Studies on vitamins B and G in growth and lactation in the rat: A, The effects of extracts of vitamins B and G; B, The distribution of vitamin G

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UMI®
STUDIES ON VITAMINS B AND G IN GROWTH AND LACTATION IN THE RAT

(a) The Effects of Extracts of Vitamins B and G

(b) The Distribution of Vitamin G

BY

James Floyd Fenster

A Thesis submitted to the Graduate Faculty for the degree of

DOCTOR OF PHILOSOPHY

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INTRODUCTION

A Brief Discussion of Fundamental Food Constituents

All animals must rely upon their food supply for materials to be used in building body tissue and for energy to be used in performing the natural functions of life. Workers in physiological and nutritional chemistry have been interested for many years in determining the dietary essentials that must be supplied, in order that an animal may attain its complete development and perform all normal functions throughout an indefinite number of generations.

Prior to the twentieth century the biological chemist was interested chiefly in the composition of plant and animal tissues. Their analyses had conclusively demonstrated the presence of proteins, carbohydrates, fats, water, lipids, inorganic ions, and molecules in living tissue. In the nineteenth century elaborate methods of analyzing foods for proteins, carbohydrates, fats, water, and ash had been developed. Most foods included in human and animal diets had been analyzed and classified according to their composition. At that time it was generally believed that the nutritional value of a diet was wholly determined by the amounts of proteins, carbohydrates, fats, water,
chlorophyll was determined in the leaves. The nutritional requirements of

-6-
compartmena.

Further oxidized to carbon dioxide and water. Until then, in the normal processes of body oxidation these processes as well as those in the brain lead to an accumulation of acids and an acid condition of the blood resulting from carbon dioxide from the lungs.

This meet these, because they are more economical than protein.

Carbohydrates and fats supply the bulk of the energy in

* In order to furnish a margin of safety, the intake is supposed to exceed the requirements very little. In addition to this, in order to maintain an optimal condition, it is necessary to know the requirements of the organism and to adjust the amount of protein to meet the needs of the body. In order to meet certain basic needs, the body must have a certain amount of protein for the production of new proteins and the synthesis of proteins. These needs can be met by a source of nitrogen in the form of free nitrogen compounds.

**Rationale**

The above statement is the basis for the following statement: 

"Since these essential are present in the body, the reason why they are present is the lack of the necessary amount of these nutrients."
value solely as a source of energy. Burr and Burr (10), (11) demonstrated the production of kidney lesions, scaly tail, loss of hair, a scaly condition of the skin, and irregular ovulation in rats, if fats were excluded from the diet. The conditions mentioned above were prevented or cured when the animals were fed fats containing the unsaturated fatty acids, linoleic acid or linolenic acid. They also found a complex unsaturated oil like corn oil, linseed oil, or cod liver oil to be more effective than a single fatty acid, in preventing the above disorders. Recent investigations have demonstrated that although rats have the ability to synthesize fats from carbohydrates they are unable to produce certain essential unsaturated fatty acids.

Inorganic elements and their salts are essential in the formation of bone, teeth, blood, and other tissues. Records show that all through the ages animals have frequented salt licks; and the supply of salt has been a factor in the location and migration of man. At the present time the following elements are recognized as being required for mammalian nutrition: carbon, hydrogen, oxygen, nitrogen, sulphur, phosphorus, chlorine, iodine, sodium, potassium, calcium, iron, copper, manganese, and magnesium. In addition other elements: fluorine, arsenic, aluminum, zinc, and silicon may be required by some animals.

A group of accessory substances, known as vitamins, are now recognized as being required for the nutrition of many ani-
mals. The chemical composition and the exact physiological functions of most of the vitamins are not fully explained.

Vitamin A is a fat soluble substance required for the normal growth of many animals; it is also thought to play some part in the ability of the body to resist disease. A deficiency of vitamin A produces a keratinization of the epithelial tissues. A secondary infection, xerophthalmia, often develops in animals on a vitamin A deficient diet. Some of the more potent sources of vitamin A are: fish liver oils, butterfat, green vegetables, and grasses. In the last few years it has been conclusively demonstrated that rats fed a vitamin A deficient diet convert carotene to vitamin A. It is now known that vitamin A is closely related to carotene in structure.

The group of accessory factors that was for a time considered as water soluble vitamin B has been divided into two factors. The more heat-labile antineuritic factor is known as vitamin B in this country and as vitamin $B_1$ in the European countries. The more heat-stable anti-dermatitis factor is now known as vitamin G in this country and as vitamin $B_2$ in the European countries. A deficiency of vitamin B produces a nervous disorder known as polyneuritis in pigeons and beriberi in man. Beriberi or polyneuritis is cured or prevented by adding a source of vitamin B to the diet. Vitamin B promotes growth by stimulating the appetite in addition to exerting a specific effect independent of the plane of nutrition. Several workers
have prepared crystalline products having antineuritic properties. Although the exact chemical structure of vitamin B has not been established, it is known to be an organic base. Block and Cowgill (6) found vitamin B as a base to be soluble in ether and as a salt to be insoluble in ether. Vitamin G is readily prepared free from vitamin B by autoclaving. The heat-stable vitamin G is a growth promoting vitamin. It also is effective in the treatment and prevention of pellagra in man and dermatitis in rats. Data published in the last few years show that vitamin G is probably not a single chemical compound but is composed of at least two; and it may contain as many as five separate compounds, each having a distinct physiological function. Vitamin G contains one factor that is destroyed by autoclaving in an alkaline medium and another factor stable to autoclaving in an alkaline medium. The factor destroyed by autoclaving in an alkaline medium is referred to as vitamin B4 by the European workers. Vitamin B4 has recently been crystallized. Crystals of vitamin B4 do not have all of the physiological properties of vitamin G.

Vitamin C, the anti-scorbutic factor, is required in the diet of only three animals. These animals are man, monkey, and guinea pig. Some other animals have the power to synthesize vitamin C. Within the last year vitamin C has been isolated and synthesized in vitro. Some of the more important sources of vitamin C are citrus fruits, tomatoes, green vegetables, and
The dietary disease, rickets, is a condition in which calcification of bone is abnormal. Rickets may be cured or prevented by adjusting the calcium phosphorus ratio in the diet to the proper value, by irradiation of the animal with ultra-violet light, or by including vitamin D in the diet. Several workers have demonstrated that ergosterol upon being irradiated with ultra-violet light takes on the physiological properties of vitamin D. This fact demonstrates that vitamin D is a sterol and is very similar to ergosterol in structure. The exact change that takes place when ergosterol is converted to vitamin D by irradiation with ultra-violet light has not been established. The distribution of vitamin D in natural foods is rather limited. The most potent natural sources of vitamin D are: fish liver oil, egg yolk, and butterfat.

Vitamin E is an anti-sterility vitamin. The manifestation of vitamin E deficiency differs in the two sexes. Females on a vitamin E deficient ration ovulate and the foeti are implanted. The sterility of the females is due to the failure of the placental function, causing the death and resorption of the foeti. In males vitamin E deficiency produces a permanent sterility, due to the destruction of the germ cells and the entire seminiferous epithelium. This sterility is cured or prevented in the female and prevented in the male by including a source of vitamin E in the diet. Vitamin E is widely distributed in natural foods, and
The most potent sources are; wheat germ oil, lettuce oil, and oils of cereal grains. Very potent preparations of vitamin E have been prepared from wheat germ oil and lettuce oil. The composition and structure of vitamin E remain unknown.
Statement of the Problem

An adequate knowledge of the vitamin content of the different food materials consumed by man and his domestic animals is required in preparing economical diets. An understanding of the animal's need for each specific vitamin at the different stages in its life cycle is of equal importance in the production of healthy animals with a minimum cost. In order to obtain data concerning the vitamin content of different foods, it is necessary to feed a diet adequate in all dietary factors except the vitamin for which the food is being tested. Diets which meet these requirements can be prepared only from highly purified materials.

The rat lends itself very well to vitamin research, because of its small size, small food consumption, and adaptability to laboratory conditions. The physiology of the rat approximates that of domestic animals and man. In addition the life cycle of the rat is completed in about one-thirtieth of the time required to complete a life cycle in man. This facilitates the collection of data concerning the vitamin requirements during the different stages in the life cycle.

The rat was used as the test animal in all experiments reported in this thesis. Rats from a strain of highly inbred animals maintained in the animal colony of the Chemistry Department of Iowa State College were employed throughout these studies. Previous researches in this laboratory have demonstrated that
diets that are adequate for normal growth and reproduction failed to support normal lactation. Certain seeds were demonstrated to be inadequate as the sole source of vitamins B and G for lactation. The studies reported here were designed to produce further information concerning the following questions:

1. Do various seeds and products from seeds when fed at certain levels furnish sufficient water soluble vitamins for growth?

2. Which is the limiting factor in seeds and seed products for growth, vitamin B, or vitamin G?

3. Do seeds and products from seeds when fed at certain levels furnish sufficient water soluble vitamins during the lactation period?

4. Is vitamin B or vitamin G the limiting factor in seeds during lactation?

5. Does the rat require an increase in vitamin B during the lactation period?

6. Does the rat require an increase in vitamin G during the lactation period?

7. Is the failure of mother rats to rear their young due to a decreased quantity or a poor quality of milk?

8. What is the vitamin G content of certain foods?

9. What is the solubility of vitamin G?

10. What are some of the properties of vitamin G?
Review of Literature

Research in nutrition has progressed at such a rate since the beginning of this century as to make it impossible to include an exhaustive review of the literature pertaining to vitamin B, vitamin G, and lactation in this paper. Hence, only the literature that has a direct connection with the study in this paper will be mentioned. For a more complete treatment of the subject of nutrition the reader is referred to "The Newer Knowledge of Nutrition", by E. V. McCollum and Nina Simmonds (63), and "The Vitamins", by H. C. Sherman and S. L. Smith (99).

The relation of the maternal diet to milk secretion and rearing of young is very important in the problem of the nutritional requirements of mammals. In the last decade considerable data have been accumulated, which demonstrate that certain diets which are satisfactory for continuous growth at a normal rate are unable to support lactation, as measured by the ability of rats to rear normal young.

Adair (1), in 1925 reviewed the literature on lastation of humans and laboratory animals. He concluded from the data available at that time that the vitamin content of milk is dependent upon the vitamins ingested. A case in the United States of an underweight infant exhibiting rigidity of limbs and neck, was referred to by Hoobler (42). The symptoms of rigidity were cured and an increase in weight was produced by feeding one-half
-15-
teaspoon of yeast concentrate daily. Hoobler suggests that there may be cases of vitamin B deficiencies in infants in this country. Some of the symptoms of this deficiency are anorexia, loss of weight, spasticity of the arms and legs, rigidity of the neck, restlessness, and fretfulness.

In a study of the vitamin B content of human milk Macy, Outhouse, Graham, and Long (65) found 25c.c. to 30c.c. were required per day to furnish vitamin B to promote normal growth in rats. They concluded that many mothers probably do not supply sufficient vitamin B to their babies. McGosh, Macy, and Hunscher (64) found that human mothers receiving their regular mixed diet did not secrete milk of as high a vitamin B (B complex) content as mothers which received a daily supplement of 10g. of yeast. Donelson and Macy (20), using the Sherman technique found pooled human milk fed at the following levels, 3.0, 5.0, 10.0, 15.0 and 20.0c.c. per day to produce gains of 19, 32, 41, 72, and 94g., respectively, in a period of eight weeks. Control animals receiving 0.4g. of autoclaved yeast per day made a gain of 154g. in eight weeks. In 1934 (19) the same workers reported that pooled human milk contained 0.2 units of vitamin G and 0.1 unit of vitamin B per c.c. They fed each woman a supplement of 10g. yeast per day and found the vitamin G content was increased to 0.3 units of vitamin G per c.c.; but the vitamin B content remained unchanged during the yeast feeding.

Anderegg (2) observed that female rats receiving a diet
containing 50 per cent of whole milk as a source of water soluble vitamins had difficulty in rearing their young. He also found wheat germ at a level below seven per cent to be inadequate for lactation. Guest, Nelson, Parkes, and Fulmer (38) fed the following grains, wheat, rye, barley, white and yellow corn at different percentages of the diet as the sole source of the vitamin B complex. The rats grew and reproduced normally on such diets. The above grains proved unsatisfactory as measured by the ability of females to rear their young. Mortality of the young was high on rations in which grains served as the sole source of vitamin B. From these experiments the authors concluded that a larger amount of vitamin B is required for lactation than for growth and reproduction. Taylor (126) found 73.5 per cent of white corn, yellow corn, barley, hulled oats, rye, or wheat in a purified ration to be unsatisfactory for rearing of young. If 10 per cent of grain was replaced by yeast the weight of the young at weaning was increased and the mortality was decreased. Ten per cent autoclaved yeast was found to be as effective as 10 per cent yeast in improving the lactation on rations containing 63.5 per cent of the following grains, white corn, yellow corn, barley, or hulled oats.

Miller (70) worked with yeast as the source of water soluble vitamin B. He found that females receiving a purified diet containing three per cent of yeast were able to rear young with an average weight of 33g, and a mortality of 50 per cent.
He found that the addition of 10 per cent autoclaved wheat embryo allowed females to wean young weighing 50-60g. at 33 days of age. The autoclaved wheat embryo employed by Miller did not support growth in young rats. Miller explained the improvement in lactation upon addition of autoclaved wheat germ as being due to the destruction of some toxic substance contained in wheat embryo and not due to an increase in the vitamin B complex content of the ration. From these data Miller concluded that an increased amount of vitamin B was not required for lactation. This view was contrary to the work of Guest and co-workers [33].

In 1928 Evans and Burr [24] reported some very interesting experiments concerning the amount of vitamin B required for lactation. Six-tenths grams of yeast daily did not allow a female to rear normal young to an age of 21 days. If the yeast was supplemented with tikitiki, eight drops daily, the young were weaned at 21 days with a normal weight. The supplement of tikitiki alone permitted females to wean about 68 per cent of their young, but the weaning weight was decidedly inferior. At the same time they reported that 15 per cent yeast, or about five times the amount required for normal growth was required for lactation. Ten per cent of rice polishings or 10 per cent of ether extracted wheat embryo, were unsatisfactory as a source of the vitamin B complex for lactation. Evans and Burr interpreted their results as demonstrating that the increase in yeast, or
tikitiki, improved lactation because of the antineuritic content of these substances. In the same year Evans and Burr (23) observed a paralysis in young produced by mothers receiving 15 per cent yeast, but deprived of vitamin E. This paralysis developed about the 19th. to 20th. day after birth. The incidence of paralysis was decreased by the addition of butter to the diet and prevented, if wheat germ oil was administered before the 15th. day. The above disturbance was due to a vitamin E deficiency according to Evans and Burr.

Macy and co-workers (66), working with a synthetic diet, in which yeast served as the source of the vitamin E complex, found three to five times as much yeast required for lactation as growth. The work of Evans and Burr (24) was in agreement with this work.

Nakahara and Inukai (67) found that females on a diet of 10 per cent fish protein, 75 per cent polished rice powder, 10 per cent butter, and 5 per cent McCollum's salt mixture supplemented with 5g. of brewers' yeast per 100g. of the above mixture grew normally but failed to suckle their young. These results differ from those of Evans and Burr (24); Macy et. al. (66), Guest et. al. (33), Taylor (126) and others who found that the mothers started out to suckle their young, but the young failed about the 14th. to 20th. day of life.

Soy beans were shown by Wilkinson and Nelson (128) to be deficient in some substance necessary for lactation. At the same
time soy beans at a 10, 20, 40, or 75.3 per cent level were
found satisfactory as a source of vitamins B and G, as judged by
their ability to promote growth in young rats. Liver from hogs
or cattle and hog kidneys were found to improve lactation on a
15 per cent soy bean ration, so as to allow females to rear their
young at a superior rate of growth. This factor contained in
liver and kidney was destroyed by heating at 120°C. for 12 hours
in an electric oven. It was not extracted with ether but was
contained in the liver residue after the ether extraction.

Five per cent of yeast was found to be the amount required
to furnish vitamins B and G for maintenance by Hussemann and
Hetler (46). They found yeast at a level of 15 per cent in-
sufficient for rearing of young. Twenty per cent of yeast was
the amount necessary to allow a female rat to rear a successful
litter. With tikitiki as a source of vitamin B and autoclaved
yeast as a source of vitamin G, Hussemann and Hetler (46) in-
vestigated the effect on lactation of the addition of vitamin
B or vitamin G to a ration containing five per cent yeast, the
maintenance level. They found that the addition of 3 drops daily
of tikitiki allowed mother rats to rear their young at a weight
of 36-39g. at 21 days. Five per cent yeast supplemented with
15 per cent autoclaved yeast was found to produce normal lac-
tation. Fifteen per cent autoclaved yeast as the sole source of
vitamins B and G did not allow females placed on such a ration
three weeks before mating to produce young. From their series
of experiments Hussemann and Hetler concluded that a diet must contain both vitamins B and G in order to promote lactation, and that an increase of either vitamins B or G above the maintenance requirement produced an improvement in lactation in the rat. They also postulated the existence of a possible quantitative relationship which exists between the requirements of the two vitamins.

Moore and Brodie (74), working with a ration in which the vitamin B complex was supplied as two per cent yeast, observed normal growth of rats to sexual maturity. They found, however, that the mortality of young born to females on this ration was 75 per cent before weaning time. Forty-five per cent of the mortality accrued during the first week after birth. The mammary glands were functioning as demonstrated by milk in the stomachs of the young at death. The young often developed paralysis of the limbs and a few had convulsive attacks preceded by a shrill squeal, followed by a rapid extension of the hind limbs. These workers found an increase of the yeast content of the diet to seven per cent prevented the occurrence of all but slight symptoms of paralysis, from which the young recovered. Moore and co-workers published a series of articles (75), (76), (77) on lactation, using, 10 per cent yeast, their stock diet, and combinations of vitamin B₁ and vitamin B₂ preparations. Their source of vitamin B₁ was an 85 per cent alcoholic extract of yellow corn; and their source of B₂ was a 20 per cent suspen-
sion of yeast autoclaved 3 hours at 15 pounds pressure. The young weaned by females on the various diets studied were of about equal weight at weaning. If the ability of a ration to promote lactation was measured by the per cent mortality, the synthetic diets were found inferior to 10 per cent yeast or their stock diet. The smaller young on the synthetic diets perished, causing the mortality of young on the synthetic diet to be high and increasing the average weight of the young weaned on these diets. Moore and co-workers (75) concluded that a third factor, probably Reader's vitamin B₄ was required for lactation in addition to vitamins B₁ and B₂.

In investigating the effect of optimal intake of vitamin G (B₂) as compared with the necessary requirements of vitamin G for growth, Sherman and Ellis (96) fed 2.5, 7.5, 15, and 30 per cent skimmed milk powder in the Sherman and Chase ration as a source of vitamin G; the above percentages of skimmed milk powder produced a diet containing 0.4, 0.9, 1.3, and 2.2 units of vitamin G, respectively, per gram of food mixture. The average weight at weaning of young whose mothers received the above rations increased in the following order for males, 33.9, 38.3, 47.4, and 51.0g. and for females, 33.6, 38.7, 44.4, and 48.4g., respectively. The ration containing 2.5 per cent of skimmed milk produced growth below the average rate. Seven and five-tenths per cent of skimmed milk powder supplied sufficient vitamin G for growth at the average rate.
Acetone-extracted wheat germ fed at a 20 per cent level was reported by Sure (113) to be unsuccessful as a ration for rearing of young rats. The addition of three per cent of the acetone extract permitted females to rear their young. The paralysis observed by Evans and Burr (23) substantiates the findings of Sure (113). In 1927 Sure published the first of a series of articles in a study of vitamin B requirements of the albino rat during lactation. An extract prepared by extracting wheat embryo with 75 per cent alcohol fed at a level equivalent to 45g. of wheat embryo per 100g. of ration was found inadequate as a source of the vitamin B complex for lactation. Sure (114) found it necessary to increase the amount of extract to 80g. per 100g. of ration, in order to wean young weighing 40g. at 28 days. In later papers Sure (111), (112) found three times the daily allowance of yeast required to produce continued growth in young rats was required for lactation if fed to the mothers. By feeding a portion of the yeast to the young Sure (112) was able to rear young with a smaller yeast supplement. From his experiments he concluded the mothers were about 40 per cent efficient in secreting vitamin B in the milk. In investigating the amount of wheat germ and rice polishings required to support successful rearing of young, Sure (110), (120) reported 30 per cent rice polishings to contain insufficient vitamin G and 20 per cent rice polishings to contain an insufficient amount of the anti-neuritis factor for rearing of young rats. Thirty per cent
wheat germ was found to support normal lactation. Young rats died with curd in their stomachs, showing the symptoms of beri-beri when 10 or 20 per cent of wheat germ served as the source of the vitamin B complex. Uncomplicated vitamin B deficiency was produced in suckling young by Sure and Smith (122), (123), and Sure, Kik and Walker (119). The symptoms exhibited by these young were hypoglycaemia, anhydremia, hemorrhages, and a disturbance in hematopoietic function. These symptoms were cleared up and growth resumed if a vitamin B concentrate was fed to the mothers.

