STUDIES IN MINK NUTRITION WITH SPECIAL REFERENCE TO SUPPLEMENTARY PROTEIN SOURCES

by

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ABSTRACT

The primary objective of this study was to compare horse meat, whale meat, chicken wastes and herring as supplementary animal protein sources in rations for mink during maintenance, reproduction and growth. Part of the rations were canned and part of the rations contained a commercial antioxidant.

The results of the maintenance experiment indicated that there were no statistical differences between the rations. In the reproductive phase of the experiment, the mink receiving the canned rations had almost complete reproductive failure whereas the mink receiving the frozen rations had a kit crop that was below average.

There were significant differences between the rations in the growth phase of the experiment. The rations containing Horse Meat, Whale Meat and Chicken Waste A were superior to those containing Chicken Waste B and Herring. The difference between the two chicken wastes has been attributed to the variability of the composition of chicken waste. The below average performance of the mink receiving the Herring ration was not due to protein of inferior quality, as evidenced by its high Net Protein Utilization value, but probably due to the results of oxidative rancidity.

The addition of the commercial antioxidant to the rations had no significant effect on the performance of the mink during maintenance and reproduction but did retard the growth of the kits. The antioxidant failed to prevent the destruction of

thiamine in the frozen and canned rations but did afford some protection for Vitamin A in the canned rations.

The Net Protein Utilization values for the fat extracted supplementary protein sources differed considerably. Herring was the highest and was followed by Horse Meat and Whale Meat, Chicken Waste A and Chicken Waste B.

The digestibility trials indicate little differences between the rations except for fat digestibility. The high fat digestibility suggests that mink can utilize high levels of fat from varying sources. The low "carbohydrate" digestibility indicates that mink may have difficulty in utilizing the cereal portion of their rations.

The variability in feed consumption of the mink receiving the canned rations was attributed to the differences in the digestible energy contents of the rations. Regression equations relating feed consumption to body weight were calculated for the various ration groups.

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I. Introduction

Mink are the most important of the semi-domesticated animals raised for the production of fur. Economical and efficient production is contingent upon maximum production per unit of feed. Large litters, heavy weaning weights, continuous and rapid growth of kits make for profit and are largely the result of adequate nutrition. Information on the nutritional requirements of mink are scanty and are in a continuous state of revision as more information becomes available.

This lack of information stems, in part, from the recent domestication of this species. As with other carnivores, such as the dog and cat, there is a surprising dearth of information needed to permit formulation of adequate practical rations. Recently, The United States National Research Council has prepared a Recommended Nutrient Allowance for Minks (91). The many incomplete sections in this publication, further testifies to the inadequacy of our information to date. The present study attempts to extend our limited knowledge of the nutritive needs of this animal. Of necessity, some of the work has had to be of an exploratory nature in the hope that the results might provide a basis for further investigations and ultimately lead to a more precise statement of the nutritive needs of the mink.

The diet of the mink is normally mixed as a semi-fluid mass and contains from 30-40 per cent dry matter. Attempts have been made to formulate dry diets but none have been developed to

the point that they will successfully carry a mink through its complete life cycle (24, 25). The failures to date may have been due to the lack of essential unknown factors such as those shown to be present in liver and hog mucosa (38).

Little or no back-log of basic information is available on the mink's nutritive needs and research in this field, of necessity, has had to follow two pathways. Investigations using synthetic diets are an essential prerequisite for the determination of absolute nutritive needs. However, such studies are likely to be time consuming and expensive. There is much that can be learnt of direct immediate benefit to the industry from experimental work conducted on natural dietaries.

The primary objective of this study has been to seek out information that would ultimately lead to the resolution of the problems created by the diminishing supply and increasing cost of raw animal protein. In the past, the main source of such protein has been horse meat. Rations formulated with a high level of this meat have given fairly satisfactory results. Recently, mink ranchers have been forced to look for alternative animal protein sources because the diminishing horse population has been unable to provide adequate supplies of meat to the expanding mink industry. The substitutes used include whale meat, chicken wastes, various types of fish, tripe, lungs and meat scraps. Little information is available on the relative worth of these substitutes in comparison to horse meat and thus the primary objective of this inves-

tigation has been to compare some of these substitutes with horse meat for maintenance, reproduction and growth of the mink.

The wet diets fed to mink present chemical and microbial problems that are not encountered in the dry rations formulated for other species. In addition, most mink rations are relatively high in fat. The fatty portions of meat and fish products become oxidized in the presence of labile oxygen and form hydroperoxides (27) which are capable of oxidizing such vitamins as A, D, E, Ascorbic Acid and Biotin (43, 61). In rations designed to test the relative worth of proteins, the destruction of such micronutrients might lead to reproductive and growth failures that could mistakenly be attributed to inadequacy of the proteins. Support for this view can be found at the practical level where evidence has been obtained to show that these oxidative changes may give rise to a variety of pathological conditions in the mink, such as "yellow fat disease." (9) Various Vitamin E preparations are being used to fortify mink cereals on the assumption that they reduce the incidence of this disease. The use of Vitamin E for this purpose has led to confusion, for it has not been possible to demonstrate whether the added Vitamin E is used as a vitamin or as an antioxidant. (9) It seems unlikely that it could be acting as a vitamin because of the high level of supplementation. If the vitamin is serving as an antioxidant it would seem more provident to use one of the many cheaper commercial antioxidants. Thus, a second objective of the present work

has been to determine if one of the commercial antioxidants would afford protection to the labile non-protein constituents of mink rations and in so doing indirectly aid in the evaluation of the proteins.

The problem of spoilage is another factor which might complicate the evaluation of the protein sources in a wet ration. The normal wet ration provides an excellent menstruum for bacterial growth. Severe losses have been recorded from botulism and such outbreaks have frequently been attributed to the microbial contamination carried by the animal protein sources. If this were the case in any of the protein sources under test, reproductive or growth failure due to bacterial activity could mistakenly be attributed to an inadequacy of the proteins. In an effort to prevent such a complication a part of the rations were canned in a manner similar to that used for commercial dog and cat feeds.

II. The Formulation and Composition of the Experimental Rations

There is a great variety of diets fed to mink. The most important ingredients used include fish, red meats, liver and cereals in various proportions. Due to the unavailability and high cost of horse meat many ranchers are omitting it from their rations and are using replacements, such as poultry by-products, rabbits, whale meat and other animal and vegetable protein sources. Day (12) has recorded the seasonal variation in feeding programmes in the Pacific North West and they are presented in Table I.

Table I

The Seasonal Variation of Mink Feeding Programmes

U.S. Pacific Northwest

Constituent	Pelting to Weaning	Weaning to Pelting
	Per Cent	Per Cent
Fish	87 to 0	90 to 40
Red Meats	5 to 75	0 to 25
Beef or Pork Liver	2 to 12	0 to 10
Cereals	6 to 15	6 to 25

The great variation in the ingredients and the proportions in which they are offered makes it difficult to formulate a representative ration.

The experimental rations developed for this study consisted of a cereal portion, liver and a supplementary protein source. Water was added to yield a feed having a hamburger-like consistency. They differ from the normal ranch mink rations in

that a single raw source of protein was used instead of the usual mixture. The National Research Council Recommended Nutrient Requirements for Foxes and Minks (49) were used as a guide for the formulation of the rations.

1. Development of the Cereal Portion

The National Research Council basic cereal mix (49) was used as a guide in formulating the cereal portion of the rations. Its constituent composition is given in Table 2.

Table 2

National Research Council Basic Cereal Mix for Mink

Wheat (cooked, dried)	1415.0
Oatmeal, cooked	462.0
Salt, iodized	12.5
Bonemeal, steamed	10.4
Limestone	20.8
Alfalfa Meal	45.9
Yeast, Brewers	20.9
Cod Liver Oil	12.5
	2000.0 lbs.

Several modifications of this mix were made before the final cereal mix was formulated. Since cooked wheat and cooked oatmeal are difficult to obtain in this area, ground wheat and oat groats were substituted. Table 3 gives what might be termed a Canadian counterpart for the United States recommendation given in Table 2.

Table 3

First Revision of the NRC Cereal Mix

Ground Wheat	1415.0
Oat Groats	462.0
Salt, iodized	12.5
Bone Meal, steamed	10.4
Limestone	20.8
Alfalfa Meal	45.9
Brewers Yeast	20.9
Fish Oil (1500 A) (400 D)	12.5
	2000.0 lbs.

Since the primary objective of the present investigation was to permit a comparison of the various raw protein sources it was necessary to ensure an adequate supply of all the other essential nutrients. For this reason the National Research Council (49) type cereal mix was further modified to include a variety of common feed ingredients in the hope that they would provide these other vitamins. The cereal was supplemented with choline, Vitamin B_{12} and folic acid since there have been suggestions that these vitamins might be required at higher levels in diets rich in fat.

The constituent composition of the cereal mix finally selected is given in Table 4.

Table 4

Composition of the Experimental Cereal Mix

Ground Wheat	680
Oat Groats	680
Fish Meal-Herring - 70%	240
Soya Bean Meal - 43%	100
Wheat Bran	100
Dehydrated Grass Meal	100
Dried Herring Solubles	30
Brewers Yeast	25
Limestone	20
Iodized Salt	12.5
Fish Oil (1500 A) (400 D)	12.5
	2000 lbs.
Supplemented/Ton	800 gm. Choline Chloride (200 gm. Choline). 4 lbs. Profactor B (40 mg. Vit. B_{12}). 20 gm. Parvo (600 mg. Folic Acid).

The dry matter content of the cereal mix was 91.4 per cent. On a dry weight basis the per cent protein was 23.8; fat 5.9; and ash 5.1. Various references were consulted for the calculation of the proximate composition of the cereal mix (3, 48, 49).

2. The Supplementary Protein Sources

The mink industry in Canada and the United States is relatively new and its early success was founded in part on a supply of readily available horse meat. For this reason, horse meat has become the standard animal protein source of the industry and other protein sources are usually compared relative to horse meat. The supplementary proteins tested in this study, were whale meat, two types of chicken waste and whole herring. Chicken Waste B differed from Chicken Waste A in that it had the gall bladder and anal portion removed from the viscera. Part of the chicken waste was obtained from the University Poultry Farm and the remainder from a local firm. Round Pacific Coast Fall Herring was obtained from a local fish reduction plant. Pacific Coast Sperm Whale meat of the type sold for mink feeding was used. The five supplementary protein sources were analyzed for dry matter, fat, ash and protein by standard procedures (14). Great difficulty was encountered in obtaining samples which were truly representative of these products because of the inherent variability in their composition. The results of duplicate analysis are presented in Table 5. These values were used for the calculations of the basic rations on the assumption that they would be indicative of the average composition.

Table 5

Partial Proximate Composition of the Five Protein Sources

Protein Source	Per Cent of Constituent							
	Protein		Fat		Ash		Dry Matter	
	Wet Basis	Dry Basis	Wet Basis	Dry Basis	Wet Basis	Dry Basis	Wet Basis	Dry Basis
Horse Meat	16.3	65.0	8.0	32.0	0.75	3.0	25.0	100.0
Whale Meat	24.0	80.0	5.1	17.0	0.87	2.9	30.0	100.0
Chicken A	17.2	44.0	17.2	44.0	3.9	10.0	39.0	100.0
Chicken B	15.9	43.0	17.0	46.0	3.6	9.8	37.0	100.0
Herring	17.9	69.0	5.5	21.0	2.3	8.8	26.0	100.0

3. Formulation of the Basic Rations

Five basic rations were formulated, one for each of the five supplementary protein sources. An attempt was made to formulate the rations so that they would be isocaloric with respect to protein, fat, and carbohydrate. The ash content was held constant.

