduction period 3.0, 6.0 and 12.0% R.O.M. respectively. Lysine at a level of 0.2% was supplemented to one ration containing the 100% level of R.O.M. to investigate the possibility of rations containing high levels of R.O.M. being deficient in this amino acid.

With pigs, the most severe growth depression resulted from the 100% substitution of R.O.M. for soybean oil meal. Group-fed pigs on the 100% R.O.M. rations gained 0.17 lb. per day slower than control animals throughout the entire experimental period. Individually-fed animals on a similar comparison gained 0.06 lb. per day less than controls. The 25% R.O.M. group was not adversely affected. At a 50% level of R.O.M. supplementation gains were slightly reduced. Animals in the starter period from 15 to 50 lb. were affected most by the inclusion of R.O.M. in the rations. An adjustment was made by the pigs during the growing period and during the finisher period no ration effects were evident.

Individually-fed pigs were restricted in feed intake as they ate only 3.7 as compared to 5.0 lb. of feed per day for group-fed animals. This restriction of feed intake was associated with a 10% improvement in efficiency of feed utilization. Swine carcass quality was improved

slightly by the high levels of R.O.M.; carcasses tended to be longer and have less backfat although they had smaller loin areas.

Rats responded similarly to pigs, although differences in rate of gain and efficiency of food utilization were much more definite with the former species.

Reproductive performance was adversely influenced by the 50 and 100% levels of R.O.M. The effects manifested were delayed onset of puberty in pigs; reduction in number of young born alive in rats and a general lactation disturbance in both species.

Energy, nitrogen and dry matter digestibility coefficients were lowered for both species as the levels of R.O.M. increased, but the differences were significant only for rats. Nitrogen retention by rats was lowered by the 50 and 100% R.O.M. diets. Pigs receiving 100% R.O.M. plus supplemental lysine had digestibility coefficients higher than the unsupplemented 100% R.O.M. diet. Additional lysine was ineffective for rats.

Standard metabolic rate determinations done during the third week of the experimental period showed a reduction of oxygen consumption by rats on the high R.O.M. diets. Blood serum protein-bound iodine values for rats were within the normal range and were not influenced by R.O.M. levels. A significant reduction in P.B.I. of swine serum was noted when pigs received 50 or 100% R.O.M. Serum P.B.I. values were negatively correlated with increased thyroid gland weight and these results provide evidence of a thyroid disturbance. Swine thyroid histological studies showed no definite trends. Rat thyroid glands were enlarged and hyperplastic after 2 to 4 weeks of R.O.M. feeding although after 9 weeks on trial this tendency had been overcome and glandular activity seemed

normal. Adrenals and livers from rats were normal in size at 12 weeks of age and after the reproduction period. Livers from market pigs tended to be enlarged at the 50 and 100% levels of R.O.M. feeding.

On the basis of these trials it may be concluded that:

1. Swine and rats will tolerate levels of R.O.M. as high as 3.9% in the total ration with no detrimental effects. Levels of 7.8 and 15.6% R.O.M. caused decreased rate of gain, lowered efficiency of food utilization and decreased digestibility of energy, dry matter and nitrogen.
2. Using criteria of rate of gain and efficiency of food utilization, both species of animals were able to adapt themselves to high levels of R.O.M. during the period of growth. This result was presumably associated with an increase in size or number of active cells in the thyroid gland.
3. R.O.M. at the levels fed may be deficient in lysine for swine although the rat trials did not confirm this observation.
4. Extreme caution should be used in feeding R.O.M. to pigs if they are to be used for reproduction purposes.

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