Using the paired feeding technique, Sure and Walker (125) found 10 per cent yeast to be more satisfactory than 10 per cent autoclaved yeast in a synthetic diet for lactation. Sure and Smith (121) reported that vitamin B assists lactation due to a specific influence unrelated to the plane of nutrition in addition to the improvement due to increased food consumption. Sure, Kik, Walker, and Smith (118) in a further investigation of the specific effect of vitamin B on lactation found a vitamin B concentrate prepared by extracting rice polishings, adsorption on charcoal and elutriation with dilute HCl increased lactation in the rat independent of an increase in food or water intake.

Mapson (69) in investigating the lactation of rats on synthetic diets found a ration containing 10 parts wheat germ and 8 parts dried yeast plus milk inferior to the same ration containing a daily supplement of 2g. of fresh liver. The factor
responsible for the increased growth was not destroyed by heating; but vitamins B<sub>1</sub> and B<sub>2</sub> were destroyed under these conditions. The factor contained in liver was extracted from autolysed liver with hot water. In about 70 per cent of the cases mothers on the synthetic diet failed to suckle their young. In a further investigation of this factor Mapson (68) found the substitution of "Glaxo Casein" for "Light white casein" produced a more marked disturbance in lactation. With "Glaxo Casein" as the source of protein the mothers which started to suckle their young were unable to rear them. Rations containing "Light white Casein" permitted the mothers, which started to care for their young, to wean them. A preparation of "physin" was found to supplement the ration containing "Glaxo Casein". The "physin" was prepared from an aqueous extract of ox-liver by precipitation with 90 per cent acetone. The active "physin" was contained in the 90 per cent acetone soluble fraction.

Smith (103) found a ration containing 15 per cent liver protein supplemented with yeast gave poor lactation. In a continuation of the above study Smith and Seegers (104), (105) demonstrated that a ration containing 18.7 or 23.2 per cent of alcohol extracted liver supplemented with 0.5g. yeast daily was inadequate for lactation. Whole dried or fresh liver allowed mother rats to rear their young. This factor required for lactation is removed from liver by hot alcoholic extraction and is not contained in yeast.
The articles mentioned in this discussion conclusively demonstrate the need of an increased amount of some or all of the factors of the vitamin B complex during lactation. The exact amount of the different factors of the vitamin B complex required for lactation is an open question.

At the present time the only generally applied methods of assaying a material for water soluble vitamin content are biological methods. The test animals most commonly used in the assay of the water soluble vitamin B complex are rats and pigeons. Rats were employed as the test animals in all the experiments reported in this paper; therefore, only the basal rations and technique used in animal experiments employing rats will be discussed.

Steenbock and Sell (108) pointed out that twice as much cereal grain was required as a source of vitamin B to produce the same amount of growth on screens as was required if rats were allowed access to feces. Sherman and Spohn (100) emphasized the importance of keeping rats from eating their feces. Coprophagy is an important factor in vitamin assay as demonstrated by the work of Kennedy and Palmer (49). The decline in weight of rats on a vitamin B deficient ration was arrested by allowing access to their feces. Guerrant and Dutcher (31) and Moore, Flymate, and White (77) demonstrated that feces from vitamin B complex deficient rats supplement a vitamin deficient ration. The references cited along with other work, which space
does not permit to be reviewed, demonstrate the importance of preventing cephalophagy in vitamin studies.

A source of water soluble vitamin free protein for use in vitamin assay has received considerable attention. Sherman and Spohn (100) purified casein by washing with 60 per cent by weight alcohol. The casein used in the diets of Sherman and Chase (12) and Sherman and Bourquin (9) was purified in essentially the same way. Palmer and Kennedy (83), Chick and Roscoe (17) emphasized the importance of rigorous purification of casein by extraction with an alcohol water mixture in addition to leaching with water acidified with acetic acid. The importance of purifying casein for vitamin studies was further pointed out by Evans and Lepkovský (26). These workers also mentioned that starch and fat have sparing effects on vitamin B and emphasized the importance of using purified sucrrose as a source of energy in vitamin B studies. Hogan and Richardson (41) also found that sucrrose is more satisfactory than starch as a source of carbohydrates in the production of dermatitis.

The literature in which the early work in the field of the water soluble vitamin B complex is described is too voluminous to permit a review of it in this paper. For a discussion of this work the reader is referred to the excellent review by Mitchell (72) on the early phases of this work. Hofmeister (39) gave a detailed discussion of the symptoms of polyneuritis resulting from a lack of water soluble vitamin B. An excellent
discussion of a method of producing pellagra-like lesions in rats, receiving a diet deficient in vitamin B<sub>2</sub>, and the pathological developments in the skin was published by Finalely (27). The later phases of the work are admirably treated by Kruse and McCollum (54) and by Sherman and Smith (99). For a more recent review of the literature of vitamins B and G the reader is referred to the works of Simonnet (101), (102). The review of literature in this paper will be confined to those articles which have a direct bearing upon the work in this paper.

Considerable data had appeared in the literature prior to 1926 which suggested the dual nature of vitamin B. Smith and Hendrick (107) demonstrated that rolled oats at a level of 40 per cent in a synthetic diet failed to produce normal growth. Yeast autoclaved 6 hours at 15 pounds pressure supplied the substance necessary for growth not furnished by rolled oats; but autoclaved yeast by itself was unable to support growth in young rats. Very shortly after the work of Smith and Hendrick was published, Goldberger, Wheeler, Lillie and Rogers (29) and Goldberger and Lillie (38) reported data which showed the P-P factor in yeast was heat stable. Yeast which had been autoclaved was found to be effective in the treatment of black tongue in dogs and pellagra in human beings; but rats fed autoclaved yeast as a source of vitamin B died unless fed an extract prepared by extracting corn with 85 per cent alcohol, which did not support growth when fed as the sole source of the vitamin B com-
lex. The addition of the above evidence to the evidence presented by many other workers established the existence of at least two water soluble vitamins which are required for growth of the rat.

The Committee on vitamin B terminology (31) appointed by the American Society of Biological Chemists has recommended: that the term "B" be restricted to designate the more heat-labile (anti-neuritic) factor and, that the term "G" be used to denote the more heat-stable, water-soluble dietary factor, called the pellagra-preventive factor by Goldberger and his associates, and which also has to do with maintenance and growth. The terminology suggested above will be used throughout this thesis. The English workers (13) and those on the continent use the term "B_1" to designate the heat-labile antineuritic factor and the term "B_2" to designate Goldberger's heat-stable P-P factor necessary for growth and prevention of dermatitis in the rat.

In determining the vitamin G content of a material it is necessary to include in the diet a source of vitamin B comparatively free from vitamin G. Chick and Boscoe (16) found yeast to be a rich source of vitamin B and G. They found the 35 per cent alcoholic extract of either yeast or wheat germ contains the antineuritic factor but is deficient in the dermatitis-preventing factor. Munsell (78) found 30 per cent white corn to be a suitable source of vitamin B for use in vitamin G tests. Hunt (44) found a strong alcoholic extract of corn to be a potent
source of vitamin B. An 80 per cent by weight alcoholic extract of wheat is used by Bourquin and Sherman (9) in their diet used for vitamin G assays. Likewise Guirrant and Dutcher (31), Sandels (91), and several other workers have found concentrated alcoholic extracts of yeast or other materials to be a suitable source of vitamin B for vitamin G assays. The works of Smith (106), and Seidell (94) show ethyl alcohol in concentrations of 70 to 80 per cent to be a suitable solvent for extracting vitamin B without removing appreciable quantities of vitamin G. Eighty per cent ethyl alcohol was found by Stiebeling and Allerman (109) to extract considerable amounts of vitamins B and G; but the extract was more potent in B than G, due to oxidation of vitamin G in a strong alcoholic solution.

Fuller's earth was used by Seidell (92), (93) to adsorb vitamin B from a yeast extract, prepared by the method of Osborne and Waksman (82). Seidell also observed the active principle could be eluted with barium hydroxide. In an extensive study of the effect of pH upon adsorption of vitamins B and G by fuller's earth, Salmon, Guerrant and Hayse (90) found maximum adsorption of vitamin B at a pH of 4.5, and adsorption decreased with increase in pH. The maximum adsorption of the F-P factor, vitamin G, was found to be at a pH of 0.08 and decreased as the pH increased. They found fuller's earth to be a more efficient adsorbent for vitamin B than vitamin G. Sherman and Halliday (97) found 50 per cent of vitamin B and 33 per cent of vitamin G
contained in protein-free milk was present in the adsorbate, and that Lloyds' reagent was more efficient in adsorbing vitamin B than vitamin G. The International standard for vitamin B, (14), is a fuller's earth adsorption product; its vitamin B₂ content is negligible. Chick and Roscoe (17) and other English workers have found activated norite prepared by the method of Kinnersley and Peters (52), (53), or a modification of their technique, to be a source of vitamin B₁ (B) suitable for use in vitamin B₂ (G) tests.

Irradiation with ultra-violet light was reported by Hogen and Hunter (40) to be a means of preparing vitamin B free from vitamin G. Chick and Roscoe (15) and Kennedy and Palmer (48) found they could not rely upon irradiation to completely destroy vitamin G. Sure (124) met with success in preparing vitamin G-free rice polishings by irradiation with ultra-violet light. The importance of a high intensity of ultra-violet light in the destruction of the anti-dermatitis factor was emphasized by Hogen and Richardson (41).

The early success of Smith and Hendrick (107) and Goldberger et. al. (28), (29) stimulated research on the effect of heat on the vitamin content of yeast. Yeast autoclaved 5 hours at 120°C was found by Chick and Roscoe (16) to be unable to support growth as the sole source of the vitamin B complex; but if supplemented with an 85 per cent alcoholic extract of wheat germ it produced normal growth. Sherman and Axtmayer (95) found auto-
oclaved yeast was unable to support growth in young rats. Their autoclaved yeast supplemented wheat which is a rich source of heat-labile vitamin B. Guarrant and Solman (32) found the vitamin B contained in yeast was destroyed along with about 20 per cent of the vitamin G by heating 4 hours at 20 lbs. pressure. Autoclaving yeast was found by Williams, Waterman, and Gurin (131) not to be entirely satisfactory as a means of preparing a source of vitamin G free from vitamin B. They found vitamin B to be incompletely destroyed and vitamin G to be destroyed in considerable amounts by autoclaving 6 hours at 15 lbs. pressure. Yeast autoclaved at 15 lbs. pressure 2 to 6 hours was found by Chase and Sherman (12) to be a suitable source of vitamin G for vitamin B assays. They found that it is necessary, however, to test the particular sample of autoclaved yeast to be sure of its vitamin B and vitamin G content. Dry yeast heated at 95° to 100°C. for 2 to 4 weeks was found to retain most of its vitamin G content, according to the work of Block and Farquhar (7). Smith (106) believes autoclaved yeast, or autoclaved yeast extract, to be the only practical source of vitamin G nearly if not completely free from vitamin B.

The differences in solubility or extractibility of vitamins B and G in organic solvents have been used in the preparation of sources of vitamin G comparatively free from vitamin B. The extraction of yeast with 90 per cent alcohol in a Soxhlet extractor was found by Kennedy and Palmer (50) to leave considerable amounts
of vitamin G in the yeast residue. Chick and Roscoe (15) confirmed the work of Goldberger and co-workers (29) that the P-P factor is insoluble in strong alcohol, but they were able to account for only about 50 per cent of the vitamin G content in the yeast residue. They explained the decrease of vitamin G as being due to destruction of vitamin G by contact with the strong alcohol. Yeast that had been extracted with 95 per cent alcohol was found by Sherman and Sandels (98) to retain practically all of its vitamin G content; but extraction with 80 per cent alcohol was found to decrease the vitamin G content of yeast by about 30 to 35 per cent. Smith (106) found 70 per cent or higher concentrations of alcohol plus 1 per cent HCl are capable of removing 80 per cent of the vitamin B contained in dried Brewers' yeast without removing appreciable quantities of vitamin G. Methyl alcohol plus five per cent HCl was found to be unsuitable for the differential extraction of vitamin B and vitamin G, since it removed the two vitamins with nearly equal facility. Day (18) found yeast residue to contain as much vitamin G after extraction with 80 to 95.5 per cent acetone or 100 per cent methyl alcohol as before extraction. He found 60 per cent acetone or 60 per cent methyl alcohol to extract about one-half of the vitamin G contained in yeast. The solution obtained by extracting skim milk powder with 80 per cent alcohol acidified with 0.1M gallic acid was found by Stiebeling and Allerman (109) to contain about 30 per cent of the vitamin G contained in the original material.
However, if the 80 per cent alcohol was acidified with 0.1M HCl, the extract contained practically no vitamin G. They explain this difference as being due to the reduction potential of gallic acid, which prevents the destruction of vitamin G rather than to a difference in extractibility. Bocher (8) found vitamin G could be extracted from whey powder with hot 93 to 94 per cent ethyl alcohol, and the general properties of the vitamin G concentrate corresponded to the properties of lactochrome of whey.

Vitamin G, along with a small quantity of vitamin B, was shown by Solman and Guerrant (89) to be precipitated from an aqueous extract of velvet beans by the addition of sufficient alcohol, to give a concentration of 82.7 per cent alcohol. By making use of precipitation from acetone and alcohol containing hydriodic acid, Levene (59), (60) prepared a fraction which, when fed at a level of 0.001-0.002g. per day as a source of vitamin G, produced normal growth in rats. Narayanan and Drummond (80) were able to prepare a potent source of vitamin G by adding lead acetate to an aqueous or 50 per cent alcoholic extract of yeast. The lead precipitate was decomposed with H₂SO₄ and the active material precipitated by adding alcohol to give a concentration of 70 per cent alcohol. Precipitation with salts of heavy metals and adsorption on fuller's earth or norite have been used with considerable success in the separation of vitamins B and G. Lead acetate added to a yeast extract was used very effectively by Kinnersley and Peters (53), and Chick and
Rescoe (15) in separating vitamins B and G.

Since the definite establishment of at least two factors in the vitamin B complex, considerable research has been done on the distribution of vitamin G. Sherman and Axtemeyer (95) found dried skimmed milk was more potent in vitamin G than vitamin B, but the reverse was true of wheat. A rather extensive study of the distribution of vitamin $B_2$ (G) was reported (3) by Aykroyd and Rescoe. Of the foods tested they found dried liver and yeast to be the most potent in vitamin G. Wheat germ and wheat bran were found less potent; and the cereal grains, such as whole wheat and whole maize, were found to be poor sources of this vitamin. Guha (35) reported bakers' yeast to be a more potent source of $B_2$ (G) than brewers' yeast. He found beef muscle to be poor in $B_2$ (G) but ox-liver and ox-liver extract to be potent sources of vitamin $B_2$ (G). Two hundred and ninety mg. per day of dried beef liver produced an increase of 102g. per rat in 30 days, when fed as the sole source of vitamin G, according to data of Block and Farquhar (7). They also found a liver concentrate to be a potent source of vitamin G. Graham and Griffith (30) found one gram of dried liver daily as a source of vitamin B was unable to support normal growth; but as a source of vitamin G they found liver to be superior to yeast. Commercial whey powder was reported by Boocher (8) as containing 20 Sherman units of vitamin G per gram.

Hunt (44) observed that 25 per cent corn or wheat was able
to support growth in rats only at a slow rate. Likewise Munsell (78) found 50 per cent corn did not produce optimal growth as a source of vitamin G. If the vitamin G content of yeast is taken as 100, Munsell (79) reports the vitamin G content of wheat germ, wheat, and rice polishings to be 15, 5, and 4.5 respectively.

Gyorgy, Kuhn, and Wagner-Jauregg (37) report 0.2-0.4g. liver, 0.2-0.4g. kidney, 0.5-1.0g. yeast, 3.0-6.0g. e. c. cows milk, 3.5g. spinach, or 5.0g. of lettuce to furnish sufficient vitamin G to produce normal growth in young rats. Mitchell (71) using the paired feeding technique found six per cent of yeast was required to produce maximum growth in young rats.

Work on the further separation of the vitamin B complex has been a field for a large number of investigations in the last few years. Only brief mention of the different factors which have been postulated as existing can be mentioned here. Hunt (43) observed that yeast residue after extraction with water made weakly acid with acetic acid was necessary for optimal growth in rats. Small amounts of this factor were contained in the fuller’s earth residue after elution of the antineuritic factor with Ba(OH)₂. Further evidence for the existence of this factor was presented by Hunt and Wilder (45). The existence of a second thermolabile water-soluble factor was reported by Reader (86). She proposed a method of assaying this factor for which she proposed the name vitamin B₄ (87). Williams and Lewis (129) observed that a factor not extracted from yeast with 50 per cent
alcohol was necessary for normal growth in the rat. Pigeons require a second heat-labile vitamin for maintenance of weight according to the work of Williams and Waterman (130). Egg white which is a good source of vitamin $B_2$ (G) was found by Chick and Copping (13) to be lacking in some factor other than the antinutritive factor, which is required for normal nutrition of the rat. Guha (36) found a factor present in egg white not contained in alkaline autoclaved yeast. His results were in agreement with those of Chick and Copping (13). Halliday (38) found the basal diet of Sherman and Chase to be deficient in a factor which is stored by young rats for five to six weeks.

This factor is contained in red winter wheat and is probably identical with the vitamin $B_4$ reported by Reader. Peters (84) concluded from the evidence in the literature that there is a possibility of five factors in the vitamin B complex. Lewis (61) presents a very good review of the factors in the vitamin B complex other than vitamins B and G reported in the literature.

The isolation of vitamin $B_4$ in crystalline form was reported by Barns, O'Brien, and Reader (4). Kinnersley, O'Brien, Peters, and Reader (51) describe a technique for large scale preparation of vitamins $B_1$ and $B_4$. Tschesche (127) confirmed the report of Barns and co-workers (4). Kuhn, Gyorgy and Wagner-Jauregg (55), (57) isolated a group of food pigments from milk and eggs for which they proposed the name flavin. Lacto-flavin was found by Kuhn and co-workers (56) to be lacking in some fac-
tor other than the antineuritic factor necessary for growth in rats. Kuhn, Ruby, and Wagner-Jauregg (59) believed the identity of lacto-flavin and vitamin B₂ to be established. They proposed the formula C₁₇H₂₀N₄O₅ for lacto-flavin.
EXPERIMENTAL

The Sources and Treatment of the Materials used in the Experiments.

Casein

Casein was used at an 18 per cent level as a source of protein throughout the feeding experiments reported. Commercial casein, purchased from the Wilkins-Anderson Company of Chicago, Illinois, was purified in one of three ways. Casein used in experiments prior to ration number 206 was washed in battery jars with distilled water acidified with acetic acid. The water was changed daily for three weeks, and at the end of this time the casein was dried at 85°C. In experiments after number 212, the casein was further purified by extraction for five days in a continuous extractor with hot 60 per cent by volume ethyl alcohol acidified with acetic acid. This product was then dried and fed in all experiments after number 212. The casein used in rations 210 and 211 was washed with distilled water for three weeks and then dissolved in water by the addition of ammonium hydroxide. Acetic acid was added to precipitate the casein. The liquid was decanted off and the above process repeated twice. This casein was then dried at 85°C, and extracted with hot 60 per
sent ethyl alcohol for five days. Casein thus treated was dried at 85°C. and fed as reprecipitated casein.

**Dextrin**

Commercial corn starch obtained from the Penick and Ford Company, Cedar Rapids, Iowa was worked into a stiff paste with thirty-seven hundredths per cent citric acid solution. This paste was autoclaved at 15 pounds pressure for two and one-half hours. It was then dried at 85°C.

**Salt mixture 185**

McCullum and Simmonds (62) salt mixture 185 fortified with two grams of potassium iodide per three and one-half kilograms salt mixture was fed at a level of 3.7 per cent throughout these experiments.

**Yeast**

Dried yeast purchased from the Fleischmann Yeast Company, was employed as a source of water soluble vitamins.

**Autoclaved yeast**

Dried yeast, moistened with distilled water to form a thick paste, was spread in one inch layers in granite pans and autoclaved five hours at fifteen pounds pressure. This product was
dried and fed as autoclaved yeast. It supplied vitamin G.

**Butterfat**

College butter was melted in four liter beakers. The water and curd settled to the bottom and the fat was promptly decanted.

**Cod Liver Oil**

Squibbs cod liver oil was used as a source of vitamin A and D.

**Wheat, Barley, White Corn, and Yellow Corn**

Wheat, barley, white and yellow corn were purchased on the local market.

**Corn germ**

Corn germ was obtained from Penick and Ford, Cedar Rapids, Iowa.

**Rice bran, and Rice polishings**

The rice bran, and rice polishings were obtained from the Arkansas Rice Growers Corp. Assoc., Stuttgart, Arkansas.

**Wheat germ**

The wheat germ was purchased from General Mills, Minneapolis, Minnesota.
Hog Liver

All of the liver used in these experiments was furnished from the meat laboratory of Iowa State College, through the courtesy of Professor F. J. Beard. Fresh liver, secured within a few hours after the hogs were butchered, was worked up immediately. The first step in all of the hog liver preparations was the grinding of the liver in a food chopper.

Whey powder

The whey powder used in these studies was furnished by the Kraft-Phenix Cheese Co, Chicago, Ill.

Preparations from Wheat Germ

Ether extracted wheat germ

Wheat germ flakes were extracted in a continuous extractor with di-ethyl ether for 72 hours. At the end of this extraction the wheat germ was practically fat free. The ether was removed from the germ by drying at room temperature. This material is referred to as ether extracted wheat germ, or fraction I, throughout this paper.