The rations consisted of cereal, liver and the supplementary protein source mixed to a hamburger-like consistency with water. The National Research Council Recommended Nutrient Requirements for Mink (49) state that the average dry matter of a mink ration is 34 per cent. They also suggest a cereal level of 20 to 30 per cent of the wet ration. Table 1 suggests that many mink ranchers prefer a lower cereal level in their rations. The one

chosen for the five basic rations was 22.5 per cent of the wet ration which is 59 per cent of the dry ration when the dry matter is 34 per cent. The chosen cereal level was then the lower limit of the suggested National Research Council (49) range. The 7 per cent, recommended maximum level of liver (49) was chosen for the five basic rations. Thus, the cereal portion and liver have been added as a definite percentage of the dry ration, 59 and 7 per cent respectively. The remainder of the dry matter then arose from the supplementary protein source and any fat or bonemeal that were added to balance the rations so they would have the same percentage of fat, protein and ash. The National Research Council (49) has recommended rations that contain 16, 22 and 28 per cent protein on a dry weight basis for maintenance, reproduction and growth. Howell (21) has shown that the National Research Council levels are too low for maximum growth and suggest a protein level of 32 per cent on a dry weight basis for the most efficient growth. Higher levels of protein did not give any greater growth. A protein level of 33 per cent of the dry ration was selected for the five basic rations.

All the rations were balanced to the 33 per cent protein level (dry basis) with the respective supplementary protein sources. Now the rations had the same protein content on a dry weight basis but had different percentages of fat, ash, and dry matter due to the variation of the proximate composition of the supplementary protein sources (Table 5). Chicken Waste B had the highest

fat percentage. This basic ration then contained 21 per cent fat on a dry weight basis. All the other rations were balanced to this level by the addition of Devon Deep Fry Fat (a rendered animal fat used for deep frying). Theoretically, it would have been desirable to add horse fat to the horse rations, chicken fat to the chicken rations and so on but this proved to be impossible. Slight adjustments had to be made in the amounts of cereal and liver to retain the protein percentage at 33 and permit the fat percentage to be adjusted to 21. In a similar manner the ash content of the rations was adjusted by the addition of bone meal. Theoretically, the resulting basic rations then contained on a dry weight basis 33 per cent protein, 21 per cent fat and 6.8 per cent ash.

The constituent composition of the five basal rations as mixed is given in Table 6. Since the amount of feed required over the experimental period was large, three separate mixings were made. The quantities of ingredients shown in Table 6 are the amounts that could be conveniently mixed at one time in the feed mixer. The amount of water that was added to the rations to bring them to a hamburger-like consistency varied with the nature of the supplementary proteins and was therefore not measured. Since the rations were full fed this variation in moisture content could not lead to limiting feed consumption on rations containing higher levels of water.

Included in Table 6 are the analytical proximate composition of the first and third ration mixes and their averages. The differences between the computed and the analytical composition was somewhat disappointing and can best be attributed to the variability in the supplementary protein sources. The fat content of the whale meat and chicken waste rations, by actual analysis, are lower than the anticipated fat contents. It then follows that the analytical values for these protein sources as presented in Table 5 must have been higher than the actual average composition and as a result the mixed rations had a lower fat content. Westock (63) considered whale meat from different sources to have a fat percentage from 10-11 and a protein percentage from 77-83, when expressed on a dry weight basis. On the basis of the actual analyses the whale meat used for the rations contained 17 per cent fat which must have been higher than the actual average fat content. It is regretable that these discrepancies occurred but as previously mentioned these products are highly variable in composition. This fact makes it very difficult to obtain a representative sample for analysis which is typical of the supplementary protein sources.

The term supplementary protein source has been used to describe the animal proteins. The reason for this term becomes apparent when the total protein in the ration arising from the different sources is examined. Of the total protein in the ration 41.5 per cent was provided by the cereal mix, 14.5 per cent by

Table 6

Constituent Composition of the Five Basal Rations as Mixed

Constituent	Horse	Whale	Chicken A	Chicken B	Herring
Horse Meat	371 lbs.				
Whale Meat		238 lbs.			
Chicken A			352 lbs.		
Chicken B				376 lbs.	
Herring					325 lbs.
Liver	93	93	92	92	92
Bone Meal	13	14			8
Edible Fat	34	53	5		47
Cereal	261	261	257	259	260
Water	To bring ration to hamburger-like consistency				

Computed Proximate Composition: Dry Weight Basis

Protein %	32.9	33.2	32.9	33.0	33.2
Fat %	21.0	21.2	21.0	20.9	21.2
Ash %	6.7	6.8	6.7	6.7	6.8

Analytical Proximate Composition: Mix 1

Protein %	33.7	34.9	37.6	39.3	31.7
Fat %	23.0	19.2	17.0	13.2	24.1
Ash %	6.1	6.1	7.8	7.8	6.2
Dry Matter %	39.4	38.4	36.3	34.3	35.8

Analytical Proximate Composition: Mix 3

Protein %	36.0	36.8	36.9	35.9	33.6
Fat %	19.0	17.2	14.2	8.8	21.6
Ash %	6.3	6.6	8.0	8.0	7.3
Dry Matter %	38.5	43.0	42.3	41.0	43.7

Analytical Proximate Composition: Average Mix 1 and 3

Protein %	34.8	35.8	37.3	37.6	32.7
Fat %	21.0	18.2	15.0	11.0	22.8
Ash %	6.1	6.3	7.9	7.9	6.7
Dry Matter %	38.9	40.7	39.3	37.7	39.8

liver and the remaining 44 per cent by the animal protein source. On this basis, the animal proteins are considered to be supplementary since they provided only a fraction of the total protein. When the various animal proteins are compared for maintenance, reproduction and growth of the mink, they can only be compared on the basis of their supplementary value and cannot be compared directly as proteins. If the animal proteins sources were tested in a semi-purified diet where they were the only protein source, then a comparison could be made between proteins. In the present work the animal proteins will be compared on the basis of their supplementary value in the ration.

4. Description of the Experimental Rations

The formulation of the five basic rations has been described. From each basic ration four distinct rations were derived. Two of these were stored as frozen rations and the other two were canned. One of the frozen rations and one of the canned rations had a commercial antioxidant added. In total, there were 20 distinct rations derived from the five basic rations. For convenience each ration was given a descriptive code. All rations when described are referred by this code which is listed in Table 7. For example, ration HAF-1 is described as frozen horse meat ration which contains the antioxidant.

Table 7

Ration Code

HAF-1	WAF-5	CAAF-9	CBAF-13	FAF-17
HAC-2	WAC-6	CAAC-10	CBAC-14	FAC-18
HF-3	WF-7	CAF-11	CBF-15	FF-19
HC-4	WC-8	CAC-12	CBC-16	FC-20

H- Horse Meat
W- Whale Meat
CA- Chicken Waste A
CB- Chicken Waste B
F- Herring
A- Antioxidant
F- Frozen
C- Canned

5. Choice of the Antioxidant for the Experimental Rations

There are a variety of antioxidants available for the stabilization of fats in feeding stuffs. The choice of an antioxidant for a particular use is contingent upon a knowledge of the oxidative mechanism and the properties of the various antioxidants and their combinations.

Antioxidants are used extensively in a variety of products for the stabilization of fats from the deleterious effects of labile oxygen (6, 8, 28, 41). Fats, in the presence of labile oxygen undergo oxidation in two well defined phases (32). In the first, or induction phase, there is no apparent evidence of oxidation as determinable by organoleptic or other tests for rancidity (19, 30, 31, 40, 50). In the second phase, the velocity of the reaction increases rapidly in a logarithmic manner. Once this phase has set in, deterioration of the fat proceeds rapidly and rancidity can easily be detected by organoleptic means (32).

The length of the induction phase depends upon the amount of natural occurring antioxidants initially present. It has been shown with pure fatty acids that there is no induction phase. (32) When antioxidants are added to a substance they lengthen the initial induction phase to such an extent that there is no appreciable oxidative rancidity for an extended period.

The mechanism of oxidative rancidity has been reviewed frequently (6, 8, 22, 27, 30, 32). It has been established that the oxidative process is a free radical reaction. Antioxidants are able to curtail the formation of free radicals and in so doing become oxidized themselves (41).

Antioxidants are of two general types, aromatic phenols and their derivatives or aromatic amines and their derivatives. Both types are used extensively for the stabilization of fats. Of the two general types, the aromatic phenols are more water soluable and are more applicable to a fat-water system.

Most antioxidants are used for stabilizing fats which may later be added to other products. Very few antioxidants have been used in fat-water systems (8, 32). Evaluating antioxidants in such products as meats, fish and cereals is complicated because of the presence of other components such as water and salts which may influence the action of the antioxidant. To date the results of using antioxidants in meats, fish and cereals have been disappointing though some progress has been made in some cases (8).

In artifical fat-water systems, citric acid is an excellent synergist for such primary phenolic antioxidants as propyl gallate and butylated hydroxy-anisole (8). Most metals and metal ions are powerful pro-oxidants and synergists owe their activity to their metal scavenging powers. They slow the oxidative process by forming chelates with metal ions and thereby, greatly reducing their concentration.

The antioxidant chosen for the experimental rations was Tenox VII, manufactured by the Eastman Kodak Company. The composition of Tenox VII is given in Table 8.

Table 8

Composition of Tenox VII

Constituent	Per Cent
Butylated Hydroxy-Anisole	28
Propyl Gallate	12
Citric Acid	6
Mixed Glycerides	20
Propylene Glycol	34

Butylated hydroxy-anisole and propyl gallate are the primary antioxidants and citric acid is the synergist. Citric acid, in the presence of these two antioxidants is a powerful acid synergist and has been used for a variety of products (28, 30). The fact that citric acid is water soluble and propyl gallate is slightly water soluble aids in the dispersion of the antioxidant to the fat-water interfaces. Since mink rations might be regarded as a complicated fat-water system, it was felt

that with the knowledge at hand, Tenox VII might serve as an adequate antioxidant. The fact that it has been approved for human and animal use suggested that it would be a safe antioxidant.

The manufacturers of Tenox VII recommend its use at 0.05 per cent of the fat content of the ration. This recommendation is for fats added to a dry ration and it was felt that the level should be increased to 0.1 per cent of the fat content of the ration since it might be difficult to obtain a uniform dispersion of the antioxidant in the wet rations.

6. Mixing, Canning and Storage of the Rations

The rations were mixed at the Pacific Fur Breeders Cooperative at New Westminster, British Columbia. Eighteen hours prior to mixing, the frozen ingredients were allowed to thaw. At mixing time they were still ice cold but workable. The rations were blended in a large mixing machine normally used for mixing commercial mink rations. The cereal mix and bonemeal were placed first into the mixer. With the mixer in motion, melted fat was slowly poured over the mixing cereal. The meat ingredients (supplementary protein source and liver) were passed through a grinder situated on top of the mixing machine. Water was added at the same time as the meat ingredients until the ration had a hamburger-like consistency. The ingredients listed for one basic ration (Table 6) were sufficient for one mix. The antioxidant had been previously incorporated into one half of the total cereal mix. After mixing, the rations were sacked in 50 pound paper bags and immediately frozen in the quick freeze. They were then stored at -10°F.

Because of the large quantities required the rations were mixed at three different times. The first mix was on Dec. 31, 1955: the second mix was on June 4, 1956 and the final mix was on Sept. 21, 1956. Each ration mix required about 40 man hours.

One half of the frozen rations (Dec. 31 mix) were transferred to Imperial Cannery at Steveston, B.C. where they were canned. The rations were allowed to thaw for 24 hours, remixed and packed by hand into 20 ounce cans. The cans were sealed mechanically and retorted for 2½ hours at 240°F. as recommended by the technical supervisor of the American Can Co.

The remainder of the frozen rations were transferred to the University where they were stored at 0°F. The canned rations were stored at the prevailing temperature of the mink sheds.

III. Experimental Design

It was intended that each of the twenty experimental groups would consist of ten adult female and two male mink. The ten females were to consist of five standards, three pastels and two silveblus with males of corresponding colour phases. In this way it was hoped to achieve a distribution of the colour phases which were typical of the industry. For reasons beyond the control of this laboratory it was not possible to obtain sufficient animals to bring all the experimental groups to the number desired.

IV. Management

All the mink were vaccinated aginst distemper prior to the experiment. Each female received one-half ml. and each male one ml. of a live virus vaccine.

All the animals were housed in the University's experimental mink unit. It can be considered to by typical of ranch conditions.

The frozen rations were set out to thaw 24 hours prior to feeding. If the rations were too dry, water was added just prior to feeding. The canned rations were fed from a specially constructed nest box as designed by Mr. C. Harvey, senior animal technician. This permitted a 20 ounce can of feed to be before the mink at all times. In general, the females ate a can of feed every three days and the males every two days. No difficulty was experienced in feeding the mink from the cans and for the most part there was very little waste.

Animals were weighed at approximately monthly intervals on a scale that was marked in 20 gm. divisions. The only departure from this weighing schedule occurred during the gestation and pre-weaning period.

V. Experimental

1. Maintenance

The maintenance requirements of an animal encompasses its need for energy and protein for respiration, circulation of the blood, digestion and other metabolic processes, for the maintenance of body temperature and the repair of worn out tissues without causing an increase or decrease in weight of the animal (2). Nutrients in excess of the maintenance requirements are used for productive purposes, such as, gaining weight in the young, development of a foetus or the production of milk and eggs.

A ration may then be said to satisfy the maintenance requirements if it maintains the animals weight during a period when the animal is not engaged in any productive process. In the mink this period covers the winter months. Weight gained or lost was used as the criterion for measuring the maintenance performance of the various rations.

The maintenance period extended from Jan. 20 to March 21, 1956 since breeding began on March 15. The initial weights were recorded on Jan. 7, 1956 and the experimental feeding commenced on Jan. 20, 1956. The average weights and the net gain or loss for the adult female mink during the maintenance period are given in Appendix A for all the rations, except those containing herring. These will be treated in a later section.

The animals on all the rations, with one exception (CBAC-14), showed a slight weight gain. In order to establish

differences between the various treatments, the data were subjected to an analysis of variance (16). The average weight gained or lost for each ration group was used as the variate. The results of the analysis for the maintenance period is given in Table 9.

Table 9

Analysis of Variance for the Maintenance Period

Source of Error	Sums of Squares	D.F.	M.S.	F.	Tabled F p = .05
Canning (C)	784	1	784	.91	10.13
Antioxidant (A)	324	1	324	.38	10.13
Proteins (P)	2879	3	960	1.11	9.28
Interaction (C.A.)	841	1	841	.98	10.13
Interaction (C.P.)	1413	3	471	.55	9.28
Interaction (A.P.)	4788	3	160	.19	9.28
Error	2577	3	859		
Total	13606	15			

Canning refers to the canned rations and the frozen rations (no level of canning). Antioxidant refers to the rations with antioxidant and those with no level of antioxidant. Proteins refer to the rations containing the five supplementary protein sources.

From the analysis of variance it is apparent that all the calculated F values are less than their corresponding tabled values which indicates that there are no statistical differences between

any of the treatments for the adult female mink during the maintenance period. According to Brody (11) the maintenance requirements, in terms of digestible nutrients are approximately twice the basal energy requirements, whereas the requirement for productive purposes are approximately five times the basal requirement. Since the rations were designed to maintain a satisfactory growth rate, it is therefore not surprising to find no differences during the maintenance phase of the experiment.

There was no statistical difference between the frozen and the canned rations. However, it was noted that the mink receiving the frozen rations were slowly reducing their feed consumption near the end of the first ration mix (middle of May). Feed consumption increased when the mink received the freshly mixed second lot of feed (June 5). This apparent decline in feed consumption of a ration stored for several months at a low temperature may reflect the destruction of some nutrient (s). For example, it is shown in a later section that thiamine is destroyed to a certain extent upon prolonged storage at 0°F.

The herring rations, as mixed, were unsatisfactory as a maintenance diet for the adult female mink. Feed consumption began to decline after two weeks and by four weeks it was practically nil. The weight losses of the herring groups are given in Table 24, Appendix B. The average weight loss of all the herring rations was 115 gm. per female. The weight loss on the canned rations was greater than that on the frozen rations. Feed consumption was immediately restored when the mink were supplemented

with a 1 per cent aqueous solution of a complete vitamin mix (Nutritional Biochemical Corporation Vitamin Mix). In order to determine the lacking vitamin, the mink from the herring groups were reallocated into four groups and received the following vitamin supplements in their drinking water from March 2 to March 15, 1956:

Group 1 - Thiamin: 2 mg. per day.
Group 2 - Folic Acid: 0.18 mg. per day.
Group 3 - Thiamine and Folic Acid: levels of Gp. 1 and 2.
Group 4 - One per cent aqueous solution of N.B.C. vitamin mix.

Each mink was given 200 cc. of each solution per day. Two hundred cc. of a 1 per cent aqueous N.B.C. vitamin mix solution contained 2 mg. of thiamine and 0.18 mg. of folic acid.

Weight data and feed consumption prior to and after vitamin supplementation are given in Table 25, Appendix B. Supplementation with thiamine and thiamine plus folic acid markedly increased the feed consumption and the weight gained whereas treatment with folic acid caused a decrease in weight, though feed consumption was increased. This latter increase in feed consumption may have been due to a carry-over effect from supplementation by the complete vitamin mix. The mink receiving the complete vitamin mix at first lost some weight but then began to gain. Their feed consumption was also markedly increased. From the above, it was concluded that thiamine was the limiting vitamin and accordingly all the mink on the four herring rations received daily, in their drinking water 2 mg. of thiamine hydrochloride for

the remainder of the experiment. As evidenced in Table 26, Appendix B, the mink on the herring rations, with thiamine supplementation, soon increased their weights above their initial weight. It must be remembered that the last weight shown may be complicated by pregnancy.

The failure of the herring rations without thiamine supplementation may be attributed to the enzyme, thiaminase, which has been shown to be present in numerous species of fish (60). Thiaminase is more frequently found in the fresh water fish than in salt water fish. (66) Thiaminase has been demonstrated to be present in Atlantic herring (53) and Pacific herring (64) but not in Baltic herring (39). The failure to demonstrate thiaminase in Baltic herring may be due in part to the crude method used for determining thiamine.

Cats fed a diet containing salt water herring develop typical thiamine deficiences (54). Chastek Paralysis (a disease in mink and foxes resulting from the consumption of fish containing thiaminase) has been produced in mink fed a diet containing 19 lbs. Pacific Coast herring and 16 lbs. of wheat (65). It has also been produced in mink fed a diet containing 50 per cent of Great Lake's herring (29). One investigator (26) could only demonstrate a 20 per cent loss of thiamine upon refrigerated storage of Great Lake's herring during a five week period. In this case the herring had been filleted prior to storage and since thiaminase is generally absent from muscle but concentrated in the viscereal organs (60), the major enzyme containing material

would have been removed. Thus, the destruction of thiamine would be greatly reduced in filleted herring as compared to ground herring where thiaminase is evenly distributed.

Thiamine added to the herring rations was rapidly destroyed. Analysis of the herring rations for thiamine four days after mixing showed that the vitamin had been completely destroyed. No thiamine was present in any of the canned rations (Table 10). When 94.5 mcg. of thiamine hydrochloride, in the form of fortified rice pellets ,were mixed with 10 gm. of ration FAF-17 and allowed to incubate for two hours at room temperature, 78.8 per cent of the thiamine was destroyed.

It has been shown that daily supplementation of the drinking water with 2 mg. of thiamine hydrochloride restored feed consumption and growth in the mink receiving the herring rations. The destruction of thiamine takes place in the feed and not in the gut for it has been shown that thiaminase is destroyed by peptic digestion (55). The destruction of thiamine by thiaminase is an enzymatic hydrolysis yielding 4-methyl-5-hydroxy methyl ethylthiazole and 2 methyl-4-amino-5-hydroxy methyl pyrimidine (66).

2. Adequacy of the Thiamine Level in a Canned Horsemeat Ration

The results of the maintenance experiment indicate that adult, female mink fed canned rations will maintain their body weight. Table 10 compares the average thiamine contents of the frozen and canned rations. It is evident that the thiamine levels in the canned rations are much lower than the frozen rations.

Table 10

Average Thiamine Content of the Frozen and Canned Basic Rations

Wet Basis

Basic Ration	Canned mcg./gm.	Frozen mcg./gm.
Horse Meat	0.17	1.05
Whale Meat	0.25	1.09
Chicken A & B	0.13	1.10
Herring	0.00	0.00

They are below those suggested by the National Research Council (49) which is 0.34 mcg. per gm. of wet ration (dry matter is 34 per cent).

The following experiment was designed to establish if the level of thiamine in a canned ration (HAC-4) was sufficient to maintain large adult mink. A canned herring ration (FC-20) was fed to confirm the results of the maintenance experiment and to investigate the possible thiamine reserves of adult male mink.

Eight mature adult male mink (4 Standards and 4 Mutants) were fed a canned horse meat ration (HAC-4) and another similar group were fed a canned herring ration (FC-20) for a period of 11 weeks from May 8 to July 24, 1956. Weekly weights and feed consumption were recorded. Each mink receiving the herring ration was allowed to lose 200-300 gm. before it received a daily supplement of 2 mg. of thiamine hydrochloride in the drinking water.

A graphic record of the average weight and feed consumption for the two groups are recorded in Fig. 1 and 2, Appendix C.

The mink receiving the herring rations suffered slight weight losses during the first two weeks but a precipitous weight loss after the third week. The weight loss was also paralleled by a decrease in feed consumption. Shortly after supplementation with thiamine the mink began to increase their weight and feed consumption. Since the weight losses were only slight during the first two weeks it would appear that the mink had some thiamine reserve. The sharp decline in weight after the third week on a thiamine deficient diet would indicate that the reserves had been depleted.

In general, the group fed the horse meat ration had a slight weight loss over the experimental period. As expected, feed consumption paralleled an increase or decrease of body weight. It would appear that the mink receiving the canned horse meat were at their maintenance requirement or slightly below. Since feed consumption had been recorded the daily thiamine intake was calculated. The average daily feed consumption per mink was 158 gm. of wet feed or 60 gm. of dry feed. The average body weight was 1390 gm. and thus the daily thiamine intake per Kilo body weight was .019 mg. The National Research Council suggested daily requirement for thiamine (49) is .001 mg. per gram of dry matter. Thus, the suggested requirement per Kilo body weight is .043 mg. This suggested daily requirement is approximately twice the amount the mink received from the canned horse meat ration. This would suggest that the National Research Council (49) recommended levels

are too high. The present results suggest that a thiamine level of approximately .02 mg. per kilo body weight or .005 mg. per pound of feed as fed can supply the maintenance requirements for adult male mink. It is realized that further work is required to validate this suggestion. Loeschke (37) is currently working on this phase of mink nutrition.

3. Reproduction

Each experimental group was treated as a breeding group of ten females and two males. An attempt was made to stay within this group for all matings. Towards the end of the breeding season some males from other experimental groups had to be used where a within group male failed to handle his females satisfactorily. The breeding period extended from March 15 to April 8, 1956 with the majority of the females being bred between March 15 and March 25.

Though the first kits were born on April 22 the majority were born during the first two weeks of May. It was intended to wean the male kits at 400 gm. and the female kits at 300 gm. At weaning, the kits were approximately 7 weeks old.

The reproductive and weaning data for the experimental groups are given in Appendix D. Kits born per female were used as the variate in the analysis of variance for the reproductive phase. The results of the analysis are presented in Table 11.

Table 11

Analysis of Variance for Reproduction

Source of Error	Sums of Squares	D.F.	M.S.	F.	Tabled F p = .05
Canning (C)	21.7	1	21.7	20.7	7.71
Antioxidant (A)	0.4	1	0.4	0.4	7.71
Proteins (P)	3.8	4	0.95	0.9	6.39
C.A.	1.6	1	1.6	1.5	7.71
C.P.	3.8	4	0.95	0.9	6.39
A.P	3.3	4	0.8	0.8	6.39
Error	4.2	4	1.05		
Total	38.8	19			

From these results it can be concluded that there were no significant differences between treatments except for Canning (C). The reproductive performance of the mink receiving the canned rations was inferior to that of the mink receiving the frozen rations at p = .05. From the mink on the canned rations, 12 kits were born from 79 females bred, whereas, from the mink on the frozen rations 194 kits were born from 88 females bred.