Fractions IA, IB, IC, and ID

One kilogram of ether extracted wheat germ was stirred into three liters of 80 per cent ethyl alcohol and refluxed four hours. The alcohol was filtered off and two liters of fresh alcohol added
to the wheat germ and refluxed for one hour. This alcohol was filtered off and the extracted wheat germ dried at 65°C. The dried residue will be known as fraction 1A. The combined extracts were concentrated under reduced pressure, below 50°C, to one liter. One-half of this solution was dried on dextrin, so that one gram of dextrin was equivalent to one gram ether extracted wheat germ. The activated dextrin will be referred to as fraction 1B. Eighty grams of fuller's earth were added to the other half, and the suspension was mechanically stirred for one hour; it was then allowed to stand over night. The next morning the fuller's earth was filtered off. Twenty grams of fuller's earth were added to the filtrate in the same manner as the first eighty grams. This suspension was stirred one hour and allowed to stand overnight; it was then filtered. The two portions of fuller's earth were combined and air dried at room temperature then a vacuum desiccator over calcium chloride. This product will be referred to as fraction 1C. One gram of fraction 1C is equivalent to five grams of wheat germ. In all cases of adsorption with fuller's earth the above technique was followed, with such modifications as are specified. Fraction 1D was made exactly like fraction 1C, except one-half the amount of fuller's earth was employed; hence fraction 1D has twice the potency of 1C.
Fractions 2A and 2B

One kilogram of ether extracted wheat germ was extracted in a continuous extractor for 24 hours with 95 per cent alcohol. The distillation was performed at such a rate as to fill the extractor with fresh alcohol about 15 times in the course of 24 hours. The extracted material was dried and fed as fraction 2A. The extract was reduced in volume below 50°C to 200ml. and then dried on dextrin; one gram of dextrin was equivalent to two grams of ether extracted wheat germ. This product is called fraction 2B.

Preparations from Yeast

Fractions 3A, 3B, 3C, and 3D.

One kilogram of yeast was extracted with two liters of 95 per cent alcohol, by shaking by hand intermittently for four hours at room temperature. The alcohol was filtered off on a Buchner funnel. To the yeast residue two liters of fresh alcohol were added and the above process repeated. The above technique is similar to that used by Sherman and Sandels (98). The volume of the combined filtrates was reduced to 200ml. by vacuum distillation below 50°C. Five hundred milliliters of water were added to the concentrate and the volume again reduced to 200ml. Adsorption was then carried out by adding 80g. and subsequently 20g. of fuller’s earth. The combined fuller’s earth preparation constitutes fraction 3A. One gram of activated fuller’s earth,
fraction 3A, is equivalent to 10g. of yeast. The residue from
the alcoholic extraction was dried and will be known as fraction
3B. Five hundred grams of fraction 3B, the 95 per cent alcohol
extracted yeast, were further extracted with two successive one
liter portions of 60 per cent alcohol. The combined extracts
were dried on dextrin; one gram of dextrin was equivalent to
one gram of fraction 3B. This activated dextrin was fed as
fraction 3C. The yeast residue from the 60 per cent alcoholic
extraction was dried and fed as fraction 3D.

Fractions 4A and 4B

One kilogram of yeast was extracted with 95 per cent alco-
hol as previously described in the preparation of fractions 3A
and 3B, except the extraction was repeated seven times in place
of once. This yeast residue was dried and fed as fraction 4A.
The combined extracts were concentrated to 200ml., the pH ad-
justed to 4.5, and vitamin B adsorbed on fuller's earth; one
gram of fuller's earth was equivalent to 10g. of yeast. This
fuller's earth product was fed as fraction 4B.

Fractions 5A and 5B

Five hundred grams of yeast were wrapped in a cloth to pre-
vent packing and to allow the flow of alcohol through the mass
of the yeast. Extraction, in an automatic extractor, with 95
per cent alcohol was continued for 36 hours at a rate so as to
fill the percolator every two hours. At the end of the extrac-
tion the yeast residue was dried and fed as fraction 5A. The 
extract was dried on dextrin so that one gram of dextrin, frac-
tion 5B, was equivalent to two grams of yeast.

Preparations from Rice Polishings

Ether extracted rice polishings

Rice polishings were extracted, in a continuous extractor 
for 72 hours, with di-ethyl ether. The ether extracted rice 
polishings were air dried, and used as a starting material in 
the following preparations.

Fractions 6A, 6B, 6C, and 6D.

One kilogram of ether extracted rice polishings was extrac-
ted by stirring intermittently by hand in a battery jar with 
four liters of 25 per cent alcohol. The alcohol was filtered 
through a Buchner funnel and the residue was washed with one 
liter of 25 per cent alcohol. The above extraction and washings 
were repeated. The combined extracts and washings were con-
centrated to a syrup under reduced pressure the temperature being 
kept below 50°C; 95 per cent ethyl alcohol was added to the syrup 
to give a concentration of 80 per cent alcohol. The addition of 
alcohol produced a gummy brown precipitate, which was removed 
and dried on dextrin; one gram of dextrin was equivalent to five 
grams of ether extracted polishings. This product is fraction 6A.
The 30 per cent alcoholic solution was freed of alcohol and the pH adjusted to 4.5. fuller's earth was then added; one gram of fuller's earth was equivalent to 40 grams of rice polishings. This product was fed as fraction 6B. The residue from the extraction of rice polishings was dried and fed as fraction 6C. The filtrate from fraction 6B was dried on dextrin, so that one gram of dextrin was equivalent to five grams of rice polishings. This was fed as fraction 6D. Fractions 6A, 6B, 6C, and 6D were prepared by a technique similar to that employed by Evans and Lepkovsky (25).

Fractions 7A, 7B, and 7C.

One kilogram of ether extracted rice polishings was extracted by stirring by hand at thirty minute intervals for 12 hours with two liters of 95 per cent ethyl alcohol. After standing overnight the alcohol was filtered through a Buchner funnel. The above extraction was repeated twice, using one and one-half liters of alcohol in place of two. The volume of the combined extracts was reduced to 500ml. under reduced pressure, so that the temperature was kept below 50°C. Five hundred milliliters of water were added, and the volume again brought to 500ml. Neutral lead acetate was then added to give the maximum precipitate. The precipitate was filtered off and sulfuric acid added to remove the lead in the filtrate. The pH of the solution, freed from lead with sulfuric acid, was adjusted to 4.5; one-half of this
solution was dried on dextrin so that one gram of dextrin was equivalent to one gram of rice polishings. This formed fraction 7A. The vitamins in the other half were adsorbed on fuller's earth; one gram of fuller's earth was equivalent to ten grams of rice polishings. This is called fraction 7B. The rice polishings after extraction with alcohol were dried and fed as fraction 7C.

Fractions 8A, 8B, and 8C.

The extraction employed in the preparation of fraction 7A was repeated, except that three kilograms in place of one kilogram of rice polishings and three times the amount of solvent and reagents were employed. The extraction was also repeated five times in place of two times. The dried rice polishings residue from three kilograms of rice polishings weighed 2841g. It is called fraction 8A. The combined extracts were concentrated below 50°C to 500 ml. At this stage an oily brown material separated out and was removed and discarded. Fifty grams of fuller's earth were added to the solution, stirred mechanically for three hours, and allowed to stand in the ice box over night. The mixture was then filtered. To the filtrate 25g. more of fuller's earth were added. The combined activated fuller's earth weighed 80.2g. this was designated fraction 8B. One gram of fraction 8B equivalent to 37.46g. of rice polishings. The filtrate from the fuller's earth was dried on dextrin at 80°C, so that one gram
of dextrin was equivalent to five grams of ether extracted rice polishings. This constitutes fraction 9C.

Fractions 9A, 9B, and 9C.

Eighty grams of ether extracted rice polishings were extracted for 48 hours with 95 per cent alcohol in a Soxhlet extractor; the extractor was filled with alcohol and siphoned twice per hour. The rice polishings residue from this extraction was dried and fed as fraction 9A. The combined extracts from one kilogram of rice polishings were reduced in volume to 100 ml. At this point an oily material separated out and was discarded. Neutral lead acetate was then added to the vitamin solution, to give the maximum precipitate. The precipitate was filtered off and 20 per cent sulfuric acid added to the filtrate to precipitate the lead. The pH of the lead free solution was adjusted to 4.5, and the vitamin B adsorbed on 25 g. of fuller's earth. The fuller's earth after being dried in a vacuum desiccator weighed 26.7 g. This constituted fraction 9B, one gram of which was equivalent to 37.45 g. of rice polishings. The filtrate from the fuller's earth was dried on dextrin, so that one gram of dextrin was equivalent to two grams of rice polishings. This formed fraction 9C.

Preparation from Wheat

Fractions 10A, a wheat extract
Whole wheat was extracted according to the method described by Bourquin and Sherman (9). Two kilograms of ground wheat were stirred intermittently by hand with 3.7 liters of 80 per cent by volume alcohol. At the end of two hours the stirring was stopped and the mixture allowed to stand over night. The alcohol was then removed, and the wheat residue was extracted with 2.5 liters of 80 per cent alcohol. The alcoholic extract was filtered off; and the wheat residue was washed on a Buchner funnel with 0.75 liter of 80 per cent by volume alcohol. The combined extracts and washings were reduced below 50°C to a volume of 300 ml. The solution was dried on dextrin, so that one gram of dextrin was equivalent to five grams of wheat. This formed fraction 10A.

Preparations from Hog Liver

Fraction 11A

Two kilograms of ground hog liver were dried at 80°C in a drying oven. The dried liver fraction 11A weighed 583g. The liver dried at this temperature represented an average of 30.5 per cent of the fresh liver; one gram of fraction 11A was equivalent to 3.27g. of fresh liver.

Fractions 11, 11B, and 11C.

One kilogram of ground hog liver was stirred into two liters of boiling water and boiled for three minutes. The mixture was filtered on a Buchner funnel and the insoluble material washed
with 500 ml. of hot water. The liver residue was dried at 80°C. and weighed 233.0g. This residue was fed as fraction II B. The dried liver residue averaged 23.5 per cent of the weight of the fresh liver. One gram of fraction II B is equivalent to 4.25g. of fresh liver. The combined extracts and washings from one kilogram of fresh hog liver were concentrated in vacuo to 150 ml. This hog liver concentrate was used as a starting material in the following preparations and is referred to as fraction II. The process employed in extracting hog liver is similar to the process reported by Guha (35). One hundred fifty milliliters of hog liver concentrate, fraction II, was dried on dextrin; so one gram of dextrin was equivalent to two grams of fresh hog liver. This formed fraction III C.

**Fractions III D and III E.**

Ninety-five per cent ethyl alcohol was added to 150 ml. of fraction II to give a concentration of 50 per cent alcohol. This treatment caused the formation of a precipitate, which was removed and discarded. The concentration of alcohol was then increased to 70 per cent, by the addition of 95 per cent alcohol. A precipitate was formed which was filtered off and dried on dextrin; one gram of dextrin was equivalent to five grams of hog liver. This activated dextrin was fed as fraction III D. The filtrate was dried on dextrin; so one gram of dextrin was equivalent to five grams of fresh hog liver and fed as fraction III E.
Fraction 11F, 11F₁, and 11G.

Ninety-five per cent ethyl alcohol was added to 150 ml. of fraction 11 to give a concentration of 80 per cent alcohol. The precipitate thus formed was separated and dried on dextrin; so one gram of dextrin was equivalent to three grams of fresh hog liver. This activated dextrin was fed as fraction 11F. The filtrate from the 80 per cent alcoholic precipitation was dried on dextrin; one gram of dextrin was equivalent to five grams of fresh hog liver. This dextrin constituted fraction 11G. Fraction 11₁ was prepared in the same manner as fraction 11F; but one gram of fraction 11₁ was equivalent to six grams of fresh hog liver in place of three grams as in fraction 11F.

Fractions 11H and 11I.

Absolute ethyl alcohol was added to 150 ml. of fraction 11 to give a concentration of 90 per cent alcohol. The precipitate was removed and dried on dextrin; so that one gram of dextrin was equivalent to three grams of fresh hog liver. This process formed fraction 11H. The filtrate was dried on dextrin; so that one gram of dextrin was equivalent to five grams of fresh hog liver. This dextrin was fraction 11I.

Fraction 11J

One hundred fifty milliliters of acetone were added to 150 ml. of fraction 11. This addition of acetone caused the formation
of an oily brown liquid. This liquid was separated, dissolved in 100 ml. of water, and then dried on dextrin; one gram of dextrin was equivalent to two grams of fresh hog liver. This dextrin was designated as fraction 11J.

Fraction 11K

To 150 ml. of fraction 11 600 ml. of acetone were added in order to produce a concentration of approximately 80 per cent by volume of acetone. A dark oily brown liquid separated. This liquid was removed, dissolved in 100 ml. of water, and dried on dextrin; one gram of dextrin was equivalent to five grams of fresh hog liver. This process formed fraction 11K.

Fraction 11L

One hundred fifty milliliters of fraction 11 were diluted with water to 250 ml. and fed as fraction 11L. One milliliter of fraction 11L was equivalent to four grams of fresh hog liver.

Fractions 12A and 12B

Fraction 11B, dried water-extracted liver, was extracted in a Soxhlet extractor with 60 per cent alcohol for eight hours. The extraction was performed at such a rate as to siphon every 25 minutes. The alcohol insoluble material left after this treatment was 81.5 per cent of the original material (fraction 11B). This material was fed as fraction 12A. One gram of
fraction 12A was equivalent to 1.227g. of fraction 11B or 5.221g. of fresh hog liver. The alcoholic extract was dried on dextrin, so that one gram of dextrin was equivalent to one gram of fraction 11B or 4.255g. of fresh hog liver. The activated dextrin was fed as fraction 12B.

Fractions 13 and 13A

Hog liver was extracted by a modification of the method employed by Mapson (69). Minced fresh hog liver was mixed with one liter of water per kilogram of liver. This mixture was made acid to congo red paper so as to have an approximate pH of 4.5, by the addition of 20 per cent sulfuric acid. The mixture was then allowed to stand 36 hours at 30°C. At the end of this time one liter of water per kilogram of liver was added. The mixture was boiled three minutes and filtered through a Buchner funnel. The residue was washed with 500 ml. of hot water per kilogram of liver. The extracts and washings were combined and reduced in volume to 200 ml. per kilogram of liver. This concentrate will be referred to as fraction 13 in the following experiments. The insoluble residue was dried at 80°C and fed as fraction 13A. One gram of fraction 13A was equivalent to 6.05g. of fresh hog liver.

Fraction 13B

Two hundred milliliters of fraction 13 were dried on dextrin;
one gram of dextrin was equivalent to two grams of fresh hog liver. The activated dextrin constituted fraction 13B.

Fractions 13C and 13D

Ninety-five per cent ethyl alcohol was added to 200 ml. of fraction 13 to produce a concentration of 80 per cent alcohol. A precipitate was formed at this point, which was removed and dried on dextrin; so that one gram of dextrin was equivalent to three grams of fresh hog liver. This was called fraction 13C. The filtrate from the above precipitation was dried on dextrin; so that one gram of dextrin was equivalent to five grams of fresh hog liver. This dextrin is designated fraction 13D.

Fractions 13E and 13F

Ninety-five per cent alcohol was added to 200 ml. of fraction 13 to give a concentration of 50 per cent by volume of alcohol. The precipitate thus formed was removed and dried on dextrin; so that one gram of dextrin was equivalent to five grams of fresh hog liver. This is fraction 13E. The 50 per cent alcoholic filtrate was dried on dextrin; so that one gram of dextrin was equivalent to three grams of fresh hog liver. This dextrin formed fraction 13F.

Preparations from Whey Powder

Fractions 14A and 14B
Whey powder was extracted in a manner similar to the process employed by Booher (3). Five hundred grams of whey powder were successively extracted with 2000 ml., 1250 ml., and 750 ml. of boiling 95 per cent by weight alcohol for 30 min., 30 min., and 15 min., respectively, with a stream of purified nitrogen bubbling through the mixture. The extracted whey powder was air dried. It weighed 441.0g. and was fed as fraction 14A. The combined extracts were concentrated under reduced pressure to about 1000 ml., and then were further concentrated in a vacuum desiccator almost to dryness. The dried extract was then dissolved in about 150 ml. of water and dried on dextrin at 80°C; so that one gram of dextrin was equivalent to two grams of whey powder. This activated dextrin was fed as fraction 14B.

Care of Animals

The rat was used as the test animal in all feeding experiments reported in this thesis. Rats were kept in cages 12" X 24" X 10". A group of six rats was usually kept in one cage, if the rate of growth was being studied. In case of the ability of females to rear their young was being studied, the female and her litter were placed in a cage by themselves. The animals were allowed shavings as litter in certain experiments; in other experiments they were kept on a false bottom galvanized screen to prevent coprophagy.
Young vigorous rats four to five weeks of age and weighing 45g. to 55g., were placed in groups of six in a cage on false screen bottoms. These rats were fed the basal ration plus tap water for a period of three weeks. At the end of three weeks the rats had started to lose weight, due to a depletion of the body stores of the vitamin B complex. These rats were then used to determine the vitamin B and vitamin G content of materials being studied.

Females used in the study of the vitamin B and vitamin G requirements in the maternal diet for successful rearing of young were obtained in two decidedly different ways. The data summarized in tables VII and VIII were obtained from females reared on the experimental ration from the time they were four to five weeks of age. All other lactation data were obtained from females transferred from the stock diet to the experimental diet at parturition. The females were allowed shavings for litter in experiments up to and including lot 193 during the full lactation period. In experiments after 193 the females were allowed shavings for litter the first seven days after parturition, and were then placed on two mesh per inch false screen bottoms.

In some of the experiments materials tested for vitamin B or vitamin G were incorporated in the basal ration, replacing a corresponding amount of dextrin. In other experiments the material being tested was fed separately in 150 ml. beakers.
as a daily supplement except Sunday.

The basal ration used throughout these studies contained: casein 18 per cent, salts (185) 3.7 per cent, cod liver oil 1.0 per cent, butterfat 4.0 per cent, and dextrin 73.3 per cent. The dextrin content of the rations was correspondingly reduced in the experiments, in which the material being tested for vitamin B or vitamin G was incorporated in the ration. For example, if ten per cent yeast was fed as a source of vitamin B and vitamin G, the dextrin content was reduced from 73.3 per cent to 63.3 per cent.

Animals used in vitamin B tests received the basal ration plus a source of vitamin G, which was sufficiently free from vitamin B to cause a loss of weight during the eight weeks of the test. The above conditions make it possible to assume the growth-limiting factor to be vitamin B in the above ration, and any growth increase, when supplemented with a material being tested, to be due to the vitamin B carried by this material. The vitamin B units per gram of material could be calculated according to the following equation. This is a modification of the Chase and Sherman (12) definition of a vitamin B unit.

\[
\text{Avg. gain grams per week} \div \frac{\text{3 (daily supplement of material tested)}}{\text{units vitamin B per gram of material tested}}
\]

The technique used in vitamin G studies was the same as
that used for vitamin B, except a source of vitamin B, deficient in vitamin G, was supplied along with the basal ration; so that the growth-limiting factor was vitamin G. The vitamin G units per gram of material tested could be calculated according to the following equation.

\[
\text{Avg. gain grams per week} - \frac{\text{Avg. gain grams per week correction}}{3} \text{ (daily supplement of material tested)}
\]

= units vitamin G per gram of material tested.

That part of the equation corresponding to "Average gain grams per week correction", is necessary because the basal ration plus the vitamin B supplement constantly produced a small gain. In order to determine the gain due to vitamin G in the material being tested, it was necessary to subtract the gain due to the vitamin G in the basal ration plus vitamin B supplement.

Some definite criterion for the measurement of the success of rearing of young is essential, in order to establish the minimum vitamin B and vitamin G content of the maternal diet required to permit normal rearing of young. In this thesis an arbitrary standard of weight of 45g. per young rat at 28 days and a mortality of 10 per cent, or less, was chosen as normal. The performance of females on a satisfactory stock ration meets these requirements readily. The relative efficiency of the different foods could then be determined by the amount of the deviation from the normal.
Vitamins B and G Experiments

Wheat germ and wheat germ preparation as sources of vitamins B and G.

The data obtained from feeding experiments with wheat germ preparations are summarized in Table I. Ration 102, negative control, and ration 99 containing 10 per cent autoclaved yeast, were sufficiently free from vitamin B to cause the death of all rats before the end of the eighth week on the experiment. Lots 102 and 99 demonstrate that the basal ration and basal ration plus autoclaved yeast are sufficiently free from vitamin B for use in testing the content of the materials used in this work. Two-tenths of a gram of fraction 10 per rat per day furnished enough vitamin G to produce a gain of 2.7g. per week and sufficient vitamin B for a gain of 7.5g. per week, according to the data of lots 96 and 97. Fraction 1A, fed to lot 101 at a level of 1.0g. per day per rat, produced an average gain of 7.6g. per week. These data show that extraction with 80 per cent alcohol does not remove vitamins B and G quantitatively. The animals in lot 103 received 1.0g. of fraction 1B per rat per day and were able to make an average weekly gain of 2.4g. The limiting factor in ration 103 was vitamin G; therefore, fraction 1B contained 0.81 units of vitamin G per gram.

Five per cent wheat germ supplied sufficient vitamins B and
G for good growth; but it seems to be more potent in vitamin B than vitamin G, as indicated in lots 119 and 120. Vitamin B was removed quantitatively from wheat germ, by extraction with 95 per cent alcohol in a continuous extractor for 24 hours, according to the data of lots 130 and 131. Some vitamin G was left in the extracted wheat germ, fraction 2A, as shown by an average weekly gain of 2.2g. in lot 132, in which vitamin G was the limiting growth factor. Five per cent of fraction 2B (lot 133) did not contain sufficient vitamin B and vitamin G to cause a gain in weight over an eight week period, but it did maintain life and body weight in five of the six rats. Lot 134 shows that fraction 2B contains 5.9 units of vitamin B per gram. Lot 135 indicates fraction 2B to contain 1.3 units of vitamin G or about one fifth as much vitamin G as vitamin B. The data presented in Table I show wheat germ to contain 7.3 vitamin B units and 6.4 vitamin G units per gram. Furthermore, extraction of wheat germ with 80 per cent alcohol and adsorption on fuller's earth produces an adsorbate about three times as potent in vitamin B as in vitamin G.

Yeast and preparations from yeast as a source of vitamin B and G

A summary of the feeding experiments using yeast is presented in Table II. It was necessary to have a source of vita-
min B, free from vitamin G, or of a small but known vitamin G content, to use in these studies. Two-tenths of a gram of fraction 15, with a vitamin G content sufficient to induce a gain of 2.7g. per week, was fed as a source of vitamin B in testing for vitamin G in the series of experiments reported in Table II. Two and seven-tenths grams were subtracted from the average gain per week, before making the calculation of vitamin G content.

Lot 100 shows that 1.0g. of dried yeast fed daily supplies sufficient vitamin B and vitamin G to produce a gain of 12.6g. per week. The vitamin B content of the same yeast was reduced by autoclaving to such a level, that it caused the death of all animals (lot 99) which received 1.0g. of autoclaved yeast per day. Extraction of yeast with 95 per cent alcohol and adsorption on fuller's earth produced a material about four and one-half times as potent in vitamin B as vitamin G, according to the data in lots 104 and 105. A comparison of lots 106 and 107 show that extraction with 95 per cent alcohol removes vitamin B more effectively than vitamin G from dried yeast. These data indicate a selective extraction of vitamin B from yeast with 95 per cent alcohol.

Sixty per cent ethyl alcohol was a good solvent for vitamin G as shown by a gain of 7.8g. per week, when the extract from 1.0 gram of fraction 33 was fed as the source of vitamin G in lot 109. Sixty per cent alcohol did not seem to be a better solvent than 95 per cent alcohol for the extraction of vitamin B,
as evidenced by a gain of only 2.5g. per week in lot 108. The data from lots 171 and 172 demonstrate that fraction 4A, consisting of the residue from eight 95 per cent alcohol extractions of yeast, contains 1.7 units of vitamin B and 3.8 units of vitamin G per gram. Fraction 4B, the activated fuller's earth prepared from the extract, contains 18.2 units of vitamin B and 5.8 units of vitamin G per gram, according to lots 173 and 174.

Fraction 5A, the yeast residue left from extraction with 95 per cent alcohol in a percolator, is a good source of both vitamins B and G, as shown by lots 175 and 176. Fraction 5B, the extract, contains no vitamin G according to the data of lot 177 and only a small per cent of the vitamin B contained in the yeast. The poor extraction of vitamins B and G with 95 per cent alcohol in a percolator was probably due to the nature of the yeast, which packed very tightly in the extractor and prevented the penetration of the solvent.

Rice polishings and preparations from rice polishings as a source of vitamins B and G.

Data obtained by feeding rice polishings and various preparations from rice polishings are presented in Table III. Ration 121, containing 5 per cent rice polishings, produced a gain of 4.9g. per week; while ration 122, containing 5 per cent polishings plus 5 per cent autoclaved yeast, produced an average
weekly gain of 12.0g. These experiments indicate the limiting faster for growth to be vitamin G. Rice polishings are about one and seven-tenths times as potent in vitamin B as vitamin G, according to these data. The animals on ration 123 made an average gain of 2.8g. per week, while those on ration 124 made an average gain of 3.5g. per week. Fraction 6B was therefore about three times as potent for vitamin B as vitamin G; which indicates that the vitamin B in the original material had been concentrated in fraction 6B to a greater extent than vitamin G. The gummy brown precipitate which appeared when the concentration of alcohol reached 30 per cent contained only a slight amount of vitamin B, as indicated by the small growth in lots 125 and 126. Extraction with 25 per cent alcohol did not remove vitamin B or vitamin G quantitatively from rice polishings, as demonstrated by a growth of 5.8g. and 7.4g. per week in lots 127 and 128 respectively.

Fraction 7B fed at the rate of 0.1g. per rat per day supplied sufficient vitamin G to produce a gain of 0.8g. per week and sufficient vitamin B to produce a gain of 9.6g. per week, according to the data of lots 163 and 164. Fraction 7B, the activated fuller's earth prepared by adsorption of the vitamins contained in the 95 per cent alcoholic extract of rice polishings, was at least ten times as rich in vitamin B as vitamin G, according to data from lots 163 and 164. Extraction of rice polishings with 95 per cent alcohol and drying the extract
on dextrin produces a product, fraction 7A, which, according to experiments 165 and 166, is three times as potent in vitamin B as vitamin G. The 95 per cent alcohol extracted rice polishings, fraction 7C, contains considerable vitamin B and vitamin G, as indicated by a growth of 9.8g. per week in lot 164. The addition of fraction 7A to fraction 7C did not improve the growth of the rats on ration 168 over the growth of those on ration 167. This indicated the growth limiting factor in fraction 7C to be vitamin G and not vitamin B. The animals on ration 169, which contained 10 per cent of fraction 7C plus 10 per cent of autoclaved yeast, made a gain of 15.3g. per week, showing fraction 7C to be a potent source of vitamin B. The autoclaved yeast is practically vitamin B free as evidenced by the 63.3 per cent mortality of the rats on ration 170. The experiments from 163 to 170 show that extraction with 95 per cent alcohol removed vitamin B more readily than vitamin G from rice polishings. This separation of vitamin B and vitamin G seems to be carried further by adsorption on fuller's earth. It appears evident from the above series of experiments that three extractions with 95 per cent alcohol does not remove vitamin B completely from rice polishings.

Experiments 184 to 188 were conducted, in order to determine if doubling the number of extractions would remove vitamin B more completely from rice polishings. Fraction 8B contains 94.3 units of vitamin B and 26.4 units of vitamin G per gram, as seen
in lots 184 and 185. The alcohol extracted rice polishings residue, fraction 8A, contained 4.5 and 4.0 units per gram, respectively, of vitamins B and G, according to lots 186 and 187. The above data indicate that increasing the number of extractions from three to six does not markedly increase the amount of vitamin B extracted. Experiment 194 gave a growth of 4.1g. per week, with 0.027g. of fraction 8B serving as the only source of vitamins B and G. In lot 195 the 0.027g. of fraction 8B was supplemented with 10 per cent of autoclaved yeast. The animals on this ration made an average gain of 7.1g. per week.

An examination of the growth obtained by feeding materials such as fraction 8B, fraction 7B, fraction 7A, or fraction 6B, which one would expect from data in the literature to be potent sources of vitamin B almost free from vitamin G, reveals a gain of two to four grams per rat per week when fed as the sole sources of vitamins B and G. The amount of growth was not increased by doubling the amount of supplement, as in lots 184 and 194. The animals in lot 184 received 0.053g. of fraction 8B and those in lot 194 received 0.027g. of fraction 8B per day; but the gain was 4.2g. per week in lot 184 and 4.1g. per week in lot 194.

The above data suggested that the basal ration might contain some vitamin G. Cassin leached with acidified water has been reported by several investigators (17), (26), and (49) to contain appreciable amounts of the water soluble vitamins.

In experiments 206 to 211 inclusive, a study was made of the
effect of purifying casein by different methods on the ability of fraction 3B to produce growth as the sole source of vitamins B and G. The source of proteins in rations 206, 207, and 210 was 18 per cent of washed casein, alcohol extracted casein, and reprecipitated casein, respectively. All rats placed on rations 206, 207, or 210 died before the end of the tenth week on the ration. Ration 208 contained alcohol extracted casein plus 0.027g. of fraction 3B. Three of the animals on ration 208 died before the end of the seventh week on the supplement. As soon as one of the animals died the other animals consumed it. After one animal died and was consumed, the surviving animals made a gain of 6 to 10 grams the next week. This suggests they were obtaining vitamin G by eating the animals which had died with vitamin G deficiency. The rats on ration 209, which contained alcohol extracted casein plus 10 per cent autoclaved yeast, had a mortality of 83.3 per cent. The one rat that survived the eight week test lost 14g. Ration 211 contained reprecipitated casein plus 0.027g. of fraction 3B. Rats on ration 211 had a mortality of 33.3 per cent and an average gain of 3.09g. per week. The results of experiments 208 and 211 show that alcohol extracted casein produces about the same amount of growth as reprecipitated casein. A comparison of the growth rates and mortality of the animals in lots 211, 208, 194, and 194 indicates alcohol extracted casein contains a smaller amount of vitamin G than casein washed only with water acidified with
acetic acid. Due to these results, it was decided to extract all casein used in further experiments with 60 per cent alcohol.

Fraction 9B, activated fuller's earth prepared from the extract obtained by extracting rice polishings in a Soxhlet, was shown in lots 212 and 213 to contain almost ten times as much vitamin B as vitamin G. The animals in lot 214, receiving 10 per cent of fraction 9A, made no gain; showing the rice polishings residue to be practically free from either vitamin B or vitamin G, or both. The data obtained from lot 215 indicate that fraction 9A is practically vitamin B free, because it does not supplement 10 per cent autoclaved yeast. Fraction 9C contains little or no vitamin B, as evidenced by the failure of animals in lots 216 and 217 to make a gain in weight and the death of five out of six animals in lot 217. These data indicate that adsorption on fuller's earth removes vitamin B quantitatively.

Rice polishings supported a gain of 6.9g. per week, when fed at a level of 10 per cent, in lot 229. They furnished sufficient vitamin B to produce a gain of 17.5g. per week, if two per cent of fraction 11A, dried hog liver, was added to assure an abundance of vitamin G in lot 230. From the data of lots 229 and 230 it is evident that rice polishings are richer in vitamin B than in vitamin G. The animals in lot 231 were placed in individual cages and fed 0.055g. of fraction 9B per rat per day as the sole supplement of vitamins B and G. They made a
gain of 2.0g. as compared to a gain of 3.2g. in lot 232, which received 10 per cent autoclaved yeast in addition to 0.053g. fraction 8B. Lot 265 received 0.053g. fraction 8B per day plus 10 per cent autoclaved yeast. The average gain per week was 14.5g. Ration 266 was the same as 265, except it contained 10 per cent of fraction 8C. The gain in lot 266 was only 11.7g. per week; therefore, it was proven that fraction 8C does not supplement 0.053g. of fraction 8B plus 10 per cent autoclaved yeast. The data from lots 267 and 268 show that fraction 8C contains only a small amount of vitamin G.

Eighty per cent alcohol extract of wheat

Fraction 10A, incorporated in the ration at a 10 per cent level as the sole source of vitamins B and G, produced an average gain of 2.4g. per week in lot 251. Ten per cent of fraction 10A plus 10 per cent autoclaved yeast produced a gain of 13.2g. per week in lot 252. The above data show that fraction 10A contains about four times as much vitamin B as vitamin G.

Source of vitamin B to use in vitamin G studies

All vitamin B preparations tested produced a small and fairly constant increase in weight of the rats, which received the basal ration plus the vitamin B preparation, as the sole source
of vitamins B and G. Since it has been impossible to find a suitable source of vitamin B, which would produce no gain when fed along with the basal ration, it was decided to use fraction 83 as a source of vitamin B in studying the vitamin G content of materials. In making the calculations of vitamin G content of materials tested, it was necessary to subtract 2.5g. from the average gain per week, in order to correct for the vitamin G contained in the basal ration plus 0.058g. of fraction 83.

Liver and preparations from liver as sources of vitamin B and G

Liver was found by Wilkinson and Nelson (128) to supplement soy beans in rearing of young rats. Guha (34) has demonstrated that liver is a potent source of vitamin G relatively free from vitamin B. These experiments suggested the possibility of using liver as a starting material in the preparation of vitamin G to be used in lactation studies.

Animals in lot 196, Table IV, received 0.1g. of fraction 11A, dried hog liver, per day and made an average gain of 9.0g. per week. Three-tenths of a gram of fraction 11A furnished sufficient vitamin G in ration 197 to produce an average gain of 11.5g. per week. As a source of vitamin B 0.3g. fraction 11A, lot 200, produced a gain of 4.6g. per week. A comparison of lots 197 and 200 shows the ratio of vitamin G to vitamin B in dried hog liver to be two and one-half to one. If lot 196
be compared to lot 200, the ratio of vitamin G to vitamin B is six to one. The latter ratio is probably more nearly correct, because the growth rate in lot 196 was normal or above; so a threefold increase in vitamin G would not be expected to produce a threefold increase in the growth rate.

Fraction 11C contained 10.0 units of vitamin G per gram or about one-half of the vitamin G contained in the liver, as demonstrated by lot 198. The animals in lot 199 make an average gain of 2.5g. per week, which proved that fraction 11C contained some vitamin B. Fraction 11B, the liver residue left after extraction of liver with hot water, furnished sufficient vitamin G, when fed at a level of 0.3g. daily, to produce a gain of 8.8g. in lot 201. Rations 219 and 220 show that fraction 8B fed at a level of 0.063g. per day furnished sufficient vitamin B to produce a gain of at least 10g. per week and sufficient vitamin G to produce a gain of 3.4g. or less per week.

The animals in lot 221 receiving 0.325g. of fraction 11D per day were unable to survive the eight weeks test period. The animals in lot 222 which received 0.325g. of fraction 11D plus 0.027g. of fraction 8B made an average gain of 4.61g. per week. Fraction 11E contains 2.6 units of vitamin B and 11.1 units of vitamin G per gram as shown by the data from lots 223 and 224. A comparison of the rate of growth in lots 222 and 224 indicates that 70 per cent alcohol precipitates more than one-fourth of the vitamin G contained in the aqueous extract of liver.
Fraction 11F contains 2.9 units of vitamin B and 4.7 units of vitamin G per gram, according to the data of lots 257 and 258. The vitamin G extracted from one gram of hog liver, fraction 11, is 5.0 units, which indicates that fraction 11F contains about one-third of the vitamin G contained in the extract. Fraction 11G contains about one-eighth of the vitamin G contained in the liver extract, according to lot 259. From the data of lots 258 and 259, it appears that 80 per cent alcohol precipitates about two and one-half times as much vitamin G as is left in solution. Five-tenths of a gram of fraction 11H daily produced an average gain of 1.7g. per week in lot 260. The same amount of fraction 11H as a source of vitamin G in lot 261 produced a gain of 7.3g. per week, which showed fraction 11H contained 5.2 units of vitamin G per gram. The data from lot 262 show fraction 11I contained 0.66 units of vitamin G per gram. A comparison of the data in lots 261 and 262 demonstrates that 90 per cent alcohol precipitated about nine times as much vitamin G as remained in solution. The above series of experiments demonstrates that as the concentration of alcohol was increased the amount of vitamin G removed by precipitation from a solution of hog liver extract increased; and the amount remaining in solution decreased.

In rations 263 and 264, fraction 10A, the alcoholic extract from whole wheat was fed as a source of vitamin B, in order to compare the vitamin G content of fraction 11F with the vitamin
G content of autoclaved yeast. From these data it appears that 0.641g. of fraction 11F per rat per day supplies slightly less vitamin G than 10 per cent autoclaved yeast mixed into the ration. Five of six animals placed on fraction 11F, lot 289, developed polynuertis and died. Lots 289 and 290 show fraction 11F contains between 10 and 15 units of vitamin G per gram. Each animal in lot 255 received 0.25 ml. of fraction 11L daily directly from a pipette. The animals in lot 256 received the same amount of fraction 11L plus 0.053g. daily of fraction 83. The animals in lot 256 gained about twice as fast as those in lot 255, indicating fraction 11L to be twice as potent in vitamin G as vitamin B.

The data from lots 225 and 226 show that 0.50g. of fraction 11J did not furnish sufficient vitamin B to maintain life throughout the eight week experimental period; but it did supply enough vitamin G to produce an average gain of 5.4g. per week. Eighty per cent acetone precipitated sufficient vitamin B from a liver extract to enable 0.2g. of fraction 11K to produce an average gain of 3.4g. per week in lot 227 and sufficient vitamin G to produce a gain of 8.3g. per week in lot 228. A comparison of the data presented in lots 225, 226, 227, and 228 shows that as the concentration of acetone was increased, the amount of vitamin G and vitamin B contained in the precipitate from a hog liver concentrate were increased.

The animals in lot 253 made an average gain of 3.2g. per
week. This gain shows that 60 per cent alcohol extracted some vitamin G from fraction 11B. The animals in lot 254 received 0.4g. of fraction 12A as a source of vitamin G. These animals made an average gain of 5.6g. per week. The above data demonstrate that extraction of fraction 11B with 60 per cent alcohol in a Soxhlet-extractor for eight hours did not remove vitamin G quantitatively.

Five-tenths of a gram of fraction 13B was fed as the source of vitamin B in ration 234. The rats on this ration made a gain of 3.7g. per week; this showed fraction 13B contained 2.4 units of vitamin B per gram. The data in lot 235 indicate that fraction 13B contained 7.2 units of vitamin G per gram. The animals in lot 237 received 0.33g. of fraction 13A as a source of vitamin G. They made an average gain of 9.5g. per week. An examination of the data obtained in lots 234, 235, and 237 proves that the method of autolysing and extracting hog liver used removed about as much vitamin G as was left in the hog liver residue, fraction 13A. Fraction 13C, fed as a source of vitamin G at a level of 0.35g. per day, produced a gain of 8.6g. per week in lot 236. In lot 238 the rats received 0.2g. of fraction 13D daily as a source of vitamin G and made a gain of 4.5g. per week. The above data indicate that 30 per cent alcohol precipitated about twice as much vitamin G from an extract of hog liver as was left in solution. The rats in lot 239 received 0.20g. of fraction 13E per day as a source of vitamin G and made
a gain of 0.1g. per week. Rats in lots 240 received 0.35g. of
fraction 13F per day and made a gain of 11.3g. per week. The
above data show that 50 per cent alcohol precipitates prac-
tically no vitamin G from a liver extract.

Ten per cent yeast in lot 242 produced a gain of 14.6g. per
week. A comparison of lots 235 and 242 reveals the fact that
the extract from one gram of hog liver fed in the form of 0.50g.
of fraction 13B per day supplied sufficient vitamin G to pro-
mote an average gain in weight almost equal to the gain in
weight of rats which received 10 per cent of dried yeast mixed
into the ration.

Whey powder and whey powder extract as sources of vitamin G.

Whey powder was found to contain 11.7, 6.1, and 12.3 units
of vitamin G per gram respectively in lots 282, 283, and 284.
The average is 10.03 units of vitamin G per gram. Fraction 14A
contains 6.4 and 3.9 units of vitamin G per gram, according to
results of lots 305 and 304, or an average of 5.62 units per
gram. The 95 per cent alcoholic extraction of whey powder, as
revealed by comparing the growth rates in lots 305 and 306 with
the growth rates in lots 283 and 282, respectively. Fraction
14B contains the extract from two grams of whey powder dried on
one gram of dextrin. The data in lots 305 and 306 show that
fraction 14B contains about ten units of vitamin G per gram. This is about one-half of the vitamin G contained in the whey powder, from which fraction 14B was prepared.

Lactation Studies

The rations employed in all of the lactation studies consisted of casein 18, salt mixture (185) 3.7, butterfat 4, cod liver oil 1, varying amounts of materials furnishing vitamins B and C, and dextrin to one hundred parts. Food and water was kept before the animals at all times. The rats used in lactation studies were kept on shavings in experiments up to number 193. The animals on the remaining experiments were kept on shavings for seven days following parturition and then on two mesh per inch screens until weaned. Females were allowed uniform litters of six young each. The young were weaned at 28 days of age.

Wheat as a source of vitamins B and G for lactation

The experiments summarized in Table VI were conducted by transferring mother rats from the stock ration to the experimental ration on the day of parturition. The per cent mortality was 55.5, and the weaning weight of the young was 34.2 g. in 1st
53A. Lots 54A and 55A received 5 and 10 per cent, respectively, of dried yeast, in addition to 20 per cent of wheat. In lots 54A and 55A the weaning weights were 55.5g. and 56.4g., and the mortalities were 16.6 and 13.9 per cent, respectively. Lot 94 received 30 per cent of wheat plus 10 per cent of autoclaved yeast. The weaning weight of the young rats in this group averaged 50.0g., and the mortality was 16.6 per cent. Ration 69 was the same as ration 53A, with the exception that the one per cent of cod liver oil in ration 53A was replaced by butterfat in ration 69. The weight of the young at weaning in lot 69 dropped to 26.0g., and the mortality increased to 77.7 per cent. These data indicate that cod liver oil supplements butterfat in supporting a more normal milk secretion on a 20 per cent wheat ration. An inspection of the data in Table VI clearly shows that 5 and 10 per cent yeast definitely supplements 30 per cent of wheat in supplying the nutritive factors required for successful rearing of young rats. Ten per cent of autoclaved yeast improves a 30 per cent ration almost as well as 10 per cent of yeast; therefore, it is likely that the improvement in lactation is due to the heat-stable vitamin or vitamins contained in the yeast.