The failure of the canned rations, can perhaps be attributed to the destruction of some vitamin (s) during the canning process. Table 12 presents the Vitamin A levels of the canned rations in I.U. per 100 gm. of ration.

Table 12

Vitamin A levels in the Canned Rations

Rations with Antioxidant			Rations without Antioxidant		
HAC-2	930	I.U./100 gm.	HAC-4	480	I.U./100 gm.
WAC-6	2860		WC-8	540	
CAAC-10	645		CAC-12	785	
CBAC-14	210		CBC-16	220	
FAC-18	2230		FC-20	1250	
Ave.	1375			655	

The maximum National Research Council requirements (49) of Vitamin A are 140 I.U. per day. Since a mink will eat about 150 gm. per day it is apparent that it will receive sufficient Vitamin A from all of the canned rations.

It has been shown that the thiamine level in the canned rations is low when compared to the frozen rations (Table 10) and that the thiamine levels in the canned rations are just barely sufficient to meet the maintenance requirement of adult male mink. It is therefore probable that the thiamine levels in the canned rations are not sufficient for reproduction and lactation. There is also the possibility of Vitamin E being destroyed during the canning process and since one of the primary functions of Vitamin E is to ensure normal reproduction, a reduced level would have a very marked effect.

Due to the complete failure in reproduction and due to difficulties in arranging for additional canning, the canned rations were discontinued for the remainder of the experiment.

The average number of kits produced per female bred from the frozen rations was 2.2. Enders (15) states that the United States National average in 1949 was 3.2 and in 1950 was 2.8 kits per female. An average of 3 kits per female was considered good. From the cited values the reproductive performance of the mink on the frozen rations was below average.

The rations fed to the experimental mink were frozen and stored, a practice which is contrary to normal ranch conditions where the rations are mixed daily. Long storage periods at freezing temperatures result in a gradual loss of nutrients through enzymatic and oxidative processes (8). It will be demonstrated in a later section that there is considerable loss of thiamine upon prolonged storage at refrigerated temperatures. It is therefore probable that the poor reproductive performance of the mink receiving the frozen rations may have been, in part, due to losses of essential nutrients by storage at refrigerated temperatures. It had been hoped that the antioxidant would have afforded some protection but there was no evidence to suggest that this was the case.

Other factors such as over aggressive males, sterile females, false pregnancies, disease, weak sperm, lack of ovulation and fat females will also reduce the average litter size. One

surveyor of ranch operations (12) found a breeding ratio of one male to 3.5 females as the most satisfactory ratio whereas one male to 5 females was considered borderline. Since the experimental groups had one male associated with 6 females this may have been another contributing factor to the lower than expected fecundity. The fact that these mink were handled as experimental animals may have influenced their reproductive performance. Since no selection pressure in the direction of increased fertility has been possible in the University Unit to this time, this may also have been a contributing factor.

The average gestation period for all the mink was 53.4 ± 4.0 days with a range of 46-64 days. The average gestation period as compiled by Enders (15) is approximately 51 days. Ranges from 37 to 91 days have been reported though the normal range is from 47 to 75 days. The gestation period of the mink on the experimental rations is considered normal.

4. Growth

Female mink, with associated males for breeding, have been maintained successfully on the various experimental rations. However, the reproductive performance of the mink receiving the canned rations was essentially a complete failure and the number of kits available from the mink fed the frozen rations was below average. As a result, the number of kits available for the growth phase was greatly reduced from what had been anticipated at the outset of the experiment. It had been hoped to have at least 10

male and 10 female kits for each experimental ration. Because of the reduced numbers available, 6 male and 6 female kits constituted an experimental group for the ten different frozen rations. As explained previously, the canned rations were not included in the growth phase of the experiment.

All the kits were weaned in the period from June 28 to July 6, 1956 with the majority being weaned between June 28 and July 3, 1956. They were fed the respective experimental rations until Nov. 11, 1956. It had been hoped to wean the male kits at 400 gm. and the female kits at 300 gm. but because of a delay in the construction of nest-boxes the average weaning weights were somewhat higher than planned.

Where possible, 6 male and 6 female kits were placed on the same ration their mothers had received but where this was not possible, because of insufficient numbers, they were chosen from a ration that had the same supplementary protein source.

The mink were weighed at approximately monthly intervals over the experimental period in order to record their progress. The average weights of the ration groups and the average weight gained from weaning to maturity for each group are presented in Table 27, Appendix E.

The average weight gained for each group between weaning and maturity was used as the variate in the analysis of variance of the growth phase. The results of this analysis are presented in Table 13.

Table 13

Analysis of Variance for the Growth Phase

Source of Error	Sums of Squares	D.F.	M.S.	F.	Tabled F. p = .05	Tabled F. p = .01
Sex (S)	1,581,469	1	1,581,469	705.4	7.7	21.2
Antioxidants (A)	24,221	1	24,221	10.8	7.7	21.2
Proteins (P)	200,561	4	50,140	22.4	6.4	16.0
S.A.	805	1	805	0.4	7.7	21.2
S.P.	65,713	4	16,428	7.3	6.4	16.0
A.P	25,479	4	6,369	2.8	6.4	16.0
Error	8,967	4	2,242			
Total	1,907,215	19				

The results of the analysis indicate several differences between treatments. There is a significant difference at p = .01 between the average gains of the males and the females which of course is expected. There is a significant difference at p = .05 but not at p = .01 between the rations containing the antioxidant and those that do not contain it. The inclusion of the antioxidant in the rations has depressed growth. There are significant differences between the supplementary protein sources at p = .01. The interaction between Sex and Proteins is significant at p = .05.

The differences within the groups composing the source of error in Table 13 are determined in the following manner. The analysis of variance has shown a significant difference between sex and antioxidants. Since there are only two sexes and two levels

of antioxidant the total gains may be directly compared and the one with the larger total gain is superior to the other. When this was done, it was established that the males gained more than the females (significant at p = .01) and that the antioxidant depressed growth (significant at p = .05).

The analysis of variance indicates that there are differences of gain associated with the various supplementary protein sources. The significance between the gains of the mink receiving the various supplementary proteins may be determined by calculating the minimum difference between any two variate means required for significance (M.S.D.). These calculations appear in Appendix F and from them the following conclusions may be drawn. There is no significant difference at p = .01 between the Horse Meat, Whale Meat and Chicken Waste A rations but these rations as a group are significantly different and superior to the Chicken Waste B and the Herring rations.

The analysis of variance for the growth phase also indicates a significant interaction between Sex and Proteins at p = .05. The minimum significance for a difference (M.S.D.) for this interaction is calculated in Appendix F. From these calculations Table 14 was prepared to facilitate the interpretation of the interaction between Sex and Proteins.

Table 14

Comparison of the Differences in Gain due to the Interaction Sex and Proteins

Significance at p = .05 with

Ration-Sex	Horse Meat	Whale Meat	Chicken Waste A	Chicken Waste B	Herring
Horse Meat-Females	none	less	none	none	none
Horse Meat-Males	none	greater	greater	greater	greater
Whale Meat-Females	greater	none	none	greater	greater
Whale Meat-Males	less	none	none	greater	greater
Chicken A-Females	none	none	none	none	greater
Chicken A-Males	less	none	none	greater	greater
Chicken B-Females	none	less	none	none	none
Chicken B-Males	less	less	less	none	none
Herring-Females	none	less	less	none	none
Herring-Males	less	less	less	none	none

In this table, the total gains of the males and females for each supplementary protein source are compared for significance at p = .05 to the males and females of the other supplementary protein sources. The following example explains the terminology used in Table 14. When the Chicken A-Males are compared to the males on the other supplementary protein sources, the table is read as follows. The total gain made by the males receiving the Chicken A rations was significantly less (less) than the total gain made by the males receiving the Horse Meat rations, significantly greater than the total gain of the males receiving the Chicken B and Herring rations but not significant from the gain (none) made by the males receiving the Whale rations.

In general, the interaction between sex and proteins indicates a parallelism between the supplementary protein sources but these differences are greater in the males than in the females. It would be anticipated that differences in the males would be greater than in the females since at any given time after birth the female is physiologically older than the male (11). In other words, under the conditions of the present experiment, females at weaning had completed 37.5 per cent of their final weight while the males had completed only 27 per cent.

The results of the analysis of variance for the growth phase has indicated significant differences between the gains of the groups receiving the different supplementary protein sources. In all probability the differences in the growth rates of the mink on the various experimental rations may be similar to the differences encountered in mink fed various ranch diets. These variations in growth rates would occur since the rations fed to ranch mink undoubtedly vary in nutritive value and as a result differences in growth rates would occur. If the average growth curves of each of the experimental rations are plotted, then the spread in weights at any given time may be an indication of the variation in weights that might be expected in ranch mink. These weight curves, one for the females, Fig. 3, Appendix E and one for the males, Fig. 4, Appendix E, were plotted from the data presented in Table 27. These curves show the magnitude in the spread of weight at any time after weaning. As the animals grow

older and the differences between rations becomes more apparent the spread in weights become progressively larger. In the case of the ranch mink it is difficult to segregate nutritional effects from inherent differences in the mature weight of the various strains. Such distinct differences are recognized in geographic strains of wild mink. Undoubtedly, man has increased the variation in size of ranch mink in the course of his selection pressures for colour phase and pelt quality. It is therefore felt that these curves represent the variation in weight that might be expected from ranch mink which are fed diets differing in nutritive value and arising from the various strains.

The rations containing Horse Meat, Whale Meat and Chicken Waste A are superior to those containing Chicken Waste B and Herring. Gunn (17) has compared Whale Meat and Horse Meat in diets for growing mink and found no difference in the total weight gained from weaning to maturity. The present experimental results also indicate no difference and suggest that Whale Meat may be substituted for Horse Meat on a dry weight basis.

The significant difference between the two Chicken Waste rations is difficult to explain. The two types of Chicken Wastes differ in one respect, Chicken Waste A contains the gall bladder and the anal portion whereas Chicken Waste B does not. The results of the Net Protein Utilization Values of the supplementary protein sources (Table 19) show that Chicken Waste A has a higher value than Chicken Waste B which indicates that the protein of the

latter is inferior to the former. In all probability the difference in the Net Protein Utilization Values of the Chicken Wastes arose from the inherent variability in the composition of this product. When compared to the other ration groups, the gains of the Herring group were less but it was certainly not due to protein of inferior quality. The results of the Net Protein Utilization experiment (Table 19) show that Herring has the highest value, even higher than casein. Robertson (52) has also obtained similar results on commercial herring meals. The poorer performance of the Herring rations might be due to an insufficient supply of thiamine though this seems rather unlikely since they were being supplemented daily or it might be due to the formation of rancidity in the ration since herring fat is easily oxidized.

The growth depressant effect of the antioxidant, Tenox VII, would seem to indicate that it possesses some degree of toxicity. The antioxidant had been added at double the recommended level for it was felt that the dispersion of the antioxidant in a wet ration might be poor. The level allowed for the addition of an antioxidant to fats containing a mixture of butylated-hydroxy-anisole, propyl gallate and citric acid (components of Tenox VII are: butylated hydroxy-anisole, .02%; propyl gallate, .01%; and citric acid, .005% (8). The level of Tenox VII added to the rations was .01% of the fat content. From the composition of Tenox VII (Table 8), the computed percentages of the component antioxidants added to the fat of the ration were butylated

hydroxy-anisole, .028%; propyl gallate, .012% and citric acid, .006%. All of these were added at levels slightly higher than recommended for use in stabilizing fats.

The above values refer to the antioxidants as percentages of the fat content of the ration and not of the total ration. A high fat diet will contain more antioxidant than a low fat diet if the percentage of antioxidant is based on the fat content. If an animal consumes a diet containing 20 per cent fat it will not receive twice as much antioxidant as if it were consuming a diet containing 10 per cent fat since the food consumption is less on the high fat diet than the low fat diet. Some caution then has to be used in evaluating data pertinent to the toxicity of antioxidants.

When a diet containing 10 per cent fat and 0.3 per cent butylated hydroxy-anisole was fed to rats there was a depression of growth (20). Growth was also depressed in rats by feeding a diet which contained 1.17 per cent propyl gallate, though other experiments showed that as much as 5 per cent was required (34). There were no deleterious effects on the performance of guinea pigs or dogs fed a diet containing 0.011 per cent propyl gallate (34).