The reproduction and lactation data presented in Table VII were obtained by placing young rats with an initial weight of 45g. to 55g. on the experimental rations. A inspection of the reproduction records shows that reproduction was normal; every
female produced from two to six litters with the exception of the females in lot 118. The rats in lot 118 were lost after they were on the experimental diet only six months. Lot 147 also fell below the other lots in the number of litters produced. This was partly due to the death of some of these females early in the experiments. The growth rate on all of the rations in Table VII was considerably above normal, indicating that 30 per cent of wheat supplied ample vitamins B and G for normal growth. These animals were allowed shavings as litter so coprophagy was probably an important factor in their nutrition.

An inspection of the per cent mortality and weaning weights of the young in lots 116 and 144 conclusively demonstrates that 30 per cent of wheat is decidedly deficient in some factor or factors necessary for normal lactation. The young on ration 145 containing 5 per cent of yeast plus 30 per cent of wheat had a weaning weight of 56.0g. and a mortality of 18.8 per cent. The lactation performance of females receiving ration 146, which contained 10 per cent of yeast plus 30 per cent of wheat, was only slightly improved over the performance of females on ration 145. The young on rations 117 and 147, which contained 5 per cent of autoclaved yeast plus 30 per cent of wheat had weaning weights of 53.6g. and 46.8g. and mortalities of 59.2 per cent and 59.2 per cent, respectively. The young on rations 118 and 148, which contained 10 per cent autoclaved yeast plus 30 per cent wheat, had weaning weights of 40.5g. and 55.0g. and mortalities of 54.7
per cent and 14.4 per cent, respectively. A comparison of the
data of the different lots in Table VII shows that 5 per cent of
yeast is more effective than 5 per cent of autoclaved yeast in
 supplementing a 30 per cent wheat diet; but 10 per cent of yeast
is no more effective than 10 per cent of autoclaved yeast in
 supplementing a 30 per cent wheat diet. These results indicate
that 30 per cent of wheat supplies sufficient vitamin B, and
that the autoclaving process destroys some of the heat stable
vitamin G; which makes autoclaved yeast less effective in supple-
menting a 30 per cent wheat ration.

All data reported in Table VIII were obtained by trans-
ferring pregnant females from the stock ration to the experimen-
tal ration on the day of parturition. Sixty per cent of wheat
allowed the mothers in lot 53 to wean their young with an aver-
age weight of 50.6g. and a mortality of 34.8 per cent. Five
per cent of yeast was used to supplement 60 per cent of wheat in
ration 54. The young on ration 54 had a weaning weight of 55.1g.
and none of them died. The young from six females on ration 55,
which contained 10 per cent of yeast plus 60 per cent of wheat,
had a weaning weight of 65.0g. and a mortality of zero per cent.
A comparison of the data of lots 53, 54, and 55 indicates that
60 per cent of wheat furnishes sufficient vitamins B and G for
fair lactation; but a supplement of 5 per cent yeast increased
the weight of the young at weaning and decreased the mortality
of the young; and a supplement of 10 per cent of yeast still
further increased the weight of the young.

All data reported in Table IX were obtained from rats raised on the experimental ration. The growth rate on every ration in this group was above normal. Reproduction was normal on these rations. Young rats produced on ration 70, which contained 60 per cent of wheat, had an average weaning weight of 49.0g. and a mortality of 50 per cent. The young on ration 115, which had the same composition as ration 70, had a weaning weight of 54.5g. and a mortality of 30.1 per cent. The young on ration 70 had the higher mortality and also the greater weaning weight. The difference in the weaning weights was probably due to the smaller young animals in lot 70 being unable to survive the 28 days of the experiment. Females on ration 71 containing 5 per cent of yeast plus 60 per cent of wheat weaned young weighing 59.3g. and having a mortality of 5.8 per cent. Ten per cent of yeast plus 60 per cent of wheat in ration 72 enabled the females that received this ration to wean young with an average weight of 62.6g. and a mortality of 5.7 per cent. Females on ration 114, which contained 5 per cent of autoclaved yeast plus 60 per cent of wheat, weaned young with an average weight of 55.0g. and a mortality of 6.2 per cent. The weaning weight and per cent mortality of the young whose mothers received ration 115, containing 10 per cent of autoclaved yeast plus 60 per cent of wheat, were 58.0g. and 5.5 per cent respectively. The data summarized in Table IX demonstrate
that a 60 per cent wheat ration is deficient in some factor or factors necessary for the rearing of normal young rats. Since the factor or factors are carried by both yeast and autoclaved yeast; the factor or factors are probably vitamin G and not heat-labile vitamin B.

Rice polishings, wheat germ, rice bran, and corn germ as sources of vitamins B and G for lactation.

The data presented in Table X were obtained from females transferred from the stock ration to the experimental ration on the day of parturition. Females on ration 140 weaned young with an average weight of 39.5g. and a mortality of 30.6 per cent. The addition of 10 per cent autoclaved yeast to 10 per cent rice polishings in the ration of lot 140A allowed the females that received this ration to wean young weighing 56.3g. and having a mortality of 8.9 per cent. The average weaning weight and per cent mortality of the young, whose mothers received ration 141, which contained 10 per cent wheat germ, were 47.4g. and 62.5 per cent, respectively. Ten per cent of wheat germ plus 10 per cent of autoclaved yeast in ration 141A enabled mothers to wean young with an average weight of 56.8g. and a mortality of 8.3 per cent. Females on ration 142, which contained 10 per cent of rice bran, weaned young with an aver-
age weight of 46.7g. and a mortality of 44.4 per cent. Ration 142A, which contained 10 per cent of rice bran plus 10 per cent of autoclaved yeast allowed mother rats to wean young with an average weaning weight 54.1g. and a mortality of zero per cent. The average weaning weight and per cent mortality of young on ration 143, containing 10 per cent corn germ, were 22.5g. and 48.6 per cent, respectively. Ten per cent of corn germ plus 10 per cent of autoclaved yeast in ration 143A allowed females to rear young with an average weight of 27.2g. and a mortality of 38.8 per cent. The data of lots 140, 141, 142, and 143 demonstrate that 10 per cent rice polishings, wheat germ, rice bran or corn germ do not supply a sufficiency of some nutritive factor or factors to support normal lactation. The data of lots 140A, 141A, and 142A demonstrate that the addition of 10 per cent autoclaved yeast to 10 per cent of rice polishings, wheat germ, or rice bran increases the weaning weight of the young to about 55.0g. and decreases the mortality to less than 10 per cent. These data indicates that 10 per cent of rice polishings, wheat germ, or rice bran, as the sole sources of vitamins B and G in the ration do not supply enough vitamin G for normal lactation but do supply enough vitamin B for normal milk secretion. The addition of 10 per cent autoclaved yeast to 10 per cent of corn germ in ration 143A did not increase the weaning weight of the young to normal; so it seems that corn germ at a level of 10 per cent does not supply sufficient vitamins B and G to support normal rearing of young.
Oatmeal, yellow corn, white corn, and barley as sources of vitamins B and G for lactation

Table XI is self-explanatory; therefore it is unnecessary to discuss these data in detail. It is sufficient to call attention to the fact that the lactation records of females on a ration containing oatmeal, yellow corn, white corn, or barley are inferior to the lactation records of females receiving the same products plus five per cent autoclaved yeast. The addition of 10 per cent autoclaved yeast to the same amount of the various grains further improves the weaning weight of the young and decreases the mortality. The data in Table XI support the theory that cereal grains are a more potent source of vitamin B than vitamin G. Each of the four grains studied supplied enough vitamin B to support lactation at a rate superior to the stock ration.

Storage of vitamin G as a factor in lactation

The inability to rear normal young from rats transferred at parturition from the stock ration to a ration containing 60 per cent of yellow corn as the sole source of vitamins B and G has been demonstrated in lot 154. In order to determine if female rats on the stock ration store vitamin G, the following experiments were conducted. Six females in lot 154A received
60 per cent yellow corn throughout the lactation period. The mortality of their young was 66.6 per cent, and the average weight at weaning was 36.7g. In lot 154B six females were fed the stock ration until the young were seven days old. At this time the females were transferred to the 60 per cent yellow corn ration. Under these conditions the mortality was reduced to 22.2 per cent, and the average weaning weight was increased to 48.0g. Six females in lot 154C were transferred from the stock ration to a 60 per cent yellow corn ration, when the young were fourteen days old. In this group of young rats the average weaning weight was 46.5g. and the mortality was 11.1 per cent. The experiments summarized in Table XII demonstrate that the weaning weight and mortality of the young rats are influenced by the time at which the mothers are transferred from the stock ration to a 60 per cent corn ration.

The effect on lactation of supplementing yellow corn, and rice polishings with various sources of vitamin G

All lactation experiments after number 193 were conducted by allowing the females to remain on shavings for seven days after parturition. At the end of seven days the females were placed upon two mesh per inch screens, and the young were placed in a small metal pan 6" X 3" with sides one inch high. The pans were removed and the young placed on the screens when they were
fourteen days of age.

Fifteen per cent of fraction 11A, dried hog liver, was the sole source of the vitamin B complex in ration 193, Table XIII. The mortality of the young was 76.4 per cent and the average weight at weaning was 26.8g. In lot 203 the source of the vitamin B complex was 3.3 per cent of fraction 11A. The mothers on this ration were able to rear only one young out of six litters. Ten per cent of autoclaved yeast in the ration does not permit a female to rear her young successfully, as demonstrated in experiment 202. Liberal amounts of vitamin G in the form of dried hog liver or autoclaved yeast as the sole source of the vitamin B complex do not enable females to rear normal young, according to the data of lots 193, 202, and 203.

Fifty per cent of yellow corn, a rich source of vitamin B but relatively poor in vitamin G, is shown to be inadequate for the normal rearing of young, according to the data from lot 218. A ration containing 50 per cent of yellow corn plus 3.3 per cent of fraction 11A permitted females to wean their young with an average weight of 56.7g. and a mortality of 6.9 per cent, as shown in lot 205. Females that receive ration 204, containing 50 per cent of yellow corn plus 10 per cent of autoclaved yeast, reared their young with an average weight of 53.0g. and a mortality of 5.6 per cent.

The young, on diets rich in vitamin B but poor in vitamin G, that die show stomachs filled with curd. The young, whose mothers
receive a diet rich in vitamin G but deficient in vitamin B, exhibit peculiar spasms. These young run and scream and very frequently go into fits; later they develop paralysis in the hind legs. Examination of the stomachs of these young showed this organ to be empty. These differences in symptoms suggested that females, which receive a diet rich in vitamin B but deficient in vitamin G, might secrete an abundance of milk of low vitamin G content.

Experiments numbers 247, 248, 249, and 250 were designed to answer the question of whether there is a deficiency of vitamin G in the milk of mothers receiving ample vitamin B but insufficient vitamin G in the ration. Young in lot 249, from females on 50 per cent yellow corn, had an average weaning weight of 37.8g. and a mortality of 50 per cent. The females in lot 250 received 50 per cent of yellow corn; in addition each young was fed daily 0.1 ml. of fraction III, a hog liver concentrate, directly by means of a pipette from the 14th. to the 28th. day. These young had an average weaning weight of 45.1g. and a mortality of 20.8 per cent. Ration 247 contained 10 per cent of rice polishings as the sole source of the vitamin B complex. Mothers in this group weaned young with an average weight of 37.0g. and a mortality of 31.9 per cent. Mothers in lot 248 received 10 per cent of rice polishings; but each young was fed 0.1 ml. of fraction III daily, from the 14th. to the 28th. day. Young in lot 248 had an average weaning weight of 45.5g. and a
mortality of 11.1 per cent. The data from lots 247, 248, 249, and 250 indicate that mother rats receiving a diet rich in vitamin B but low in vitamin G secrete milk with a low vitamin G content; and direct feeding of a source of vitamin G is beneficial to the young. Young were reared as successfully in lots 248 and 250 as in lot 243, which received the stock ration.

The effect of fraction 8B, fraction 11A, fraction 11F, and fraction 11F₁ as sources of vitamins B and G on lactation

Data presented in lots 232 and 265 Table III, demonstrate that 0.053g. of fraction 8B per rat per day together with autoclaved yeast supplied sufficient vitamin B to produce an average gain in young rats of at least 10.0g. per week. Experiments 231 Table III and 219 Table IV show that the same amount of fraction 8B when fed with out the addition of autoclaved yeast produces an average gain of about 2.5g. per week. These data prove fraction 8B to be at least four times as potent in vitamin B as vitamin G. Data presented in lot 196 show that 0.1g. of fraction 11A per rat per day as a source of vitamin G produces a gain of 9.0g. per week. Lot 200 Table IV demonstrates that 0.3g. of fraction 11A supplies enough vitamin B for a gain of 4.6g. per week. The results from lots 257, 258, 264, 288, 289, and 290, Table IV show fraction 11F and fraction 11F₁ to be relatively potent sources of
vitamin G, with a small or negligible vitamin B content.

A summary of the lactation data obtained by feeding fraction 8B plus fraction 11A to pregnant females, transferred from the stock ration to the experimental ration on the day of parturition, is presented in Table XIV. Females on ration 189 were unable to rear a single young. In experiments 274 and 294 each female received 0.318g. of fraction 8B per day as the sole source of vitamins B and G. This ration is unsatisfactory for normal rearing of young, as is disclosed by a weaning weight of 20.4g. and a mortality of 30.5 per cent in lot 274 and a weaning weight of 19.3g. and a mortality of 38.8 per cent in lot 294. Fraction 11A (dried hog liver) at a level as high as 15 per cent has been proven, lot 198, Table XIII, to be inadequate for normal lactation. Experiment 296 shows that 1.66g. of fraction 11F per day does not furnish sufficient of the vitamin B complex to allow a female to rear normal young. These young developed paralysis of the hind legs.

In experiment 192 each female received 0.063g. of fraction 8B plus 0.2g. of fraction 11A. The average weaning weight was 28g. and the mortality was 95.7 per cent. Mothers in lot 244 received 0.106g. of fraction 8B plus 0.5g. of fraction 11A. Their young had an average weaning weight of 30.7g. and a mortality of 43.5 per cent. The sources of the vitamin B complex in ration 245 was 0.218g. of fraction 8B plus 0.500g. of fraction 11A per day. These young had an average weaning weight of 28.9g.
with a mortality of 44.4 per cent. In ration 246 the source of the vitamin B complex was 0.106g. of fraction 8B plus 1.0g. of fraction 11A per female per day. Young rats on this ration had an average weaning weight of 29.0g. and a mortality of 93.3 per cent. Females that received 0.218g. of fraction 8B plus 1.0g. of fraction 11A in lot 269 were able to rear young with an average weight of 31.9g. at 28 days and with a mortality of 44.4 per cent. The average weaning weight and per cent mortality in lot 270 was 45.2g. and 19.4, respectively. The source of the vitamin B complex was 0.318g. of fraction 8B plus 1.0g. of fraction 11A.

The source of the vitamin B complex in ration 272 was 0.318g. of fraction 8B plus 10 per cent autoclaved yeast. Young in this lot had an average weight of 35.2g. at 28 days and a mortality of 16.6 per cent. In the experiments 273, 275, 295, and 297 each female received 0.318g. of fraction 8B per day. Each female in lot 273 received 1.32g. of fraction 11F per day. The females in lots 275 and 295 received 1.33g. of fraction 11F per day. The allowance of fraction 11F was 1.66g. per female per day in lot 297. The average weaning weight were 31.2g., 34.1g., 35.3, and 39.3g., respectively, in lots 273, 275, 295, and 297. The per cent mortality was 6.94, 29.2, 16.6, and 8.3 in the respective lots. The above data demonstrate that, as the amount of fraction 11F or fraction 11F was increased, the average weight of young at weaning increased, but the per cent mortality did
not decrease in all cases.

The data presented in Table XIV demonstrate that a source of vitamin B, such as fraction 8B, does not promote successful rearing of young rats when fed as the sole source of the vitamin B complex. It is also shown by the data in Table XIV that sources of vitamin G, as fraction 11A, fraction 11F₄, or fraction 11F fed as the sole sources of the vitamin B complex, do not allow mother rats to rear young at a normal rate. These data further demonstrate that at least six times as much vitamin B plus five or six times as much vitamin G as are necessary for normal growth are required for successful rearing of young.

Studies on yellow corn and wheat supplemented with fraction 8B and fraction 11F₄ as sources of vitamins B and G for lactation.

Fraction 8B fed at a level of 0.318g. per female per day was not able to supplement a 60 per cent yellow corn ration, as is demonstrated by data from lots 309 and 311 Table XIV. Data from lot 310 show that 1.66g. of fraction 11F₄, per female per day, is effective in supplementing a diet containing 60 per cent of yellow corn. A 60 per cent wheat ration was only slightly improved by feeding 0.318g. of fraction 8B, per female per day, according to the data in lots 312 and 314. The addition of a supplement of 1.66g. of fraction 11F₄ per female per day, to a
60 per cent wheat ration, in lot 313, allowed six female rats to wean their young with an average weight of 53.8g. and zero per cent mortality. The data in Table XV conclusively demonstrate the failure of female rats to rear their young on a wheat or yellow corn ration to be due to a deficiency of vitamin G and not a deficiency of vitamin B.

**Hemoglobin Studies**

The average hemoglobin content of the blood of normal rats on a mixed grain and milk diet was reported by Mitchell and Miller (73) to be 16.2g. per 100c.c. of blood. The same workers found the hemoglobin values dropped to 13.0g. per 100c.c. at parturition; the anemia gradually disappeared during the lactation period. This anemia was considered by Mitchell and Miller to be a physiological rather than a pathological condition. A deficiency in the vitamin B complex was reported by Sure, Kik, and Walker (119) to produce a disturbance in the hematopoietic function of nursing young. Sure, Kik, and Smith (116) observed a marked anemia in rats that developed skin lesions on a vitamin G deficient diet.

The experiments presented in Table XVI were designed to determine if the hemoglobin of lactating females was affected by the vitamin G content of the ration. In the experiment lots 179 to 183 six young vigorous females and two males, four months
old, were transferred from the stock ration to the experimental ration. Hemoglobin determinations were made weekly by the Newcomer (51) method. The males had an average hemoglobin value of 16.3g. per 100c.c. of blood at the beginning and 15.5g. per 100c.c. after seven weeks on the various rations. The average for the females at the beginning was 15.6g. of hemoglobin per 100c.c. of blood. The hemoglobin remained practically constant until the females commenced producing litters; and then it dropped to 15.9, 14.2, 12.8, and 12.4g. hemoglobin per 100c.c. of blood in lots 179, 180, 181, 182, and 183, respectively. The hemoglobin values remained low in all the lots during the lactation period; but there appears to be no appreciable differences in the hemoglobin content of the rats on the different rations.

In the series of experiments 309, 310, 311, 312, 313, and 314 females were transferred from the stock ration to the experimental ration on the day their young were born. Hemoglobin determinations were made on the day of parturition and each week during the lactation period. The average hemoglobin values at parturition were 13.4g., 13.9g., 15.5g., 15.8g., 14.1g., and 14.0g. per 100c.c. of blood in lots 309, 310, 311, 312, 313, and 314, respectively. After parturition the hemoglobin content of the blood increased until at the end of the lactation period the values were 15.5g., 16.6g., 16.6g., 16.5g., 17.0g., and 16.6g. per 100c.c. of blood in lots 309, 310, 311, 312, 313, and 314, res-
pectively.

The data presented confirm the results of Mitchell and Miller (73), who showed that the hemoglobin value dropped to 13g. of hemoglobin per 100c.c. of blood at parturition and gradually increased during the lactation period. The hemoglobin content of the rat during pregnancy reported in this thesis was not as low as that stated by Beard and Myers (5). These investigators reported that hemoglobin during pregnancy fell to a level of 10 to 11g. hemoglobin per 100c.c. of blood.
Discussion of Results

Wheat germ was found (Table I) to be somewhat more potent in vitamin B than in vitamin G. This is in general agreement with data presented in the literature; however, the value of 6.4 units of vitamin G per gram of wheat germ is twice as great as that reported by Munsell and De Vaney (79). Flimmer, Raymond, and Lowndes (85) found 5.0 per cent to 7.0 per cent of wheat germ to required to supply sufficient vitamin B for maintenance in pigeons. Five per cent wheat germ as a source of vitamin B produced normal growth in rats. The data presented in Table II confirm the results of Aykroyd and Roscoe (3), Chick and Roscoe (16), and Sherman and Sandles (93) which showed yeast to be a potent source of both vitamins B and G.

Rice polishings are approximately twice as potent in vitamin B as vitamin G, according to the data in Table III. Munsell (79) found rice polishings to contain 5.0 units of vitamin B and 1.0 unit of vitamin G per gram. The sample of rice polishings used in this work had a vitamin B content of one and two-thirds times as great and a vitamin G content six times as great as the average values reported by Munsell. The difference in the vitamin B and vitamin G content was greater in rice polishings than in wheat germ or yeast. Vitamin B is present in a higher concentration in rice polishings than in wheat germ or
yeast. This inequality of vitamin B and vitamin G content in rice polishings should cause rice polishings to be a more desirable starting material than wheat germ or yeast, for the preparation of potent extracts of vitamin B free from vitamin G.

By making use of the differences in distribution, solubility in strong alcohol, and adsorption on fuller's earth or norite, several investigators have been able to obtain sources of vitamin B, with a vitamin G content low enough to cause loss of weight in rats. An extract obtained by extracting wheat germ with 35 per cent alcohol was found by Chick and Roscoe (16) to supply vitamin B but to produce no growth. Bourquin and Sherman (9) use an 80 per cent alcoholic extract of whole wheat as a source of vitamin B in assaying materials for vitamin G. Tikiti, a strong alcoholic extract of rice polishings was found by Evans and Burr (22) to be able to promote only a slight amount of growth as the sole source of the vitamin B complex. Evans and Lebkovsky (25), by making use of the differences in solubility in alcohol and adsorption on fuller's earth, were able to prepare a good source of vitamin B free from vitamin G. The adsorption on fuller's earth of vitamins B and G was found by Salmon and Guerrant (90) to be influenced by the pH of the solution; maximum adsorption of vitamin B was at pH 4.5 and maximum adsorption of vitamin G was at pH 0.08. Extraction with 95 per cent alcohol, fractionation with lead acetate, and adsorption on fuller's earth was reported by Guha (34) to be effective in
preparing a source of vitamin B free from vitamin G. With evidence as the above it is difficult to explain the gain in weight made by animals in lots 103, and 96, (Table I); lots 104, and lots 173, (Table II); lots 122, 163, 165, 184, 194, 203, 211, 231, and 251, (Table III). The average gain in weight made by the rats in the different lots was fairly constant. It is interesting to note that animals in lots 194, 208, and 211 received 0.027g. fraction 3B per rat per day and made an average gain of 4.1g., 4.0g., and 3.1g. per week, respectively. Animals in lots 184 and 231 received 0.065g. fraction 3B per rat per day and made a gain of 4.2g. and 2.0g., respectively. These data suggest that the amount of the vitamin G which the animals received was not proportional to the amount of fraction 3B fed per animal per day. These data suggest that the basal ration might furnish a small amount of vitamin G.

Experiments 206 to 211 (Table III) indicate that alcohol extracted casein contains less vitamin G than casein leached with water acidified with acetic acid. This is in agreement with the results of Chick and Roscoe (17), Palmer and Kennedy (23), Sherman and Spohn (100), and Evans and Lepkovsky (26), who have shown the purification of casein to be of the utmost importance in vitamin studies. Fraction 3B plus the basal ration containing alcohol extracted casein supplies a small amount of vitamin G as shown by the results in lots 208, and 251, (Table III) and lot 219 (Table IV). It is possible that
the source of the vitamin G is the dextrin used. Evans and Lepkovsky (26), and Hegen and Richardson (41) found that corn starch contains vitamin G, and it is necessary to use recrystallized sugar as a source of carbohydrate in vitamin B studies. Sherman and co-workers (100), (98), (9), and (12) find corn starch to be suitable as a source of carbohydrate in vitamin B and G studies. The English workers Chick and Roscoe (17) use rice starch in vitamin B and G studies. The growth obtained in lots 219 and 231, is possibly due to vitamin G contained in the basal ration. The difficulties encountered in securing a ration that supplies vitamin B without vitamin G suggests that a study of the vitamin content of the carbohydrate used in the basal ration might show this constituent to contain vitamin G.

Data presented in Table II show extraction with 95 per cent alcohol to be more effective in removing the vitamin B than the vitamin G contained in yeast. The yeast residue, fraction 22, was almost as potent in vitamin G as the original yeast. Sixty per cent alcohol is a more effective solvent than 95 per cent alcohol for the extraction of vitamin G from yeast. These results confirm the data of Sherman and Sandle (98), which showed 95 per cent alcohol extracted yeast to have as high a vitamin G content as the original yeast. Sherman and Sandle showed extraction of yeast with 60 per cent alcohol reduced the vitamin G content to about one-third of the original vitamin G content of the yeast. Data in Table II are in harmony with the results
of Smith (106), in so far as extractibility of vitamin G with 95 per cent alcohol is concerned; but the data in Table II indicate that extraction of yeast with eight portions of 95 per cent alcohol does not remove vitamin B quantitatively from yeast. This does not substantiate the work of Smith (106), who found vitamin B was almost completely removed from yeast by extraction with 95 per cent alcohol.

Fraction II A, dried hog liver, is a potent source of vitamin G, according to the results in lots 196 and 197 (Table IV). Fraction II A at a level of 0.1 g. per rat per day, lot 196, furnished sufficient vitamin G to produce a gain of 9.0 g. per week. A comparison of these data with data obtained by Aykroyd and Rescoe (3) shows the vitamin G content of dried hog liver and dried beef liver to be very similar. A gain of 102 g. in 30 days made by rats receiving 290 mg. of dried beef liver per day was reported by Block and Farquhar (7). This is a much greater rate of gain than that made by rats, which received 0.30 g. of dried hog liver as a source of vitamin G in lot 197. Extraction of hog liver by the method reported by Guha (35) does not remove vitamin G quantitatively. The hog liver extract thus obtained fed at a rate equivalent to 0.50 g. fresh hog liver per rat per day as a source of vitamin G produced a gain of 7.5 g. per week. The vitamin G content of the hog liver extract is similar to the vitamin G content of the beef liver extract prepared by Guha (35). The aqueous hog liver extract is shown by a gain of 2.5 g. per week.
in lot 199 to contain a small amount of vitamin B.

The series of experiments 219 to 290 (Table IV) demonstrates that as the concentration of alcohol was increased the amount of vitamin G precipitated from a hog liver solution increased. With the exception of the data from lot 257 there is little or no indication of an increase in the amount of vitamin B precipitated with an increase in the concentration of alcohol. Animals receiving fraction 11F₁ as the sole source of the vitamin B complex developed polyneuritis. Fraction 11F₁, supplemented with a source of antineuritic vitamin B, supports normal growth in rats. These results suggest that fraction 11F₁ might be used as a source of vitamin G in studying the vitamin B content of natural foods or vitamin B concentrates. Narayanan and Drummond (80) observed that the addition of alcohol to give a concentration of 70 per cent alcohol precipitated vitamin G from a yeast extract. Sherman and Sandals (95), Chick and Roscoe (15), and Smith (105) have demonstrated that strong alcohol is not an effective solvent for the extraction of vitamin G from yeast.

A comparison of the data in this thesis with the findings of the above workers indicates that the solubility in strong alcohol of vitamin G contained in an aqueous hog liver extract is very similar to the solubility of vitamin G contained in an aqueous yeast extract or yeast.

Bocher (8) found boiling 93 per cent to 94 per cent ethyl alcohol extracted vitamin G from whey powder. Eighty per cent
of ethyl alcohol was found by Stiebeling and Alleman (109) to extract vitamins B and G at almost equal rates from skimmed milk powder. They attributed the low vitamin G content of the extract prepared with 0.1M HCl in 50 per cent by weight alcohol to the oxidation of vitamin G in the presence of strong alcohol. They found 0.1M gallic acid decreased the losses of vitamin G in strong alcohol solutions, due probably to the reduction potential of gallic acid. Chisk and Reese (15) observed that about one-half of the vitamin G contained in yeast was destroyed by contact with strong alcohol. The failure to recover quantitatively (Tables IV and V) vitamin G contained in whey powder or liver extract after treatment with strong alcohol is probably due to destruction of a portion of the vitamin G through contact with the strong alcohol.

Levene (59), (60) reported that acetone precipitated vitamin G from yeast extract. In studying the extractability of vitamin G from yeast with various acetone-water mixtures Day (18) found 60 per cent acetone removes about one-half of the vitamin G contained in yeast. If the concentration of acetone was 80 per cent or greater the residue contained the total vitamin G content of the original yeast. The data from lots 225 and 226 (Table IV) demonstrate that 60 per cent acetone precipitates about one-third of the vitamin G contained in a hog liver extract. Over one-half of the vitamin G contained in a hog liver extract was precipitated by 80 per cent acetone, according to the data in lots 227 and 228.
These data show the solubility in acetone-water mixtures of vitamin G in a hog liver extract to be similar to the solubility in acetone-water mixtures of vitamin G in yeast extracts and yeast, as reported by Levens (59), (60) and Day (13).

Extraction of hog liver by the method reported by Mapson (69) did not remove vitamin G more completely than extraction of hog liver by the method reported by Guha (35). A larger percentage of the total hog liver solids is removed by the Mapson extraction. The above facts suggest that the extract obtained by the method reported by Guha (35) should be the better starting material to use in attempting to obtain pure preparations of vitamin G.

The experiments reported Tables VI, VII, VIII, and IX were performed to determine if the length of time a female is on a ration before parturition affected the rearing of young. A comparison of the data in Table VI with that in Table VII and data in Table VIII with data in Table IX indicates that females reared on purified rations do not rear young as successfully as females transferred to the purified ration on the day of parturition. This is in agreement with the finds of Taylor (126). The data in Tables VI and VIII demonstrate that females transferred from the stock ration to a 30 per cent wheat ration or a 60 per cent wheat ration are unable to rear young normally. These conditions shows that females transferred from the stock diet to a purified diet on the day of parturition fail to rear young at a normal rate if the purified diet is deficient in some dietary
factor essential for normal lactation. This fact shows that pregnant females transferred from the stock ration to the experimental ration on the day of parturition are suitable test animals to be used in studying the requirements of water-soluble vitamins B and G during lactation.

Data presented in Table X demonstrate that rice polishings, wheat germ, rice bran, or corn germ incorporated in a purified ration at a level of ten per cent fail to supply sufficient vitamins B and G for lactation. The lactation performance of females in lot 141 is very similar to the data found by Taylor (126) using 12 per cent wheat germ as the source of the vitamin B complex. Sure and Schilling (120) observed that ten per cent defatted wheat embryo as a source of the vitamin B complex was entirely unsatisfactory for lactation. They reported that the young died of beriberi, with their stomachs filled with milk. Young in lot 141 often had screaming, running fits, and died with curd in their stomachs. The data from lot 141A prove that sufficient vitamin B is supplied by 10 per cent of wheat germ, if supplemented with 10 per cent autoclaved yeast as a source of vitamin G. In view of these data it seems likely that conditions noted by Sure and Schilling (120) were not true beriberi and the improvement in lactation produced by increasing the defatted wheat germ to 30 per cent was due to the increase of vitamin G rather than an increase in vitamin B. Evans and Burr (84) and Sure (110) found 10 per cent of rice polishings fail-
ed to furnish sufficient vitamin B for lactation. These workers believed vitamin B to be the limiting factor. The data in Table X indicate that the limiting factor in a 10 per cent rice polishings ration is vitamin G rather than vitamin B.

Guest, Nelson, Parks, and Fulmer (33) and Taylor (126) observed various cereal grains fed at different levels were unable to support normal lactation in rats. Taylor (126) found 10 per cent of autoclaved yeast supplemented a purified diet containing 63.3 per cent of white corn, yellow corn, barley, or hulled oats as a source of the vitamin B complex for lactation. The data in Table XI confirm the data of the above workers; and demonstrate oatmeal, yellow corn, white corn, or barley at a level of 60 per cent in a purified ration fail to furnish vitamin G in sufficient quantities to meet the requirements of lactation. It is also shown that the seeds studied in Table XI are supplemented by five per cent autoclaved yeast. Ten per cent autoclaved yeast further supplements the various seeds as a source of the vitamin B complex for lactation. These facts suggest the limiting factor in the various seed rations to be vitamin G.

The nature of the deficiency of rations, in which yellow corn or rice polishings are the sole sources of vitamins B and G, is further elucidated by the data in Table XIII. It is evident that increased amounts of both vitamins B and G are required for normal rearing of young. This corroborates the find-
The yield of 60 per cent wheat at 10 per cent uncoated yeast as effective in improving late season containing 60 per cent yellow or other yeast concentrations enhance the late season performance. This is why do not increase the possibility of the yeast productions suggested the efficiency produced in uncoated yeast to be used.

Infection such as those reported by the above mentioned workers a direct link in wheat with uncoated yeast.

As far much the ocean water in human as well it is likely that the limiting growth factor is human. But each woman 100 of yeast-days. In view of these facts, a considerable increase in the wheat at a low and uncoated very similar to a low water content. They have found in the water content of human milk observed that women on the day of delivery

A young to thrive is due to the poor quantity of the milk. Don't depend on the secretion and care for the child. But the child on the least needed feeding. Also in the ease of the mother's milk.

III. A source of vitamin G, the monosaccharides that mother must re-

and other inventions. The marked improvement in the reading
The nitrogen content of fraction 11F₁ is 1.51 per cent. It is unlikely that the marked improvement in lactation is due to the protein supplied by as little as 1.66g. of fraction 11F₁ per female per day. Fraction 9B, a potent source of vitamin B, was entirely unsatisfactory as a supplement to rations containing yellow corn or wheat. These data further demonstrate the unsatisfactory results obtained with yellow corn or wheat as the sole source of the vitamin B complex to be a deficiency of vitamin G rather than vitamin B.

Hussemann and Ketler (46) observed that five per cent of yeast supplied the maintenance level of vitamins B and G; and an increase of either vitamin B in the form of tikiti or vitamin G as autoclaved yeast produced an improvement in lactation. Moore and co-workers (75), (76) used an 85 per cent alcoholic extract of corn as a source of vitamin B and yeast autoclaved three hours at fifteen pounds pressure as a source of vitamin G. They obtained poor lactation with all synthetic diets studied. They concluded that a factor contained in yeast, but not in the alcoholic extract of corn or autoclaved yeast is required for lactation; and that this factor is probably identical with the vitamin B₄ described by Reader (97). Sure and Kik (117) found vitamin B exerted a specific effect upon growth in addition to increasing the food consumption and the plane of nutrition. Sure and Walker (125) found the vitamin B in yeast exerted a specific effect upon lactation. Sure and Smith (121), Sure,
Kik and Walker (118) observed that a vitamin B concentrate prepared from rice polishings supplemented lactation on a ration in which 10 per cent autoclaved yeast supplied vitamin G. In Table XIV lactation performances of females receiving fraction 8B, fraction 11A, fraction 11F, fraction 11F₁, or autoclaved yeast and combinations of these sources of vitamins B and G are recorded. It is evident that sources of vitamin G, as autoclaved yeast, fraction 11F₁, and fraction 11A, which are potent sources of vitamin G relatively free from vitamin B, are unsatisfactory as the sole sources of vitamins B and G for lactation. These data confirm the work of Sure and Smith (123), Sure, Kik and Walker (118), and Russewann and Hetler (46).

The series of experiments 194 to 297 (inclusive) Table XIV demonstrates that vitamins B and G fed at the rate required to support normal growth fail to support lactation. It is further demonstrated that increased amounts of both vitamins B and G are required for rearing of young. An increase in vitamin G in the form of fraction 11A produced an improvement in lactation only if accompanied by an increased amount of vitamin B in the form of fraction 8B. Likewise it is demonstrated that an increase of vitamin B in the form of fraction 8B can not decrease the mortality of the young and increase the weaning weight of the young unless accompanied by an increased amount of vitamin G as autoclaved yeast, fraction 11A, fraction 11F, or fraction 11F₁. The minimum amounts of vitamins B and G required
for normal rearing of young are demonstrated to be at least six times the amounts required to produce normal growth in young rats.

The results summarized in Table XIV show that, although extracts and concentrates of vitamins B and G, when fed at six times the rate required to produce normal growth in young rats, support rearing of young rats at about the same rate as the stock ration, they do not allow rearing of young at as satisfactory a rate as purified rations in which the vitamin B complex is supplied by a combination of cereal grains, with autoclaved yeast, fraction II A, or fraction II F₁. The sources of vitamin G, fraction II A, II F₁, or autoclaved yeast, used to supplement fraction SB in rearing of young were demonstrated by data in Tables XIII and XIV to support lactation at a superior rate when supplemented with rice polishings, yellow corn, or wheat. These experiments suggest that a factor, or factors, other than anti-neuritic vitamin B and vitamin G are required for successful rearing of young.

The method of preparing fraction SB suggests the limiting factor to be similar to the third factor described by Hunt (45), and Hunt and Wilder (45). This factor was reported to be contained in yeast residue after extraction with 0.1 per cent acetic acid. They further found a factor which was adsorbed on fuller's earth but difficultly eluted with barium hydroxide. It is possible that the third factor described by Hunt (45) is
identical with vitamin $B_4$ reported by Reader (66), (87). The heat and alkali stability of the third factor of Hunt is similar to that of vitamin $B_4$ as described by Reader (66), (87). Moore, Plymate, Andrew, and White (76), Moore, Plymate, and Andrew (75) observed that an 85 per cent alcoholic extract of corn plus autoclaved yeast did not support normal lactation. They suggested the poor lactation was probably due to a deficiency of vitamin $B_4$ described by Reader. Hussenmann and Hetler (46) suggest that there is a definite quantitative relationship which exists between the amounts of vitamins $B$ and $G$ required for successful lactation. This is in accord with the view of Kellogg and Eddy (47) who suggest the ratio of vitamin $B$ to $G$ is more important than the absolute amounts of the two vitamins. Evans and Burr (24), Macy (66), and Sure (111) found three to five times the amount of yeast required to support growth in young rats were required to support lactation. In light of the data in Tables XIII, XIV and XV, and the data reported in the literature, it is likely that the limiting factor, or factors, in lots 270, 272, and 297 is similar to the third factors reported by Hunt (43) and Reader (66), (87).
Summary and Conclusions

The vitamin B and vitamin G content of wheat germ, rice polishings, and dried yeast were investigated. Five per cent of rice polishings or wheat germ in a purified ration did not produce normal growth in young rats. Five per cent of rice polishings supplemented with five per cent autoclaved yeast produced normal growth in young rats. Wheat germ at a level of five per cent furnishes sufficient vitamins B and G to support growth at almost a normal rate. Five per cent autoclaved yeast supplements five per cent of wheat germ so that growth is normal. This shows wheat germ to be more potent in vitamin B than vitamin G. The vitamin B and vitamin G content of dried yeast appear to be almost equal.

Autoclaved yeast is a good source of vitamin G free, or practically free, from vitamin B.

Of the several extracts and concentrates prepared fraction 8B and fraction 9B (the extract prepared by extracting rice polishings with 95 per cent alcohol, concentrating and adsorbing on fuller's earth) were the most potent sources of vitamin B.

All extracts and concentrates of vitamin B tested produced some growth as the sole source of the vitamin B complex. The rate of growth was slow and fairly constant. This constant rate of growth suggests that the basal ration or the preparation employed
might contain a small amount of vitamin G.

Dried hog liver is a potent source of vitamin G. The vitamin B content of dried hog liver is much lower than the vitamin G content.

The extraction of hog liver by the methods employed in this thesis does not remove vitamin G quantitatively from hog liver. The hog liver extracts prepared were rich in vitamin G and relatively poor in vitamin B.

The addition of alcohol to an aqueous liver extract causes the precipitation of vitamin G. As the concentration of alcohol was increased the amount of vitamin G precipitated was increased. Fraction 11F, prepared by adding alcohol to an aqueous hog liver concentrate and drying the precipitate on dextrin, is a good source of vitamin G. Polyneuritis is readily produced in rats by feeding the basal ration plus 0.33g. of fraction 11F (vitamin G preparation) per rat per day as a source of vitamin G.

Acetone at a concentration of 80 per cent precipitates vitamin G from an aqueous hog liver concentrate. Acetone at a concentration of 60 per cent precipitates only a small amount of vitamin G from an aqueous hog liver concentration.

Whey powder is a good source of vitamin G.

Extraction of whey powder with boiling 95 per cent ethyl alcohol removes considerable amounts of vitamin G.

The solubility of vitamin G of whey powder in boiling 95 per
cent ethyl alcohol is greater than the solubility of vitamin G contained in dried yeast or a hog liver concentrate in 80 per cent alcohol or alcohol of a greater concentration at room temperature.

The vitamin G contained in whey powder or a hog liver concentrate is not fully accounted for in the various fractions produced by treatment with ethyl alcohol. This suggests that strong ethyl alcohol destroys vitamin G to a certain extent.

Thirty per cent wheat or 60 per cent wheat as the sole source of vitamins B and G does not produce normal lactation. Five per cent yeast, 10 per cent yeast, and 5 per cent autoclaved yeast, or 10 per cent autoclaved yeast supplement a ration containing 30 or 60 per cent wheat as a source of vitamins B and G for lactation.

Oatmeal, yellow corn, white corn, or barley incorporated in the basal diet at a level of 60 per cent do not, as the sole sources of vitamins B and G, support normal rearing of young. Addition of five per cent autoclaved yeast to the various rations in which the above grains were included improved the lactation of females. Ten per cent of autoclaved yeast in supplementing the various grain rations was more effective than five per cent of autoclaved yeast.

Rice polishings, wheat germ, rice bran, or corn germ at a 10 per cent level as the sole source of vitamins B and G do not support normal lactation. The addition of 10 per cent of autoc-
claved yeast markedly supplements 10 per cent of rice polishings, wheat germ, or rice bran for lactation. Ten per cent of autoclaved yeast plus 10 per cent corn germ do not produce normal lactation.

Ten per cent of autoclaved yeast or 15 per cent of dried hog liver, (fraction 11A) as the sole sources of vitamins B and G, do not support lactation.

Fraction 11A (dried hog liver) at a level of 5.3 per cent is as effective as autoclaved yeast at a level of 10 per cent in supplementing a ration containing 50 per cent of yellow corn.

Mother rats on a diet containing 50 per cent yellow corn or 10 per cent of rice polishings secrete milk of a low vitamin G content. Direct feeding of fraction 11L (hog liver extract) to the young increases the weight of the young at weaning and decreases the mortality of the young.