It is apparent that the levels of butylated hydroxy-anisole and propyl gallate which cause a growth depression in the rat are higher than those causing a similar depression in the mink. The problem of toxicity of antioxidants for the mink requires further study in order to establish safe levels.

5. Fur Quality

No quantitative comparison of fur quality was made on the mink receiving the experimental rations. At present, the number of mink at the University is relatively small and in order to ensure sufficient numbers for future experiments none of the females and only 41 from a possible 56 males were pelted. In general, the retained mink were of a superior quality to those pelted since a selection pressure was exerted in favour of size and pelt quality. The pelts were scored by an arbitrary system where a value of 1 was a pelt of superior quality and a value of 5 was inferior. On this system, the average score of all the pelts was 2.2 which indicates a pelt quality slightly above the average. From the small number of experimental animals pelted, no quantitative comparison of fur quality could be made though there is an indication that fur quality could be considered as average.

6. The Destruction of Thiamine in the Experimental Rations

(a) Introduction

Studies concerned with the destruction of vitamins during the various processing and storage stages of a feed product are important in the final evaluation of its nutritional adequacy. In this regard, the destruction of thiamine is particularly important since one of the first signs of a thiamine deficiency is anorexia. When feed consumption is decreased by a limiting supply of thiamine, other vitamins and nutrients may then too become limiting.

Losses of vitamins in modern processing and storage are primarily due to enzymes, chemical oxidation and extraction (43). The most prominent changes in animal, fish and dairy products stored for prolonged periods at refrigerated temperatures are due to oxidative rancidity (8, 43).

Mapson (43), in a review, indicated that there was little change in the vitamins of the B groups when they were stored at refrigerated temperatures. However, Lehrer (36) reported a 40 per cent loss of thiamine in pork chops stored at 0°F. for 6 months whereas Lee (33) reported no loss of thiamine in pork chops stored at 0°F. for 6 months. Lehrer (35) also reported a 50 per cent loss of thiamine in lamb chops stored for 6 months at 0°F. Liver slices stored for 2 months at refrigerated temperatures lost 27 per cent of their thiamine. The apparent conflict of published results on the destruction of thiamine upon prolonged refrigerated storage may be explained in part by the method and type of wrapping used for the stored product. Lee (33) demonstrated no thiamine loss in pork chops wrapped in 300 MSAT cellophane and butchers paper. This type of wrapping is vapour proof and therefore does not allow any atmospheric oxygen to come in contact with the meat surface and presumably greatly reduces any oxidative rancidity. Other wrappings which are not vapour proof or which are used carelessly would allow access of oxygen to the meat surface and hence encourage oxidation.

Further evidence that thiamine is destroyed by oxidation has been demonstrated with distilled water solutions of thiamine which were stored in sealed ampoules for a year. Thiamine loss was 59 per cent but no loss occurred when the ampoules contained an atmosphere of nitrogen (13). Addition of antioxidants to the thiamine solutions decreased the thiamine loss and it was concluded that oxygen was the destructive factor.

It has also been demonstrated that there is a marked instability of thiamine in certain purified diets (62). An unsaturated oil, such as linseed oil, was more deleterious than a hydrogenated vegetable oil and the addition of ascorbic acid or hydroquinnone, serving as antioxidants, increased the stability of thiamine (22). This again is evidence for an oxidative destruction of thiamine. However, in certain types of salt mixtures it has been domonstrated that dipotassium phosphate is chiefly responsible for the destruction of thiamine because it is very hygroscopic and upon hydrolysis is alkaline, providing conditions in which thiamine is very unstable (62).

Two separate studies were made on the destruction of thiamine in the experimental frozen rations in order to establish if the loss of thiamine prior to eating would be great enough to make thiamine a limiting factor in the rations. In one study, the destruction of thiamine was measured over a four month storage period at 0^{o}F. while in the other, the destruction of thiamine was measured from the time the frozen rations were beginning to thaw

until they were consumed by the mink. These studies were also designed to show the quantitative loss of thiamine upon storage and the effect of the antioxidant upon thiamine destruction.

(b) The Effect of Prolonged Storage at 0°F. on the Thiamine Content of the Experimental Frozen Rations.

Duplicate thiamine determinations were made on samples from each of the experimental frozen rations at 0, 33, 77, and 110 days of storage at 0°F. during the period from June 6 to Sept. 24, 1956.

The thiochrome procedure as recommended by the Association of Vitamin Chemists, Inc. was used for the determination of thiamine (14). The modifications of the procedure and the sampling methods are presented in Appendix G.

Duplicate results, expressed as mcg. of thiamine per gram of dry ration, are presented in Table 15. The average per cent destruction of thiamine for all the rations over the 110 day period of storage was 29.2. The average per cent destruction of thiamine in the rations containing the antioxidant was 30.2 whereas the average per cent destruction of thiamine in the rations containing no antioxidant was 28.2. There is then no evidence to suggest that the antioxidant exerted any protective effect upon the destruction of thiamine. The apparent failure of the antioxidant to afford protection may be due to the fact that citric acid is the only water soluble component of the antioxidant mixture. Since the concentration of citric acid in Tenox VII is only

6 per cent, the resulting concentration of it in the water phase would be very low and for this reason may have been insufficient to permit the destruction of thiamine, especially if there were some interference by the food components.

The Herring rations contained no thiamine since it probably was destroyed during mixing by the enzyme, thiaminase. At the time of the first analysis, the rations were only seven days old.

(c) The Destruction of Thiamine Between Thawing and Feeding.

All the experimental rations fed to the mink at this time were frozen and required thawing prior to feeding. The thawing process, at room temperature, took about 24 hours after which the rations were remixed by hand to ensure uniformity. If the mink were given an excess of feed, the rations could conceivably remain on the wire 24 hours after thawing. Thus, the maximum time the rations had to deteriorate between thawing and consumption was 48 hours. In order to determine the thiamine loss between thawing and eating, samples from each ration were analysed for thiamine at 24 hour intervals for a three day period. The herring rations were excluded from the study since they contained no thiamine. Table 16 shows the destruction of thiamine in the experimental rations between thawing and eating at 24 hour intervals. The per cent thiamine destruction for each ration is also given.

Table 15

Thiamine Levels[1] of the Experimental Rations During Refrigerated Storage

Rations	June 6	July 9	Aug. 22	Sept. 24	Per Cent Destruction
HAF-1	4.7	4.4	3.1	2.9	38.3
	4.7	4.5	3.3	2.9	
HF-3	5.9	4.6	3.5	3.5	41.6
	5.9	4.6	3.5	3.4	
WAF-5	3.9	3.1	4.3	3.6	7.7
	3.9	3.0	4.3	3.6	
WF-7	5.7	4.8	4.4	4.1	28.7
	5.8	4.8	4.4	4.1	
CAAF-9	6.8	5.2	5.3	3.9	42.6
	6.7	5.2	5.3	3.8	
CAF-11	5.6	5.8	6.2	4.9	13.3
	5.7	6.1	6.2	4.9	
CBAF-13	5.5	5.2	4.8	3.6	32.4
	5.3	5.2	4.8	3.7	
CBF-15	5.3	4.7	4.3	3.8	29.4
	5.6	5.1	4.4	3.9	
FAF-17	0.0	0.0	0.0	0.0	0.0
	0.0				
FF-19	0.0	0.0	0.0	0.0	0.0
	0.0				

1 Expressed as mcg. thiamine per gm. of dry ration.

Table 16

The Destruction of Thiamine Between Thawing and Feeding

Ration	0 hrs.	24 hrs.	48 hrs.	Per Cent Destruction
HAF-1	2 4.8	4.2	4.0	16.7
HF-3	4.8	4.5	4.2	11.1
WAF-5	4.2	4.0	3.7	12.5
WF-7	5.3	5.0	4.2	20.0
CAAF-9	4.8	4.2	3.5	27.8
CAF-11	5.6	5.3	5.0	9.8
CBAF-13	4.0	3.5	3.2	20.0
CBF-15	5.0	5.0	4.5	10.5

2 Micrograms thiamine per gm. of dry ration.

The average per cent destruction of thiamine for all the rations was 16.0; the average per cent destruction of thiamine in the rations containing the antioxidant was 19.3 and the average per cent destruction of thiamine in the rations containing no antioxidant was 12.8. There was no evidence of any protection of thiamine by the antioxidant. Though the rations containing the antioxidant had a larger thiamine destruction than the rations containing no antioxidant, the difference was not significant.

The average thiamine loss for all the rations stored at 0°F. for a four month period was 28.8 per cent and the average thiamine loss for all the rations between thawing and consumption

was 16.0 per cent. Accordingly, the average loss, calculated from the initial thiamine content, due to a maximum storage period and the period between thawing and consumption would be 40.2 per cent of the initial level.

The lowest initial thiamine level was 3.9 mcg. per gram of dry ration (Table 15). The maximum average thiamine destruction was 40.2 per cent and presumably 2.3 mcg. of thiamine per gram of dry ration would be left for the mink. If an average feed consumption of 60 grams of dry matter per day is assumed for adult females, then they would consume at a minimum, 0.14 mg. of thiamine per day. The upper level of thiamine recommended by the National Research Council (49) is 0.1 mg. per day. This, then suggests that the thiamine levels in the rations even after four months of refrigerated storage are adequate for the mink. It is therefore suggested that thiamine should not have become the limiting nutrient factor in the frozen rations.

The results of the studies on the destruction of thiamine in the experimental rations indicate that the antioxidant had no beneficial effect in preventing the destruction of thiamine. Investigations (13, 22) have shown that the destruction of thiamine can be oxidative and that the amount of thiamine destroyed can be decreased by the exclusion of oxygen or the addition of an antioxidant. The application of antioxidants to fresh meats, fish and poultry has not generally proved satisfactory (8) and there are cases of success (28, 41) and failure (5, 18).

Perhaps the ineffectiveness of the antioxidant in preventing the destruction of thiamine may be due to the difficulty of dispersing the antioxidant or to the low water solubility of the components of Tenox VII in the ration mixture. The influence of food components may have rendered the antioxidant ineffective (8).

7. Digestibility Studies on the Experimental Rations

The term per cent digestibility as applied to a feedstuff refers to that percentage of the feedstuff which is absorbed from the small intestine (44). The usual method is to measure the total feed consumed and faeces voided over a definite time interval. The ratio of the feed absorbed (total feed consumed less the faecal loss) to the feed consumed, expressed as a percentage is taken to be the per cent digestibility. This term should be distinguished from true digestibility which takes into consideration the metabolic portion of the faecal output, largely arising from gastrointestinal secretions, cells of the intestinal mucosa and intestinal microflora (1).

The collection method for determining the per cent digestibility of a nutrient involves the measurement of the total intake of the nutrient and the total faecal output over a predetermined time interval. This method is labourious and in most instances requires special apparatus. A simpler and less time consuming method based on the use of certain exogenous or indogenous indicators has been proposed (44). The indicator may be a simple chemical compound such as chromic oxide which is

indigestible and non-absorbable. In addition, it must be non-toxic to the animal and pass through the digestive tract at a uniform rate. The indicator is mixed uniformly with the feed at a low level and fed to the animal. After a certain time interval the feed and faeces are analysed for the indicator. From the concentration of the indicator in the feed and faeces the per cent digestibility may be calculated. The digestibility of a specific nutrient, such as nitrogen, may also be determined from the ratio of the concentration of the indicator to that of nitrogen in the feed and the same ratio in the faeces resulting from the feed (44).

There have been few published studies concerned with the digestibilities of mink rations and specific nutrients in these rations. It will be shown that the feed consumption of the mink receiving the different rations varied markedly (Table 20). For example, the mink receiving the Chicken Waste rations ate more than the mink receiving the Horse or Whale Meat rations. These differences in feed consumption may possibly arise from differences in digestibility. This study was then initiated to establish the digestibility coefficients of the various rations in order to attempt to explain the differences in feed consumption.