Lactation is not improved by supplementing a 60 per cent wheat or a 60 per cent yellow corn ration with 0.318g. of fraction 8B (source of vitamin B) per female per day. Lactation is improved by supplementing a 60 per cent wheat or a 60 per cent yellow corn ration with 1.66g. of fraction 11F (source of vitamin G) per female per day. A ration containing 60 per cent of wheat or 60 per cent of yellow corn is deficient in vitamin G for lactation.

Fraction 8B fed at the rate of 0.318g. per female per day, does not allow a female to rear normal young.
Fraction 11F₁ (source of vitamin G) fed at the rate of 1.66g per female per day, does not allow a female to rear normal young.

As the sole sources of vitamins B and G, fraction 8B (source of vitamin B) and fraction 11A (source of vitamin G) must be fed simultaneously and at a rate six times the rate required to give normal growth in young rats, in order to produce lactation approaching the normal. Fraction 8B (source of vitamin B) and fraction 11F₁ (source of vitamin G) must be fed together and at about six times the rate required to give normal growth in young rats, in order to produce lactation at a nearly normal rate.

Combinations of fraction 8B with fraction 11A, fraction 8B with fraction 11F, fraction 8B with fraction 11F₁, or fraction 8B with autoclaved yeast at the rates fed did not give as satisfactory lactation as a combination of wheat or corn with fraction 11F₁, or corn with fraction 11A, or autoclaved yeast with oatmeal, yellow corn, white corn, barley, rice polishings, rice bran, or wheat germ. It is very probable that the seeds and products from seeds studied contain a factor, or factors, other than antineuritic vitamin B necessary for lactation and not contained in a purified diet in which autoclaved yeast, fraction 11A, fraction 11F, or fraction 11F₁ are the sources of vitamin G.

The hemoglobin content of rats blood decreases in the last stages of pregnancy and during part of the lactation period. The hemoglobin content of the blood gradually increases during the
later part of the lactation period. There is no apparent relationship between the severity of the anemia of pregnancy and the vitamin G content of the diet.
ACKNOWLEDGMENT

I wish to express my appreciation of the assistance and counsel of Professor V. E. Nelson in this work.
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Table I

Wheat germ and Preparations from Wheat germ as sources of vitamin B and G

<table>
<thead>
<tr>
<th>Lot No.</th>
<th>Supplement</th>
<th>Avg. Gain</th>
<th>Units per gram per rat per week</th>
<th>B of rats</th>
<th>G of rats</th>
<th>Died of rats</th>
</tr>
</thead>
<tbody>
<tr>
<td>102</td>
<td>No supplement</td>
<td>10%</td>
<td>2.7g</td>
<td>4.3</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>99</td>
<td>Yeast Auto.</td>
<td>10%</td>
<td>2.7g</td>
<td>4.3</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>96</td>
<td>Fraction IC</td>
<td>0.2g</td>
<td>7.3g</td>
<td>12.5</td>
<td>6</td>
<td>1</td>
</tr>
<tr>
<td>97</td>
<td>Fraction IC</td>
<td>0.2g</td>
<td>7.3g</td>
<td>12.5</td>
<td>6</td>
<td>1</td>
</tr>
<tr>
<td>101</td>
<td>Fraction IA</td>
<td>1.0g</td>
<td>7.6g</td>
<td>2.5</td>
<td>6</td>
<td>1</td>
</tr>
<tr>
<td>102</td>
<td>Fraction IB</td>
<td>1.0g</td>
<td>2.4g</td>
<td>0.61</td>
<td>6</td>
<td>0</td>
</tr>
<tr>
<td>119</td>
<td>Wheat germ</td>
<td>5%</td>
<td>9.2g</td>
<td>6.4</td>
<td>6</td>
<td>0</td>
</tr>
<tr>
<td>120</td>
<td>**Wheat germ</td>
<td>5%</td>
<td>11.7g</td>
<td>7.3</td>
<td>6</td>
<td>0</td>
</tr>
<tr>
<td>130</td>
<td>Fraction 2A</td>
<td>10%</td>
<td>2.2g</td>
<td>1.1</td>
<td>6</td>
<td>0</td>
</tr>
<tr>
<td>131</td>
<td>**Fraction 2A</td>
<td>10%</td>
<td>2.2g</td>
<td>1.1</td>
<td>6</td>
<td>0</td>
</tr>
<tr>
<td>132</td>
<td>Fraction 2B</td>
<td>10%</td>
<td>0.0g</td>
<td>0.0</td>
<td>6</td>
<td>0</td>
</tr>
<tr>
<td>133</td>
<td>Fraction 2B</td>
<td>5%</td>
<td>5.9g</td>
<td>5.0</td>
<td>6</td>
<td>0</td>
</tr>
<tr>
<td>134</td>
<td>**Fraction 2B</td>
<td>5%</td>
<td>1.5g</td>
<td>1.3</td>
<td>6</td>
<td>0</td>
</tr>
</tbody>
</table>

* Denotes 10% autoclaved yeast added as source of vitamin G.
** Denotes 5% autoclaved yeast added as source of vitamin G.
# Denotes 0.1g fraction 1D per rat per day added as source of vitamin B.
Table II

Yeast and preparation from yeast as sources of vitamin B and G.

<table>
<thead>
<tr>
<th>Lot:</th>
<th>Material tested</th>
<th>Avg. gain: Units per:</th>
<th>per rat:</th>
<th>Gram:</th>
<th>No.:</th>
<th>No. of rats:</th>
<th>B:</th>
<th>G:</th>
<th>rats:</th>
<th>died</th>
</tr>
</thead>
<tbody>
<tr>
<td>100: Yeast</td>
<td>1.0g:</td>
<td>12.6g:</td>
<td>4.2:</td>
<td>4.2:</td>
<td>6:</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>99: Auto. yeast</td>
<td>1.0g:</td>
<td></td>
<td></td>
<td></td>
<td>6:</td>
<td>6</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>104: Fraction 3A</td>
<td>0.2g:</td>
<td>1.2g:</td>
<td>2.0:</td>
<td>4:</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>105: **Fraction 3A</td>
<td>0.2g:</td>
<td>5.7g:</td>
<td>9.4:</td>
<td>4:</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>106: Fraction 3B</td>
<td>1.0g:</td>
<td>8.4g:</td>
<td>2.9:</td>
<td>4:</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>107: # Fraction 3B</td>
<td>1.0g:</td>
<td>10.9g:</td>
<td>3.5:</td>
<td>4:</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>108: Fraction 3C</td>
<td>1.0g:</td>
<td>2.3g:</td>
<td>7.8:</td>
<td>4:</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>109: # Fraction 3C</td>
<td>1.0g:</td>
<td>7.8g:</td>
<td>2.6:</td>
<td>4:</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>171: Fraction 4A</td>
<td>10%:</td>
<td>3.6g:</td>
<td>1.7:</td>
<td>6:</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>172: # Fraction 4A</td>
<td>10%:</td>
<td>10.6g:</td>
<td>3.8:</td>
<td>6:</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>173: Fraction 4B</td>
<td>0.1g:</td>
<td>1.8g:</td>
<td>5.8:</td>
<td>6:</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>174: * Fraction 4B</td>
<td>0.1g:</td>
<td>5.5g:</td>
<td>18.2:</td>
<td>6:</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>175: Fraction 5A</td>
<td>10%:</td>
<td>10.2g:</td>
<td>13.28:</td>
<td>6:</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>176: # Fraction 5A</td>
<td>10%:</td>
<td>11.4g:</td>
<td>3.0:</td>
<td>6:</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>177: Fraction 5B</td>
<td>5%:</td>
<td></td>
<td></td>
<td>6:</td>
<td>5</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>178: * Fraction 5B</td>
<td>5%:</td>
<td>2.8g:</td>
<td>3.9:</td>
<td>6:</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Denotes 10% autoclaved yeast added as source of vitamin G.

** Denotes 1g. autoclaved yeast per rat per day added as source of vitamin G.

# Denotes 0.2g. fraction 1D per rat per day added as source of vitamin B.

### Denotes 1.0g. fraction 7A per rat per day added as source of vitamin B.
Table III

Rice polishings and preparations from rice polishings as sources of vitamin B and G.

<table>
<thead>
<tr>
<th>Lot No.: Material tested</th>
<th>Avg. gain: Units per gram: per week</th>
<th>No. of : No. of rats: died</th>
</tr>
</thead>
<tbody>
<tr>
<td>121: Rice polishings</td>
<td>5%: 4.9g: 5.9: 6 : 1</td>
<td></td>
</tr>
<tr>
<td>122: **Rice polishings</td>
<td>5%: 12.0g: 10.0: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>123: Fraction 6B</td>
<td>0.05g: 2.8g: 18.8: 6 : 1</td>
<td></td>
</tr>
<tr>
<td>124: ***Fraction 6B</td>
<td>0.05g: 5.6g: 56.6: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>125: Fraction 6A</td>
<td>0.35g: 3.6g: 3.3: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>126: ***Fraction 6A</td>
<td>0.35g: 2.7g: 2.6: 6 : 1</td>
<td></td>
</tr>
<tr>
<td>127: Fraction 6C</td>
<td>0.9 g: 5.8g: 2.1: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>128: ***Fraction 6C</td>
<td>0.9 g: 7.4g: 2.8: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>163: Fraction 7B</td>
<td>0.1 g: 0.8g: 2.7: 6 : 1</td>
<td></td>
</tr>
<tr>
<td>164:* Fraction 7B</td>
<td>0.1 g: 9.6g: 31.6: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>165: Fraction 7A</td>
<td>10%: 2.8g: 1.4: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>166:* Fraction 7A</td>
<td>10%: 10.6g: 4.4: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>167: Fraction 7C</td>
<td>10%: 9.6g: 5.4: 6 : 2</td>
<td></td>
</tr>
<tr>
<td>168: ***Fraction 7C</td>
<td>10%: 7.7g: 3.2: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>169: # Fraction 7C</td>
<td>10%: 15.3g: 4.4: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>170: Auto. yeast</td>
<td>10%: 7.9g: 6 : 5</td>
<td></td>
</tr>
<tr>
<td>184: Fraction 3B</td>
<td>0.055g: 4.2g: 26.4: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>185: * Fraction 3B</td>
<td>0.055g: 15.0g: 94.2: 6 : 1</td>
<td></td>
</tr>
<tr>
<td>186: Fraction 3A</td>
<td>10%: 10.7g: 4.0: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>187: * Fraction 3A</td>
<td>10%: 14.6g: 4.5: 6 : 2</td>
<td></td>
</tr>
<tr>
<td>188: No supplement</td>
<td>-0.5g: -0.5: 6 : 5</td>
<td></td>
</tr>
<tr>
<td>194: Fraction 3B</td>
<td>0.027g: 4.1g: 50.0: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>195: * Fraction 3B</td>
<td>0.027g: 7.1g: 36.0: 6 : 0</td>
<td></td>
</tr>
<tr>
<td>208: No supplement</td>
<td>-0.5g: -0.5: 6 : 5</td>
<td></td>
</tr>
<tr>
<td>209: Auto. yeast</td>
<td>10%: 3.0g: 49.3: 6 : 3</td>
<td></td>
</tr>
<tr>
<td>210: No supplement</td>
<td>-0.5g: -0.5: 6 : 5</td>
<td></td>
</tr>
<tr>
<td>211: Fraction 3B</td>
<td>0.027g: 3.1g: 36.1: 6 : 2</td>
<td></td>
</tr>
</tbody>
</table>

* Denotes 10% autoclaved yeast added as a source of vitamin G

** Denotes 5% autoclaved yeast added as a source of vitamin G

*** Denotes 1.0g. autoclaved yeast per rat per day added as a source of vitamin G

## Denotes 10% fraction 7A added as a source of vitamin B
Table III Continued

Rice polishings and preparations from rice polishings as sources of vitamin B and G.

<table>
<thead>
<tr>
<th>Lot</th>
<th>Material tested</th>
<th>Avg. gain: Units per: No.: No. of</th>
<th>per rat: gram: of: rats: per week: B: C: rats: died</th>
</tr>
</thead>
<tbody>
<tr>
<td>212</td>
<td>Fraction 9B</td>
<td>0.027g.: 0.8g.: 10.4: 6: 2</td>
<td></td>
</tr>
<tr>
<td>213*</td>
<td>Fraction 9B</td>
<td>0.027g.: 7.8g.: 96.0: : 6: 1</td>
<td></td>
</tr>
<tr>
<td>214</td>
<td>Fraction 9A</td>
<td>10%; -1.2g.: : 6: 5</td>
<td></td>
</tr>
<tr>
<td>215*</td>
<td>Fraction 9A</td>
<td>10%; -0.2g.: : 6: 4</td>
<td></td>
</tr>
<tr>
<td>216</td>
<td>Fraction 9C</td>
<td>5%; : : 6: 6</td>
<td></td>
</tr>
<tr>
<td>217*</td>
<td>Fraction 9C</td>
<td>5%; -0.16g.: : 6: 5</td>
<td></td>
</tr>
<tr>
<td>229</td>
<td>Rice polishings</td>
<td>10%; 6.9g.: 4.2: 6: 0</td>
<td></td>
</tr>
<tr>
<td>229</td>
<td>Fraction 11A</td>
<td>2%; : : : :</td>
<td></td>
</tr>
<tr>
<td>229</td>
<td>Rice polishings</td>
<td>10%; 17.5g.: 7.03: : 6: 0</td>
<td></td>
</tr>
<tr>
<td>231</td>
<td>Fraction 8B</td>
<td>0.055g.: 2.0g.: 12.4: 6: 1</td>
<td></td>
</tr>
<tr>
<td>232*</td>
<td>Fraction 8B</td>
<td>0.055g.: 8.2g.: 61.2: : 6: 0</td>
<td></td>
</tr>
<tr>
<td>233*</td>
<td>Fraction 8B</td>
<td>0.055g.: 14.5g.: 96.6: : 6: 2</td>
<td></td>
</tr>
<tr>
<td>233*</td>
<td>Fraction 8B</td>
<td>0.055g.: : : : :</td>
<td></td>
</tr>
<tr>
<td>266</td>
<td>Fraction 8C</td>
<td>10%; 11.7g.: : : 6: 0</td>
<td></td>
</tr>
<tr>
<td>267</td>
<td>Fraction 8C</td>
<td>10%; -1.5g.: : : 6: 5</td>
<td></td>
</tr>
<tr>
<td>268</td>
<td>Fraction 8C</td>
<td>10%; 1.3g.: 1.8: 6: 1</td>
<td></td>
</tr>
<tr>
<td>269</td>
<td>Fraction 10A</td>
<td>10%; 2.4g.: 1.8: 6: 0</td>
<td></td>
</tr>
<tr>
<td>262*</td>
<td>Fraction 10A</td>
<td>10%; 13.8g.: 6.11: : 6: 0</td>
<td></td>
</tr>
</tbody>
</table>

# Denotes 0.053g. fraction 8B per rat per day added as a source of vitamin B.

* Denotes 10% autoclaved yeast added as a source of vitamin G.
Table IV

Preparations from hog liver as sources of vitamins B and G.

<table>
<thead>
<tr>
<th>Lot No.</th>
<th>Material tested</th>
<th>Avg. gain (g)</th>
<th>Units per gram per rat per week</th>
<th>No. of rats</th>
<th>No. of rats died</th>
</tr>
</thead>
<tbody>
<tr>
<td>196:##</td>
<td>Fraction IIA</td>
<td>0.1g±</td>
<td>9.0g±</td>
<td>30.7</td>
<td>6</td>
</tr>
<tr>
<td>197:##</td>
<td>Fraction IIA</td>
<td>0.2g±</td>
<td>11.5g±</td>
<td>12.7</td>
<td>6</td>
</tr>
<tr>
<td>198:##</td>
<td>Fraction IIC</td>
<td>0.25g±</td>
<td>7.5g±</td>
<td>10.0</td>
<td>6</td>
</tr>
<tr>
<td>199:##</td>
<td>Fraction IIA</td>
<td>0.25g±</td>
<td>7.5±</td>
<td>5.5±</td>
<td>6</td>
</tr>
<tr>
<td>200:##</td>
<td>Fraction IIA</td>
<td>0.3g±</td>
<td>4.6g±</td>
<td>3.5±</td>
<td>6</td>
</tr>
<tr>
<td>201:##</td>
<td>Fraction IIB</td>
<td>0.3g±</td>
<td>8.8g±</td>
<td>9.8±</td>
<td>6</td>
</tr>
<tr>
<td>219:##</td>
<td>Fraction IIIB</td>
<td>0.05g±</td>
<td>3.4±</td>
<td>20.9±</td>
<td>6</td>
</tr>
<tr>
<td>220:*</td>
<td>Fraction IIIB</td>
<td>0.053g±</td>
<td>10.2g±</td>
<td>61.4±</td>
<td>6</td>
</tr>
<tr>
<td>221:</td>
<td>Fraction IID</td>
<td>0.025g±</td>
<td>4.5±</td>
<td>4.6±</td>
<td>6</td>
</tr>
<tr>
<td>222:##</td>
<td>Fraction IID</td>
<td>0.325g±</td>
<td>4.5±</td>
<td>4.6±</td>
<td>6</td>
</tr>
<tr>
<td>223:##</td>
<td>Fraction IIE</td>
<td>0.2g±</td>
<td>1.5g±</td>
<td>2.5±</td>
<td>6</td>
</tr>
<tr>
<td>224:##</td>
<td>Fraction IIE</td>
<td>0.2g±</td>
<td>6.7±</td>
<td>11.1±</td>
<td>6</td>
</tr>
<tr>
<td>267:##</td>
<td>Fraction IIF</td>
<td>0.041g±</td>
<td>4.7±</td>
<td>2.9±</td>
<td>6</td>
</tr>
<tr>
<td>259:##</td>
<td>Fraction IIF</td>
<td>0.541g±</td>
<td>7.5±</td>
<td>4.7±</td>
<td>6</td>
</tr>
<tr>
<td>250:##</td>
<td>Fraction IIG</td>
<td>0.325g±</td>
<td>5.1±</td>
<td>3.1±</td>
<td>6</td>
</tr>
<tr>
<td>251:##</td>
<td>Fraction IIG</td>
<td>0.6g±</td>
<td>1.7±</td>
<td>1.1±</td>
<td>6</td>
</tr>
<tr>
<td>252:##</td>
<td>Fraction IIG</td>
<td>0.5g±</td>
<td>7.8±</td>
<td>5.2±</td>
<td>6</td>
</tr>
<tr>
<td>253:###</td>
<td>Auto. yeast</td>
<td>10%</td>
<td>10.7±</td>
<td>4.5±</td>
<td>6</td>
</tr>
<tr>
<td>264:###</td>
<td>Auto. yeast</td>
<td>0.05g±</td>
<td>9.8±</td>
<td>6.1±</td>
<td>6</td>
</tr>
<tr>
<td>288:##</td>
<td>Fraction IIF</td>
<td>0.035g±</td>
<td>1.1±</td>
<td>1.1±</td>
<td>6</td>
</tr>
<tr>
<td>292:##</td>
<td>Fraction IIF</td>
<td>0.035g±</td>
<td>9.8±</td>
<td>9.8±</td>
<td>6</td>
</tr>
<tr>
<td>290:##</td>
<td>Fraction IIF</td>
<td>0.166g±</td>
<td>6.3±</td>
<td>12.6±</td>
<td>6</td>
</tr>
<tr>
<td>255:##</td>
<td>Fraction IIF</td>
<td>0.251±</td>
<td>4.8±</td>
<td>6.3±</td>
<td>6</td>
</tr>
<tr>
<td>256:##</td>
<td>Fraction IIF</td>
<td>0.251±</td>
<td>7.8±</td>
<td>10.6±</td>
<td>6</td>
</tr>
<tr>
<td>225:##</td>
<td>Fraction IIG</td>
<td>0.5g±</td>
<td></td>
<td>6</td>
<td>5</td>
</tr>
<tr>
<td>226:##</td>
<td>Fraction IIG</td>
<td>0.5g±</td>
<td>5.4±</td>
<td>3.6±</td>
<td>6</td>
</tr>
<tr>
<td>227:##</td>
<td>Fraction IIG</td>
<td>0.2g±</td>
<td>3.4±</td>
<td>5±</td>
<td>6</td>
</tr>
<tr>
<td>228:###</td>
<td>Fraction IIG</td>
<td>0.2g±</td>
<td>9.5±</td>
<td>13.6±</td>
<td>6</td>
</tr>
</tbody>
</table>

* Denotes 10% autoclaved yeast added as a source of vitamin G.

# Denotes 0.053g. fraction IIIB per rat per day added as a source of vitamin B.

## Denotes 0.027g. fraction IIIB per rat per day added as a source of vitamin B.

### Denotes 10% fraction IIA added as a source of vitamin B.
Table IV Continued

Preparations from hog liver as sources of vitamins B and G.

<table>
<thead>
<tr>
<th>Lot No.</th>
<th>Material tested</th>
<th>Avg. gain: Units per:</th>
<th>per rat:</th>
<th>gram:</th>
<th>No. of rats:</th>
<th>died</th>
</tr>
</thead>
<tbody>
<tr>
<td>232‡</td>
<td>Fraction 12B</td>
<td>0.55g.:</td>
<td>3.2g.:</td>
<td>3.2:</td>
<td>6: 0</td>
<td></td>
</tr>
<tr>
<td>254‡</td>
<td>Fraction 12A</td>
<td>0.4g.:</td>
<td>5.6g.:</td>
<td>4.7:</td>
<td>6: 0</td>
<td></td>
</tr>
<tr>
<td>234‡</td>
<td>Fraction 13B</td>
<td>0.5g.:</td>
<td>3.7g.:</td>
<td>2.4:</td>
<td>6: 2</td>
<td></td>
</tr>
<tr>
<td>236‡</td>
<td>Fraction 13B</td>
<td>0.5g.:</td>
<td>10.8g.:</td>
<td>7.2:</td>
<td>6: 0</td>
<td></td>
</tr>
<tr>
<td>237‡</td>
<td>Fraction 13C</td>
<td>0.33g.:</td>
<td>8.6g.:</td>
<td>8.6:</td>
<td>6: 0</td>
<td></td>
</tr>
<tr>
<td>238‡</td>
<td>Fraction 13A</td>
<td>0.33g.:</td>
<td>9.5g.:</td>
<td>9.5:</td>
<td>6: 0</td>
<td></td>
</tr>
<tr>
<td>239‡</td>
<td>Fraction 13B</td>
<td>0.2g.:</td>
<td>4.5g.:</td>
<td>7.5:</td>
<td>6: 0</td>
<td></td>
</tr>
<tr>
<td>240‡</td>
<td>Fraction 13F</td>
<td>0.33g.:</td>
<td>11.3g.:</td>
<td>11.3:</td>
<td>6: 0</td>
<td></td>
</tr>
<tr>
<td>242‡</td>
<td>Yeast</td>
<td>10%:</td>
<td>14.6g.:</td>
<td></td>
<td>6: 0</td>
<td></td>
</tr>
</tbody>
</table>

‡ Denotes 0.055g. fraction 8B per rat per day added a source of vitamin B.
Table V

Whey powder and preparations from whey powder as sources of Vitamin G

<table>
<thead>
<tr>
<th>Lot</th>
<th>Material tested</th>
<th>Avg. gain; Units per</th>
<th>per rat; gram; of</th>
<th>No. of rats; died</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.</td>
<td></td>
<td>per week; B; G</td>
<td></td>
<td></td>
</tr>
<tr>
<td>232</td>
<td>Whey powder</td>
<td>0.166g; 5.9g</td>
<td>11.7; 5; 1</td>
<td></td>
</tr>
<tr>
<td>263</td>
<td>Whey powder</td>
<td>0.125g; 2.6g</td>
<td>6.1; 6; 0</td>
<td></td>
</tr>
<tr>
<td>264</td>
<td>Whey powder</td>
<td>0.083g; 3.1g</td>
<td>12.3; 6; 0</td>
<td></td>
</tr>
<tr>
<td>303</td>
<td>Fraction 14A</td>
<td>0.166g; 3.2g</td>
<td>6.4; 6; 0</td>
<td></td>
</tr>
<tr>
<td>304</td>
<td>Fraction 14A</td>
<td>0.333g; 3.9g</td>
<td>3.9; 6; 0</td>
<td></td>
</tr>
<tr>
<td>305</td>
<td>Fraction 14B</td>
<td>0.065g; 2.3g</td>
<td>9.2; 6; 0</td>
<td></td>
</tr>
<tr>
<td>306</td>
<td>Fraction 14B</td>
<td>0.166g; 5.9g</td>
<td>10.6; 6; 0</td>
<td></td>
</tr>
</tbody>
</table>

* Indicates 0.052g. fraction 3B fed per rat per day added as a source of vitamin E.
Table VI

Lactation on diets containing wheat, yeast and autoclaved yeast.

<table>
<thead>
<tr>
<th>Lot No.</th>
<th>Sources of vitamins</th>
<th>No. of dams</th>
<th>No. of liters</th>
<th>Avg. wt.</th>
<th>Percent young weaned</th>
<th>Weaning mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>53A</td>
<td>Wheat</td>
<td>30%</td>
<td>6 : 36</td>
<td>16 : 34.2</td>
<td>55.5</td>
<td></td>
</tr>
<tr>
<td>54A</td>
<td>Wheat</td>
<td>30%</td>
<td>6 : 36</td>
<td>30 : 55.5</td>
<td>15.6</td>
<td></td>
</tr>
<tr>
<td>55A</td>
<td>Yeast</td>
<td>30%</td>
<td>6 : 36</td>
<td>31 : 55.4</td>
<td>13.9</td>
<td></td>
</tr>
<tr>
<td>69</td>
<td>*Wheat</td>
<td>30%</td>
<td>6 : 36</td>
<td>3 : 26.0</td>
<td>77.7</td>
<td></td>
</tr>
<tr>
<td>94</td>
<td>Yeast Auto.</td>
<td>30%</td>
<td>6 : 36</td>
<td>30 : 50.0</td>
<td>16.6</td>
<td></td>
</tr>
</tbody>
</table>

* Contained 5% butterfat and no cod liver oil.
Table VII

Lactation on diets containing wheat, yeast and autoclaved yeast.

<table>
<thead>
<tr>
<th>Lot No.</th>
<th>Sources of vitamins</th>
<th>No. of B and G</th>
<th>No. of litters</th>
<th>No. of young</th>
<th>Avg. Wt. at Percent</th>
<th>Mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>116</td>
<td>Wheat</td>
<td>30%</td>
<td>27</td>
<td>161</td>
<td>41</td>
<td>31.3</td>
</tr>
<tr>
<td></td>
<td>Wheat</td>
<td>30%</td>
<td>27</td>
<td>161</td>
<td>41</td>
<td>31.3</td>
</tr>
<tr>
<td>117</td>
<td>Yeast Auto.</td>
<td>5%</td>
<td>36</td>
<td>206</td>
<td>35</td>
<td>53.6</td>
</tr>
<tr>
<td>118</td>
<td>Yeast Auto.</td>
<td>10%</td>
<td>9</td>
<td>49</td>
<td>32</td>
<td>40.5</td>
</tr>
<tr>
<td>144</td>
<td>Wheat</td>
<td>30%</td>
<td>27</td>
<td>158</td>
<td>74</td>
<td>32.9</td>
</tr>
<tr>
<td>145</td>
<td>Yeast</td>
<td>5%</td>
<td>20</td>
<td>115</td>
<td>94</td>
<td>56.0</td>
</tr>
<tr>
<td>146</td>
<td>Yeast</td>
<td>30%</td>
<td>4</td>
<td>139</td>
<td>117</td>
<td>58.4</td>
</tr>
<tr>
<td>147</td>
<td>Yeast Auto</td>
<td>5%</td>
<td>15</td>
<td>79</td>
<td>43</td>
<td>46.8</td>
</tr>
<tr>
<td>148</td>
<td>Yeast Auto</td>
<td>10%</td>
<td>27</td>
<td>159</td>
<td>156</td>
<td>55.0</td>
</tr>
</tbody>
</table>

Data reported in this table were obtained from rats reared on the experimental rations.
Table VIII

Lactation on diets containing wheat and yeast.

<table>
<thead>
<tr>
<th>Lot</th>
<th>Sources of vitamins</th>
<th>No. of B and G</th>
<th>No. of:</th>
<th>Avg. Wt.:</th>
<th>Percent:</th>
<th>Weaned:</th>
<th>Weaning:</th>
<th>Mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>53</td>
<td>Wheat</td>
<td>60%: 11</td>
<td>66</td>
<td>43</td>
<td>50.6</td>
<td>34.8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>54</td>
<td>Wheat</td>
<td>60%:</td>
<td>6</td>
<td>36</td>
<td>55.1</td>
<td>0.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>55</td>
<td>Yeast</td>
<td>60%:</td>
<td>6</td>
<td>36</td>
<td>65.0</td>
<td>0.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>56</td>
<td>Wheat</td>
<td>10%:</td>
<td>6</td>
<td>36</td>
<td>63.0</td>
<td>0.0</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>


**Table IX**

Lactation on diets containing wheat yeast and autoclaved yeast.

<table>
<thead>
<tr>
<th>Lot No.</th>
<th>Vitamins</th>
<th>No. of B and G</th>
<th>No. of Mice</th>
<th>No. of Young</th>
<th>Avg. Wt. in g.</th>
<th>Percent Weaned</th>
<th>Weaning Mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>70</td>
<td>Wheat</td>
<td>32</td>
<td>188</td>
<td>94</td>
<td>48.0</td>
<td>60.0</td>
<td></td>
</tr>
<tr>
<td>71</td>
<td>Wheat</td>
<td>21</td>
<td>120</td>
<td>113</td>
<td>59.3</td>
<td>5.8</td>
<td></td>
</tr>
<tr>
<td>72</td>
<td>Yeast</td>
<td>28</td>
<td>174</td>
<td>164</td>
<td>62.6</td>
<td>5.7</td>
<td></td>
</tr>
<tr>
<td>113</td>
<td>Wheat</td>
<td>19</td>
<td>113</td>
<td>79</td>
<td>34.5</td>
<td>30.1</td>
<td></td>
</tr>
<tr>
<td>114</td>
<td>Yeast</td>
<td>31</td>
<td>177</td>
<td>166</td>
<td>55.0</td>
<td>6.2</td>
<td></td>
</tr>
<tr>
<td>115</td>
<td>Yeast</td>
<td>29</td>
<td>164</td>
<td>155</td>
<td>58.0</td>
<td>5.5</td>
<td></td>
</tr>
</tbody>
</table>

Data reported in this table were obtained from rats raised on the experimental rations.
Table X

Rice polishings, wheat germ, rice bran and corn germ as sources of vitamins B and G for lactation.

<table>
<thead>
<tr>
<th>Lot No.</th>
<th>Sources of vitamins</th>
<th>% of</th>
<th>No. of</th>
<th>Avg. Wt.</th>
<th>Liters</th>
<th>No. of young</th>
<th>in g.</th>
<th>at</th>
<th>Percent</th>
<th>Weaned</th>
<th>Weaning</th>
<th>Mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>140</td>
<td>Rice polishings</td>
<td>10%</td>
<td>12</td>
<td>72</td>
<td>50</td>
<td>39.5</td>
<td>30.6</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>140A</td>
<td>Yeast Auto.</td>
<td>10%</td>
<td>12</td>
<td>72</td>
<td>70</td>
<td>56.3</td>
<td>2.9</td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>141</td>
<td>Wheat germ</td>
<td>10%</td>
<td>12</td>
<td>72</td>
<td>27</td>
<td>47.4</td>
<td>62.5</td>
<td></td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>141A</td>
<td>Yeast Auto.</td>
<td>10%</td>
<td>12</td>
<td>72</td>
<td>66</td>
<td>56.8</td>
<td>8.3</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>142</td>
<td>Rice bran</td>
<td>10%</td>
<td>12</td>
<td>72</td>
<td>40</td>
<td>46.7</td>
<td>44.4</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>142A</td>
<td>Yeast Auto.</td>
<td>10%</td>
<td>12</td>
<td>72</td>
<td>72</td>
<td>54.1</td>
<td>0.0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>143</td>
<td>Corn germ</td>
<td>10%</td>
<td>12</td>
<td>72</td>
<td>37</td>
<td>22.5</td>
<td>44.6</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>143A</td>
<td>Yeast Auto.</td>
<td>10%</td>
<td>12</td>
<td>72</td>
<td>44</td>
<td>27.2</td>
<td>38.3</td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>
Table XI

Oatmeal, yellow corn, white corn and barley as sources of vitamins B and G for lactation

<table>
<thead>
<tr>
<th>Lot No.</th>
<th>Sources of vitamins</th>
<th>No. of:</th>
<th>wt.</th>
<th>No. of:</th>
<th>Avg. wt.</th>
<th>Percent of survivors</th>
<th>Percent of young weaned</th>
<th>Weaning mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>150</td>
<td>Stock ration</td>
<td>6</td>
<td>36</td>
<td>30</td>
<td>44.4</td>
<td>16.6</td>
<td></td>
<td></td>
</tr>
<tr>
<td>151</td>
<td>Oatmeal</td>
<td>60%</td>
<td>8</td>
<td>48</td>
<td>45.2</td>
<td>16.6</td>
<td></td>
<td></td>
</tr>
<tr>
<td>152</td>
<td>Oatmeal</td>
<td>60%</td>
<td>6</td>
<td>36</td>
<td>49.0</td>
<td>16.6</td>
<td></td>
<td></td>
</tr>
<tr>
<td>153</td>
<td>Yeast Auto.</td>
<td>5%</td>
<td>6</td>
<td>36</td>
<td>49.0</td>
<td>16.6</td>
<td></td>
<td></td>
</tr>
<tr>
<td>154</td>
<td>Yeast Auto.</td>
<td>10%</td>
<td>8</td>
<td>48</td>
<td>51.3</td>
<td>0.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>155</td>
<td>Yellow corn</td>
<td>60%</td>
<td>12</td>
<td>72</td>
<td>36.0</td>
<td>72.2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>156</td>
<td>Yeast Auto.</td>
<td>5%</td>
<td>12</td>
<td>72</td>
<td>48.4</td>
<td>9.7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>157</td>
<td>White corn</td>
<td>60%</td>
<td>12</td>
<td>72</td>
<td>41.1</td>
<td>62.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>158</td>
<td>Yeast Auto.</td>
<td>5%</td>
<td>12</td>
<td>72</td>
<td>55.2</td>
<td>8.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>159</td>
<td>Yeast Auto.</td>
<td>60%</td>
<td>12</td>
<td>72</td>
<td>55.2</td>
<td>8.5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>160</td>
<td>Barley</td>
<td>60%</td>
<td>12</td>
<td>72</td>
<td>38.1</td>
<td>54.7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>161</td>
<td>Yeast Auto.</td>
<td>5%</td>
<td>12</td>
<td>72</td>
<td>44.7</td>
<td>11.1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>162</td>
<td>Yeast Auto.</td>
<td>10%</td>
<td>12</td>
<td>72</td>
<td>54.3</td>
<td>8.3</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Table XII

Influence of storage of vitamin G on lactation.

<table>
<thead>
<tr>
<th>Lot</th>
<th>No. of vitamins</th>
<th>No. of young</th>
<th>in g. at</th>
<th>Percent</th>
<th>Weaned</th>
<th>Weaning</th>
<th>Mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>154A: Yellow corn</td>
<td>60%</td>
<td>6 : 36 : 12</td>
<td>36.7</td>
<td>66.6</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>154B: Yellow corn</td>
<td>60%</td>
<td>6 : 36 : 28</td>
<td>43.0</td>
<td>22.2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>154C: Yellow corn</td>
<td>60%</td>
<td>6 : 36 : 32</td>
<td>46.5</td>
<td>11.1</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Denotes stock ration for seven days after parturition.
** Denotes stock ration for fourteen days after parturition.
Table IX

<table>
<thead>
<tr>
<th>Time (h)</th>
<th>Percent</th>
<th>Recovery</th>
<th>Recovery</th>
<th>Percent</th>
<th>Recovery</th>
<th>Recovery</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
<td>10.0</td>
<td>60.0</td>
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<td>60.0</td>
<td>6.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
</tr>
<tr>
<td>3.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
</tr>
<tr>
<td>4.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
</tr>
<tr>
<td>5.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
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<tr>
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<td>60.0</td>
<td>6.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
</tr>
<tr>
<td>7.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
</tr>
<tr>
<td>8.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
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<tr>
<td>9.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
<td>10.0</td>
<td>60.0</td>
<td>6.0</td>
</tr>
</tbody>
</table>

Note: Percent recovery in E. coli: 100.

Footnote: 1200 mg of E. coli, 100 mg of E. coli.

Table IX. Recovery of E. coli from 10 mg of E. coli.
Table XIV

Lactation on diets containing fraction 8B, fraction 1LF1, and fraction 1LF2.

<table>
<thead>
<tr>
<th>Lot:</th>
<th>Source of vitamins</th>
<th>% of</th>
<th>B and C</th>
<th>% of</th>
<th>Young chicks feeding mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td>274:</td>
<td>Fraction 8B</td>
<td>0.055:</td>
<td>4 : 24 : 0</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>294:</td>
<td>Fraction 8B</td>
<td>0.055:</td>
<td>6 : 56 : 0</td>
<td>1 : 28.6</td>
<td>18.6 : 51.6 : 58.3</td>
</tr>
<tr>
<td>186:</td>
<td>Fraction 1LF1</td>
<td>0.055:</td>
<td>10 : 60 : 0</td>
<td>34 : 20.7</td>
<td>43.8 : 44.4 : 97.7</td>
</tr>
<tr>
<td>187:</td>
<td>Fraction 1LF1</td>
<td>0.055:</td>
<td>10 : 60 : 0</td>
<td>35.2 : 28.9 : 44.4</td>
<td></td>
</tr>
<tr>
<td>244:</td>
<td>Fraction 8B</td>
<td>0.212:</td>
<td>10 : 60 : 0</td>
<td>34 : 20.7</td>
<td>43.8 : 44.4 : 97.7</td>
</tr>
<tr>
<td>246:</td>
<td>Fraction 1LF1</td>
<td>0.212:</td>
<td>10 : 60 : 0</td>
<td>34.2 : 28.9 : 44.4</td>
<td></td>
</tr>
<tr>
<td>298:</td>
<td>Fraction 8B</td>
<td>0.212:</td>
<td>10 : 60 : 0</td>
<td>34.2 : 28.9 : 44.4</td>
<td></td>
</tr>
<tr>
<td>276:</td>
<td>Fraction 8B</td>
<td>0.212:</td>
<td>10 : 60 : 0</td>
<td>34.2 : 28.9 : 44.4</td>
<td></td>
</tr>
<tr>
<td>296:</td>
<td>Fraction 1LF1</td>
<td>0.212:</td>
<td>10 : 60 : 0</td>
<td>34.2 : 28.9 : 44.4</td>
<td></td>
</tr>
</tbody>
</table>

*Note: The table contains data on the percentage of young chicks and feeding mortality for different lots and sources of vitamins.*
Table XV
Lactation on diets containing wheat, yellow corn, 
fraction 8B and fraction 11F1.

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>309</td>
<td>Yellow corn Fraction 8B 0.318g.</td>
<td>60%: 6</td>
<td>36 :</td>
<td>4 :</td>
<td>30.2:</td>
<td>38.8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>310</td>
<td>Yellow corn Fraction 11F1 1.66g.</td>
<td>60%: 6</td>
<td>36 :</td>
<td>32 :</td>
<td>54.5:</td>
<td>11.1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>311</td>
<td>Yellow corn</td>
<td>60%: 5</td>
<td>30 :</td>
<td>10 :</td>
<td>27.2:</td>
<td>66.6</td>
<td></td>
<td></td>
</tr>
<tr>
<td>312</td>
<td>Wheat Fraction 8B 0.318g.</td>
<td>60%: 6</td>
<td>36 :</td>
<td>22 :</td>
<td>29.8:</td>
<td>38.8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>313</td>
<td>Wheat Fraction 11F1 1.66g.</td>
<td>60%: 6</td>
<td>36 :</td>
<td>36 :</td>
<td>53.8:</td>
<td>0.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>314</td>
<td>Wheat</td>
<td>60%: 6</td>
<td>36 :</td>
<td>16 :</td>
<td>27.5:</td>
<td>55.5</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Table XVI

Hemoglobin values at parturition and during lactation.

<table>
<thead>
<tr>
<th>Lot</th>
<th>Sources of</th>
<th>Hemoglobin g. per 100c.c. blood</th>
<th>Partu-</th>
<th>7th</th>
<th>14th</th>
<th>21th</th>
<th>28th</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.</td>
<td>vitamins B and G</td>
<td>ration; day; day; day; day</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>179</td>
<td>No supplement</td>
<td>13.9; 14.3; 12.1; 11.3; 13.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>180</td>
<td>Yellow corn</td>
<td>60%; 14.2; 12.0; 11.3; 11.9; 13.0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Yellow corn</td>
<td>60%;</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>181</td>
<td>Yeast Auto.</td>
<td>5%; 12.3; 12.9; 13.6; 13.3; 13.9</td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td></td>
<td>Yellow corn</td>
<td>60%;</td>
<td></td>
<td></td>
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<td></td>
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</tr>
<tr>
<td>182</td>
<td>Yeast Auto.</td>
<td>10%; 12.8; 13.6; 12.2; 13.1; 14.3</td>
<td></td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
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<td>Stock ration</td>
<td>12.4; 13.5; 12.6; 12.6; 11.7</td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>309</td>
<td>Fraction 8B</td>
<td>0.218g.</td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Yellow corn</td>
<td>60%; 13.4; 13.8; 14.6; 15.3; 15.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>310</td>
<td>Fraction 11F</td>
<td>1.66g.</td>
<td>13.9; 14.3; 15.6; 16.4; 16.6</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Yellow corn</td>
<td>60%;</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>311</td>
<td>Yellow corn</td>
<td>60%; 13.3; 14.5; 14.9; 15.8; 16.6</td>
<td></td>
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<td></td>
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<td></td>
</tr>
<tr>
<td></td>
<td>Wheat</td>
<td>60%;</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>312</td>
<td>Fraction 8B</td>
<td>0.318g.</td>
<td>13.8; 14.4; 15.2; 16.3; 16.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Wheat</td>
<td>60%;</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>313</td>
<td>Fraction 11F</td>
<td>1.66g.</td>
<td>14.1; 15.0; 15.8; 16.4; 17.0</td>
<td></td>
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<td></td>
</tr>
<tr>
<td></td>
<td>Wheat</td>
<td>60%;</td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>314</td>
<td>Wheat</td>
<td>60%; 14.0; 15.2; 15.1; 16.2; 16.6</td>
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</tbody>
</table>