Accordingly, the following digestibility studies were initiated. Total digestibility, nitrogen digestibility and fat digestibility were determined for each of the 10 experimental frozen rations, using chromic oxide as the indicator. In addition

"carbohydrate" digestibility was calculated by assuming that the mineral content of the ration was non-digestible. The details of the methods and the individual calculations for the fifty adult female mink are given in Appendix H. The average total digestibility, nitrogen digestibility, fat digestibility and "carbohydrate" digestibility with their standard deviations are presented in Table 17.

Table 17

Per Cent Total, Nitrogen, Fat and "Carbohydrate" Digestibilities of the Frozen Experimental Rations

Ration No.	Total Digestibility	Nitrogen Digestibility	Fat Digestibility	"Carbohydrate" Digestibility
HAF-1	64.5 ± 5.9	73.8 ± 6.6	91.5 ± 1.8	27.0 ± 4.7
HF-3	71.4 ± 6.4	77.7 ± 8.0	94.5 ± 1.1	27.3 ± 4.4
WAF-5	63.5 ± 1.7	75.0 ± 6.8	87.2 ± 2.3	27.5 ± 3.3
WF-7	60.0 ± 4.9	71.2 ± 8.3	83.4 ± 1.8	27.5 ± 3.5
CAAF-9	61.0 ± 6.3	69.3 ± 8.9	84.3 ± 2.6	30.3 ± 4.8
CAF-11	60.9 ± 5.1	68.8 ± 2.8	87.9 ± 1.8	30.6 ± 4.3
CBAF-13	62.4 ± 2.7	68.1 ± 5.3	84.0 ± 1.9	31.6 ± 1.8
CBF-15	58.2 ± 3.3	69.6 ± 5.3	87.8 ± 1.7	22.3 ± 2.0
FAF-17	52.2 ± 5.3	60.0 ± 9.7	86.4 ± 1.7	22.5 ± 4.7
FF-19	60.6 ± 9.7	72.0 ± 7.5	91.5 ± 1.8	22.7 ± 5.5
Average	61.5 ± 5.1	70.6 ± 6.9	87.9 ± 1.9	26.9 ± 4.0

The "t" test (16) was used to test the significance between the rations at p = .05. When the rations were compared with respect to total digestibility, there was a significant difference between ration HF-3 and FAF-17 but the differences between the other rations were not significant. There were no significant differences between the rations when the nitrogen digestibilities were compared. However, when the fat digestibilities were compared the following rations were significantly different with one another at p = .05. HAF-1 with WF-7 and CBAF-13; HF-3 with WAF-5, WF-7, CAAF-9, CAF-11, CBAF-13, CBF-15 and FAF-17; FF-19 with WF-7, CAAF-9, CBAF-13 and FAF-17. Only CBAF-13 was significantly different from CBF-15 when the "carbohydrate" digestibilities were compared.

In general, total digestibility, nitrogen digestibility and "carbohydrate" digestibility were similar for all the rations except for the few significant differences. The differences in fat digestibility between the rations was greater and in general both horse meat rations and the one herring ration (FAF-19) had higher fat digestibilities than the others. It would be erroneous to conclude that horse fat is more easily digested than the other fats since the fat in any one ration is a composite mixture of the fat derived from the supplementary protein source, the cereal mix and any added fat. Table 18 relates the per cent of the fat derived from the ration components. It is apparent from the high digestibility and the variety of the sources of fat that mink, in

general, have the ability to utilize fats with great facility and by so doing, the fat portion of the diet can provide the major portion of the energy required for maintenance.

Table 18

Per Cent of the Fat Derived from the Ration Components

	Rations				
Component	Horse	Whale	Chicken A	Chicken B	Herring
Horse Meat	36.5				
Whale Meat		.7			
Chicken A			59.0		
Chicken B				55.0	
Herring					27.1
Liver	7.2	8.3	9.9	13.5	6.5
Added Fat[3]	39.8	71.9	8.2		51.1
Cereal	16.5	19.1	22.9	31.5	15.3
	100.0%	100.0%	100.0%	100.0%	100.0%

3 Devon Deep Fry Fat

There are many factors influencing digestibility coefficients which make comparisons difficult between investigations (44). Crude fibre tends to lower the digestibility of all nutrients. A short time of passage of feed tends to lower the apparent digestibility of all nutrients. In this respect, the time of passage of the mink is extremely short and ranges from

1½ to 2½ hours (42) and consequently digestibility coefficients would tend to be reduced.

Worthen (65) fed a ration to mink which contained 19 parts of herring to 16 parts of wheat and found the average total digestibility to be 52.7 per cent and the average nitrogen digestibility to be 83.7 per cent. Madramootoo (42) used two different cereal rations and found the total digestibility to be 52.1 and 60.2 per cent. The average total digestibility of the present experimental rations was 61.5 per cent which was higher than the values cited by the above workers. This is probably due to the fact that their rations contained very high levels of cereal and consequently were less digestible.

Bernard (7) found that the protein digestibility ranged from 83.6 to 90.6 per cent and the fat digestibility varied from 84.0 to 97.9 per cent when mink were fed diets containing 75 per cent horse meat. McCay (45) cites that the protein digested by foxes on various diets ranged from 71 to 91 per cent. In dogs, about 80 per cent of the protein is digested in a mixed feed. In a series of 19 digestibility trials, total digestibility varied from 53 to 84 per cent; protein digestibility varied from 51 to 80 per cent and fat digestibility varied from 66 to 94 per cent.

It is evident from the digestibility coefficients cited for the mink, fox and the dog that there is great variability in these coefficients within the same species. These differences are mainly due to variability in ration composition.

When the average digestibilities of the experimental rations, as presented in Table 17, are compared to those cited for the mink, dog and the fox, the agreement is good except for the nitrogen digestibility which seems to be somewhat lower. Perhaps the difficulty in utilizing plant proteins would lead to this relatively low nitrogen digestibility.

The low "carbohydrate" digestibility suggests that the mink has difficulty in utilizing "carbohydrates" and in particular, the cereal portion of the ration. Dry diets for mink which contain high percentages of cereal have not proved successful for raising commercial mink. The main difficulty, according to Kifer and his associates (83, 86) is that such rations are relatively indigestible. Alberta ranchers (63) claim that cooking rations, which contain a high percentage of cereal, permits adequate growth of commercial mink. They claim that cooking the cereal makes it more digestible. However, Bernard (7), in a study of the digestibility of starch from various cereals fed to mink, found that cooking increases the utilization of cornstarch and corn but did not increase the digestibility of starch from wheat and oats. It is apparent that the problem of the utilization of cereals by mink requires further careful study. Because of the relatively low digestibility of cereals, an attempt is being made in this laboratory, to increase their utilization by the use of certain carbohydrases.

8. The Net Protein Utilization of the Supplementary Protein Sources

The primary function of dietary protein is to furnish a mixture of amino acids of the proper pattern for the synthesis of tissue proteins (1). The theoretical minimum requirement of protein is the amount actually stored by the body plus the endogenous losses. The actual dietary requirement must be much higher to provide for digestive losses and the biological value of the protein. Digestibility coefficients take into account the losses due to incomplete digestion of the dietary protein. The biological value of a protein assesses the efficiency with which the absorbed amino acids are utilized in forming body protein. It represents the percentage of absorbed protein that is stored by the body. The term net protein utilization or sometimes referred to as net protein value embodies both the digestibility and biological value of a dietary protein in that it represents the percentage of the ingested protein that is stored in the body. The net protein value (N.P.U.) is then the product of the biological value (B.V.) and the digestibility (D.) of the protein.

Biological value measurements on proteins have received considerable attention because the digestibility of a protein for a species on a particular ration tends to be relatively constant for each class of proteins and insofar as is known the digestibility of a protein is not related to its amino acid composition (56). The biologic value is a direct measure of the amino acid composition.

There are numerous methods for measuring biological value and these have been reviewed recently (1, 56) but the basis of comparison for all methods is the well established Thomas-Mitchell procedure (44). This procedure is tedious and time consuming in that it requires numerous nitrogen determinations before a value is obtained.

Recently Miller and Bender (47) have proposed a much shortened method for measuring net protein utilization. Their method compares favourably to the Thomas-Mitchell procedure. The advantage of this method over the classic one lies in the small number of measurements made over a short experimental period. For example, the evaluation of 3 proteins by the Thomas-Mitchell method with 12 rats involves 120 nitrogen estimations of the urine and faeces as well as 3 food analyses and 60 measurements of food consumption over a period of 6 weeks. The Miller and Bender (47) method can evaluate 7 proteins, using 32 rats, 8 feed consumption measurements in a period of 10 days and nitrogen determinations on the feed.

The Miller and Bender method (47) consists essentially of measuring the net nitrogen stored in a test group of rats over a 7-10 day period for which the total nitrogen intake is measured. The net nitrogen stored of the test group is the difference between the total body nitrogen of the test group and that of a control group fed a nitrogen free diet. A correction is made for the control group if the nitrogen free diet contains traces of

nitrogen. Body nitrogen is determined from the body water content of the rats since Miller and Bender (47) have shown a high correlation between body nitrogen and body water.

The net protein utilization (N.P.U.) is calculated by applying the following equation:

$$N.P.U. = \frac{B - (B_k - I_k)}{I}$$

where B and B_k are total body nitrogen of the test group and non-protein group respectively, and I and I_k are the nitrogen intake of these two groups.

One of the objectives of this experiment was to test the supplementary value of the various test proteins with respect to maintenance, reproduction and growth in the mink. Since the test proteins only made up 44 per cent of the total protein in the rations, it is difficult to arrive at any true assessment of the amino acid complement supplied by the supplementary proteins because of the associated protein from the liver and cereal portion of the ration. The net protein utilization values of the five supplementary protein sources were measured by the Miller and Bender method (47) in order to arrive at an assessment of the amino acids supplied by these proteins. The net protein values of the five supplementary protein sources were measured on the rat and it may be argued that the values obtained may not be applicable to the mink. Generally, values for the biological estimation of proteins in the rat are applicable to other species

if the proteins are compared over the same physiological age period. The use of the mink as the test animal would, of course, be far too costly.

The net protein utilization values (N.P.U.) for the five supplementary protein sources and casein as a control, as determined by the Miller and Bender method (46) are presented in Table 19. The details of the method and the calculation are given in Appendix I.

Table 19

Net Protein Utilization Values of the Supplementary Proteins

Supplementary Protein	N.P.U.
Horse Meat	.54
Whale Meat	.54
Chicken Waste A	.43
Chicken Waste B	.33
Herring	.65
Casein	.61
Casein (72)	.60

The above values for the net protein utilization of the supplementary protein sources indicate that whole herring was the superior protein for the rat, even more favourable than casein. Horse meat and whale meat are identical but much lower than herring; the chicken wastes A and B are the lowest. Both horse meat and whale meat are essentially muscle tissue and

since the amino acid composition of muscle of all species is similar (56) it would be expected that the values for horse meat and whale meat should be alike. If the muscle of herring were used as the protein source instead of whole herring, the net protein utilization would still be higher because the presence of bone and skin reduces the digestibility. Both the chicken wastes are low as would be expected because of the relatively large amount of heads and feet present. These fibrous types of proteins are poorly utilized by most species.

It is difficult to explain the difference between the values for the two types of chicken wastes which only differ from one another in that Chicken A does not have the gall bladder and anal portion removed whereas Chicken B does. The difference in these values can best be explained on the basis of the variability of composition since the chicken waste used in the experiment was obtained from different sources and at different times of the year.

9. The Relationship Between Maintenance Feed Consumption and Body Weight in Adult Mink

Mink are normally fed a wet ration which has a hamburger-like consistency. The normal practice is to feed the ration on top of the wire cage and let the mink pull at the ration with its teeth. The mink often pulls more than it can eat in one bite and invariably some of the ration drops through the wire bottom of the cage onto the ground. Some mink pack their feed into the nest box where they will often leave some on the bottom of the box.

If the mink is given an excess of feed, some will be left on the wire to dry and harden. It is difficult to remove quantitatively unless the mink is taken from the cage. Practical observations over an extended period suggests that the feed loss from these various causes may be from 10-25 per cent of the feed offered. Accurate feed consumption data for the mink are extremely difficult to obtain unless time consuming techniques are employed. If such techniques are used one is severely limited in the number of animals that can be studied at any one time.

Since one half of the rations used for the maintenance experiment were canned, there was an opportunity to record feed consumption with some degree of accuracy. It was soon observed that the mink ate uniformly into the canned feed and that there was little or no wastage. The daily feed consumption could easily be recorded by daily weighing of the feed cans.

Daily feed consumption for all the adult mink (82 mink) on the canned rations, with the exception of those mink receiving the canned herring rations, were recorded for a five day period from Feb. 25 to March 1, 1956. The mink receiving the herring rations were excluded since they all had lost weight previous to thiamine supplementation and at this time they were regaining their lost weight. For this reason they had to be excluded from the maintenance category. A five day period was selected for recording feed consumption in the hope that it would permit some assessment of the variability in the daily intake values.

It was assumed that the body weights of the mink would not change over the short experimental period and as a result the mink were only weighed at the beginning of the experiment. The results of the maintenance experiment which showed no significant weight changes during this period further validated this assumption.

Since temperature changes can influence feed consumption (57) the daily minimum and maximum temperatures were recorded. The average minimum temperature over the five day period was 33.6°F. whereas the average maximum daily temperature was 40.4°F. The small variation in the daily temperature over the experimental period would reduce the day to day variation in feed intake that might have occurred had the temperatures been more variable. It is pertinent to record that the animals nest boxes were bedded with shavings and hence no opportunity was offered to produce a nest. More recent evidence suggests that feed consumption is markedly influenced by nesting conditions.

Feed consumption was recorded on a wet weight basis since the dry matter content of the canned rations averaged 37.7^{+}-2.1 per cent. The error in weighing a 1000 gm. mink is at least 2 per cent since the scale used was marked off in 20 gm. deviations.

The average daily feed consumption of the average adult mink for each canned ration is given in Table 20.

Table 20

Average Daily Feed Consumption of the Adult Mink

Ration	Ave. Feed	Ave. Group Weight
HAC-2	130 gm.	951 gm.
HC-4	121	929
WAC-6	134	828
WC-8	130	903
CACA-10	163	913
CAC-12	165	803
CBAC-14	167	903
CBC-16	177	807
Average	148	880

The feed consumption of the mink on the horse meat and whale meat rations are similar and lower than those of the animals on the chicken waste rations. It is pertinent to note that the average weights of the mink on the horse meat and the whale meat rations are higher than those on the chicken waste rations but their feed consumption is lower. If the rations were of the same nutritive value, the mink with the higher body weight would be expected to eat slightly more than those of lower body weight. Thus, there is an indication from Table 20 that there are differences in the nutritive value of the various rations.

The method of presenting the feed consumption data in Table 20 does not give any relationship between feed consumption

and body weight nor does it give any indication of variability. In order to establish the relationship between food consumption and body weight, the average feed consumption for each mink over the five day period was plotted against its body weight on log-log paper as shown in Fig. 5. The regression line relating feed consumption, in g.m. to body weight in gm. was calculated by the method of least squares for the 82 mink (11).

The relationship between feed consumption and body weight for all the mink in the trial may be expressed by the following equation:

$$Fl = 9.97\ W^{.40};\ +S_r = 25.9\%;\ -S_r = 20.6\%$$

where Fl is the feed consumption in gm. and W is body weight in gm. The correlation coefficient p = .867.

Regression lines relating feed consumption to body weight for the mink on the various supplementary protein groups were also calculated. These equations with their standard error of estimate are presented in Table 21.

Table 21

Relationship Between Feed Consumption and Body Weight

Protein Source	Rations	Equation	Standard Error
Horse Meat	HAC-2;HC-4	$Fl = 2.39\ W^{.58}$	+Sr = 21.8%;-Sr = 17.9%
Whale Meat	WAC-6; WC-8	$Fl = 6.69\ W^{.44}$	+Sr = 21.8%;-Sr = 17.9%
Chicken A	CACA-10; CAC-12	$Fl = 3.94\ W^{.55}$	+Sr = 18.8%;-Sr = 15.1%
Chicken B	CBAC-14; CBC-16	$Fl = 10.33\ W^{.40}$	+Sr = 21.8%;-Sr = 17.2%
ALL	ALL	$Fl = 9.97\ W^{.40}$	+Sr = 25.9%;-Sr = 20.6%

Fl = feed consumption in gm.
W = body weight in gm.

The regression lines in Table 21 with their standard errors indicate that there is considerable fluctuation in feed consumption at any given body weight. This is also bourne out by the spread of points in Fig. 5. Another point to consider is that the mink were eating out of cans and though they were checked daily there was the possibility that a mink might have been without feed for a short period. As a result the feed consumption on the following day would be considerably higher than the previous day. It was hoped that this daily fluctuation could, in part, be eliminated by averaging the feed consumption over the five day period.

The exponent for body weight of the equation relating the feed consumption of all the mink to their body weight is .40 and the exponents of the body weights for the equations of the protein sources range from .40 to .58 (Table 21). The National Research Council Nutrient Requirements for Foxes and Minks (49) have a suggested dry matter intake for mature mink on a maintenance diet. Their suggested dry matter intake is presented in Appendix J, Table 31 and Table 33.

The regression line relating body weight and dry matter intake was calculated for the National Research Council's recommendation. The regression line was $Fl = 4.33\ W^{.42}$; + Sr = 6.3%; - Sr = 5.9% where Fl is the dry matter intake in gm. and W is the body weight in gm. Their exponent is in close agreement with the one calculated from the experimental data (Table 21) which

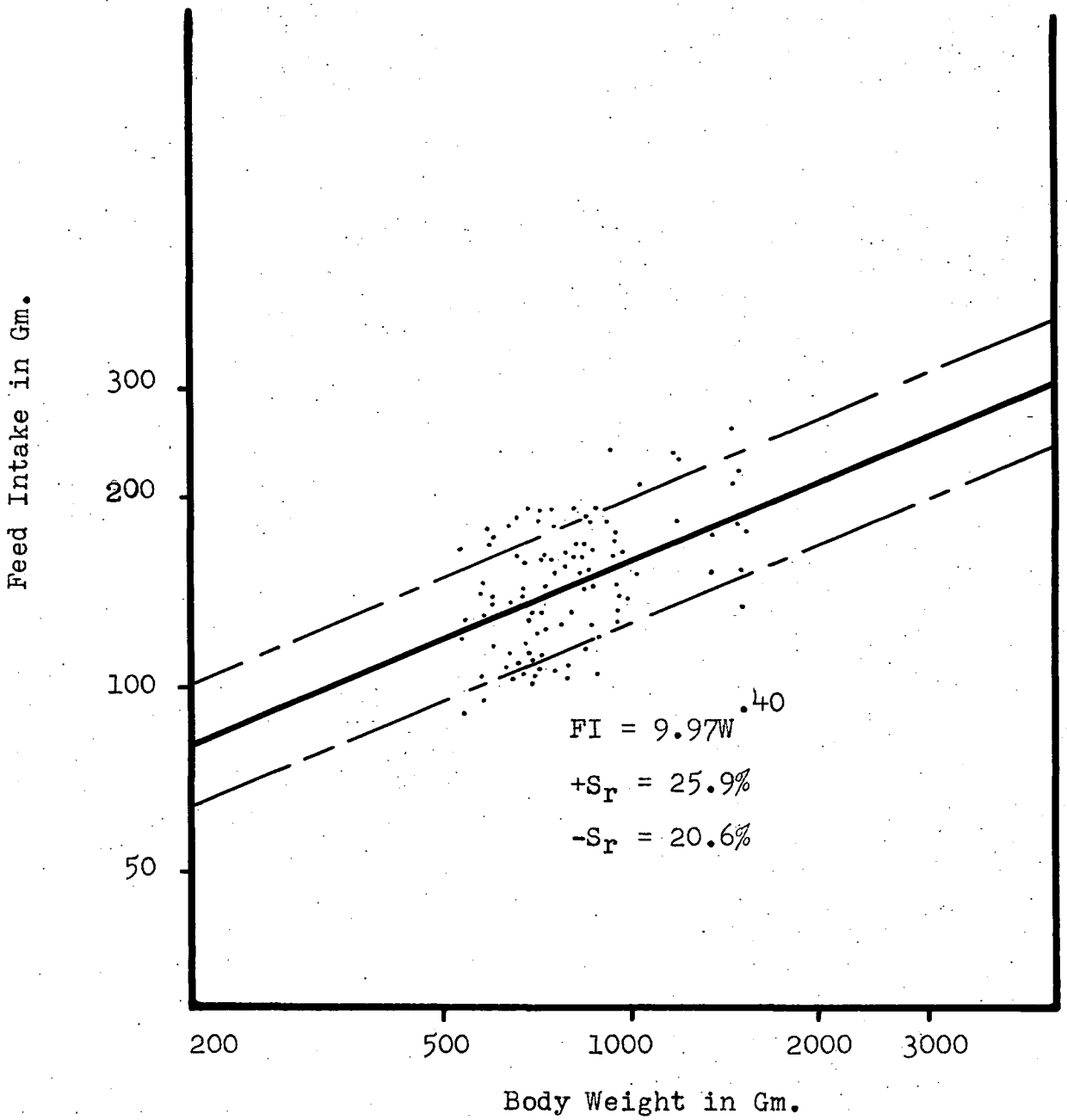
relates the feed consumption of all the experimental mink to their body weights. The exponent obtained from all the experimental mink suggest that for adult mature mink on a maintenance diet, a 100 per cent increase in body weight should lead to a 40 per cent increase in feed consumption. The exponents of W obtained for the various experimental rations range from .40 to .58. It is apparent that further studies on the relationship between feed consumption and body weight are required before any sound interpretation of the present results can be offered.

Brody (11) cites work in which it was found that feed consumption varied as the .50 power of body weight in non-laying domestic fowls, presumably on a maintenance diet. He comments that these results have to be confirmed before they can be accepted and he suggests that the expected exponent should be .70, since feed consumption should be the same function of body weight as basal metabolism.

Kansky (23) has shown that the relationship between the dry matter intake and body weight of mature Shorthorn cows is $DM = .17\ W^{.7}$ where DM is the dry matter intake of grass in lbs. and W is the body weight in lbs. Stephenson (59) has measured the feed consumption of mature beaver on two different rations. Recalculation of Stephenson's data (59) for his normal animals gave the following regression lines relating feed intake to body weight.

Fig. 5

The Relationship Between Feed Consumption and Body Weight



For his ration No. 204, the regression line was $Fl = 17.7\ W^{.90}$; + Sr = 15.3%; - Sr = 12.2% and for his ration No. 8, the regression line was $Fl = 20.6\ W^{.77}$; + Sr = 13.0%; - Sr = 11.5% where Fl is the daily feed intake in gm. and W is the body weight in pounds. Essentially the same animals were used in both trials. Thus, two different exponents of body weight were derived from feeding two nearly identical groups of animals two different rations. These rations presumably differed in nutritive value since ration 204 proved unsatisfactory whereas Ration 8 was adequate for the maintenance of the beaver.

The exponent of body weight in the regression between feed consumption and body weight has been shown to vary considerably. The biological significance of these differences is not understood. The present results and those of Stephenson (59) suggest further experimentation is necessary in order to permit an adequate understanding of the relationship between body weight and feed consumption.

In the present work it was possible to estimate the intake of digestible calories from the ration composition and the values obtained in the digestibility studies. The digestibility trials were performed on the frozen rations and not on the canned rations. It may be argued that the digestible calories per gram of dry matter should be higher in the canned rations. Canning could make the cereal portion more digestible

and hence increase the digestible energy content of the ration. This reasoning is supported by reports from Alberta ranchers who have been able to raise mink successfully on diets containing a high percentage of cooked cereal (63). However, Madramootoo (70) has shown with two different rations that cooking for 30 minutes at 15 lbs. pressure does not increase the digestibility. The question of cooking cereals for mink is then still debatable.

In order to calculate the digestible calories per gm. of ration, the average digestibilities from Table 17 were used. The average per cent composition of the rations was taken from Table 6. The following values for digestible energy were used (11); Protein 4.0 Cal./gm.; Fat 9.0 Cal./gm.; and Carbohydrate 4.0 Cal./gm. The digestible Calories per gm. of dry ration are presented in Table 22 for each ration group. Also presented in Table 22 are the total daily intake of digestible calories for each ration group whose average daily feed consumption was recorded in Table 20.

The daily intake of digestible Calories for each ration group tends to be similar. The horse meat rations have a high digestible caloric value per gm. and as a result the feed consumption is lower. The reversal is true with the chicken waste rations in that they have a low caloric value per gm. and as a result feed consumption is high. This data suggests that feed intake is governed by the digestible caloric content of the rations and the variations of feed consumption as presented in Table 20 are due to this difference.

Table 22

Digestible Calories per Gram of Dry Matter for the Ration Groups

Group	Digestible Cal./gm.	Daily Digestible Calories
Horse Meat	3.21	152
Whale Meat	2.87	143
Chicken Waste A	2.68	166
Chicken Waste B	2.24	145
Herring	3.04	
	Ave.	152 ± 10.8

Another study of feed consumption of mink receiving the canned rations was made during a six week period from Mar. 6 to April 15, 1956. In this study, cans of feed consumed per week were recorded for all the ration groups. At this time the Herring groups were supplemented daily with thiamine. The average weekly feed consumption is recorded in Table 34, Appendix J. The average daily dry matter intake per mink was calculated from the average number of cans of feed consumed, the number of mink per group and the dry matter content of the canned rations. The daily intake of digestible Calories was calculated from the dry matter intake and the digestible Calories per gm. of dry ration as presented in Table 22.

This estimation of the daily intake of digestible Calories should be an estimation of the digestible energy required

to maintain an average mink (The mink were in the ratio of 5 females to one male). An estimation of the requirement for digestible Calories may be made from a theoretical basis as described by Brody (11). The maintenance energy in digestible Calories tends to be twice the basal energy as calculated from B.M. = $W^{.73}$ where B.M. is the basal energy in Calories and W is body weight in Kilograms. In order to facilitate this calculation the mean body weights for each ration group were recorded. This calculation and the one mentioned above are presented in Table 23 which compares the calculated digestible energy consumed to that derived from twice B.M. = $70.5^{.73}$.

The average daily intake of digestible Calories per mink was 151 $\pm$ 7.9. The same value, as computed from the five day feeding trial was 152 $\pm$ 10.8 Calories (Table 22). The agreement between these two trials is excellent.

In all cases, the recorded daily intake of digestible Calories per mink was greater than that computed from theory. On the average, the mink received 16.7 per cent above the theoretical value. However, the theoretical value does not take into account any energy required for activity. A normal allowance for activity is about 10 per cent of the caloric intake but this allowance would probably be too low for the mink since it is very active. A more reasonable value would be 15 per cent of the caloric intake. If this assumption were correct, then the experimental mink were consuming sufficient Calories to meet their energy requirements for maintenance and activity.

An estimation of the daily intake of digestible Calories for a 1000 gm. mink may be calculated. The daily feed consumption was computed from the equation relating feed consumption to body weight (Table 21). This was 165 gm. of wet feed or 62.3 gm. of dry matter, assuming 37.7 per cent dry matter in the feed. The average digestible Calories per gm. of dry matter is 2.81 Calories. Then the estimated daily intake of digestible Calories for a 1000 gm. mink would be 175.

It is realized that the calculations performed in this section are subject to error because of the assumptions made but they are only intended to offer an approximation until studies on the energetics of mink nutrition are initiated.

Table 23

Comparison of the Consumed Digestible Calories to the Computed Calories for Maintenance

Canned Ration Group	Per Cent Dry Matter	Ave. Dry Matter Consumed per day	Group Mean Body Weight	Ave. Digestible Cal. Consumed per day	[9]Computed Dig. Cal. for Maintenance
Horse Meat	39.5	45.4 ± 2.7 gm.	899 gm.	146	130
Whale Meat	38.5	52.4 ± 8.2	826	150	120
Chicken A	37.7	59.2 ± 4.2	774	159	117
Chicken B	35.0	62.7 ± 11.1	932	140	134
Herring	39.8	53.3 ± 4.1	916	162	130
			827	151 ± 7.9	

9 From 2 times B.M. = $70.5W^{.73}$

VI. Summary

The primary objective of this study was to compare horse meat, whale meat, two types of chicken waste and herring as supplementary animal protein sources in rations for mink during maintenance, reproduction and growth. Part of the rations were canned in an attempt to prevent microbial spoilage. Another part, contained a commercial antioxidant in the hope that it would prevent oxidative rancidity.

The experimental results may be summarized as follows:

1. One hundred and seventy one female mink of assorted colour phases were fed the experimental rations during the maintenance period. There were no statistical differences between any of the rations.

2. In the reproductive phase of the experiment, the mink receiving the canned rations had essentially complete reproductive failure. Only 12 kits were born from 79 females bred. From the mink receiving the frozen rations, 194 kits were born from 88 females bred. The average of 2.2 kits per female was below normal. The average gestation period for all the mink was 53 ± 4.0 days.

3. Sixty female and 60 male kits were fed the frozen rations during the growth phase. The rations containing Horse Meat, Whale Meat and Chicken Waste A were statistically superior to those containing Chicken Waste B and Herring at $p = .01$. The difference between the two chicken wastes has been attributed to

the variability of the composition of chicken wastes. The below average performance of the mink on the Herring rations was not due to protein of inferior quality, as evidenced by its high Net Protein Utilization value, but probably due to the results of oxidative rancidity.

4. The antioxidant depressed growth of the kits. This suggests that at the level used, the antioxidant possessed some degree of toxicity.

5. The average percentage of thiamine destroyed in the rations during a 110 day storage period at 0^{o}F. was 29.2. The average percentage of thiamine destroyed between thawing the rations and subsequent consumption was 16.0. The antioxidant did not protect the destruction of thiamine in either of these two trials.

6. The average per cent digestibility for various ration components were determined by the chromic oxide method for 50 adult female mink receiving the frozen rations. They were:

Digestibility Class	Per Cent Digestibility
Total	61.5 ± 5.1
Nitrogen	70.6 ± 6.9
Fat	87.9 ± 1.9
"Carbohydrate"	26.9 ± 4.0

7. The Net Protein Utilization values of the fat extracted supplementary protein sources were determined in order to have an

assessment of their amino acid composition. They were:

Horse Meat	.54
Whale Meat	.54
Chicken Waste A	.43
Chicken Waste B	.33
Herring	.65

8. The relationship between feed consumption and body weight for all the mink receiving the canned rations was: $Fl = 9.97\ W^{.40}$ where Fl is the feed consumption in gm. and W is the body weight in gm. Regression equations were also determined for the other ration groups.

9. The variability in feed consumption of the mink receiving the canned rations was attributed to the differences in the digestible energy contents of the rations. The average daily digestible Calories for mink whose average weight was 827 gm. was calculated to be 151 ± 7.9.

Appendix A

Maintenance [4] Weight Data For The Female Adult Mink

Ration	No. of Females	Ave. Wt. Jan. 6	Ave. Wt. Feb.20	Ave. Wt. Mar. 21	Ave. Wt. Gained
HAF-1	9	792	801	860	68
HAC-2	10	756	799	779	23
HF-3	10	762	875	796	34
HC-4	9	712	770	745	33
WAF-5	10	711	696	740	29
WAC-6	10	717	802	811	94
WF-7	9	768	702	807	39
WC-8	10	732	749	743	11
CAAF-9	10	748	708	752	4
CAAC-10	8	704	743	709	5
CAF-11	9	705	711	784	79
CAC-12	7	788	771	814	26
CBAF-13	8	829	796	829	0
CBAC-14	8	835	806	816	-19
CBF-15	8	789	769	832	43
CBC-16	7	783	761	794	11
Ave. or Total	142	758	766	788	30

[4] All weights are in gm.

Appendix B

The Herring Mink During Maintenance

Table 24

Ration	No. of Females	Ave. Initial Wt.	Ave. Wt. Mar. 2	Ave. Wt. Loss
FAF-17	8	815 gm.	804 gm.	11 gm.
FAC-18	8	874	741	133
FF-19	8	899	839	60
FC-20	5	858	604	254
Ave. or Total	29	862	747	115

Table 25

Weight Data and Feed Consumption After Vitamin Supplementation

Supplement	Females	Ave. Wt. Data-gms. Mar.2	Mar.10	Mar.15	[5]Ave. Feed in Gms. per Day Prior Vitamins	After Vitamins
Thiamine	9	721	749	791	15	132
Folic & B_1	6	743	788	823	62	134
Folic	8	740	693	684	58	157
1% N.B.C.	6	870	805	837	60	154

5 Mink on Canned Rations Only

Table 26

Average Weight of Herring Female Mink

Ration	Jan. 20	Mar. 2	April 15
FAF-17	815 gm.	804 gm.	951 gm.
FAC-18	874	741	879
FF-19	899	839	995
FC-20	858	604	875

Appendix C

Mean Body Weight and Feed Consumption of Male Mink Receiving a Canned Horse Meat and Herring Ration

Fig. 1

The Horse Meat Group

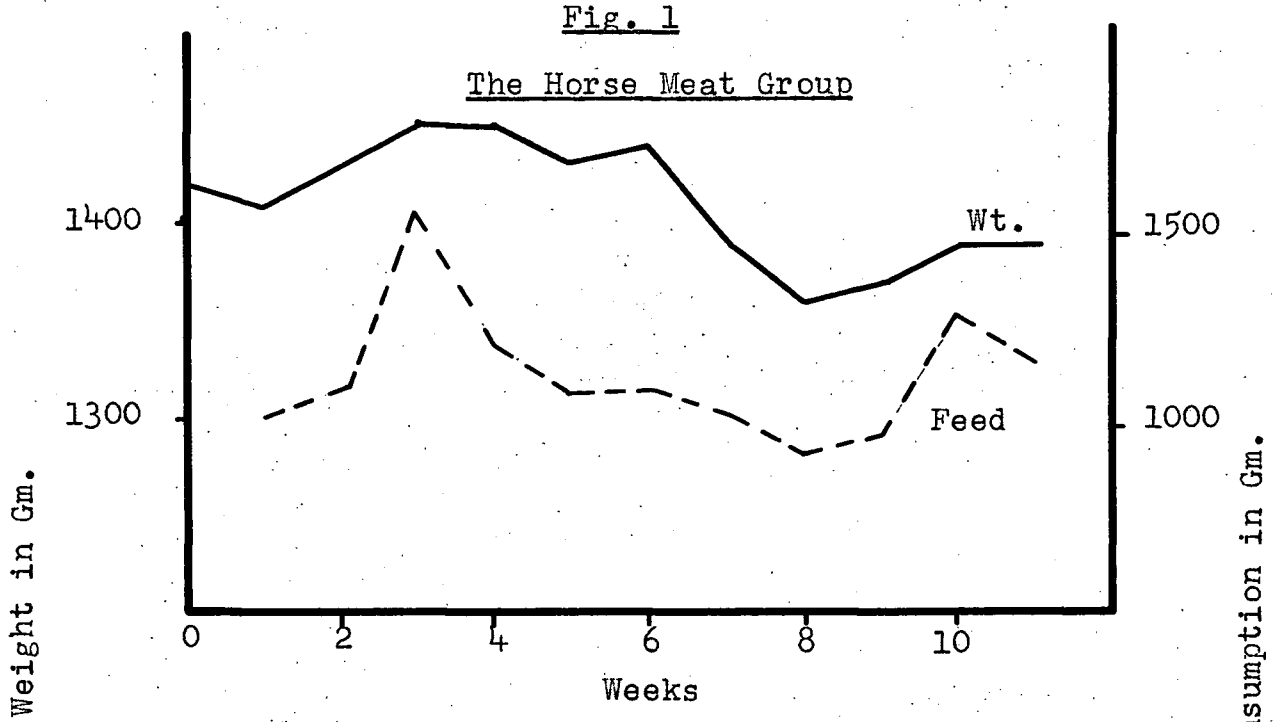
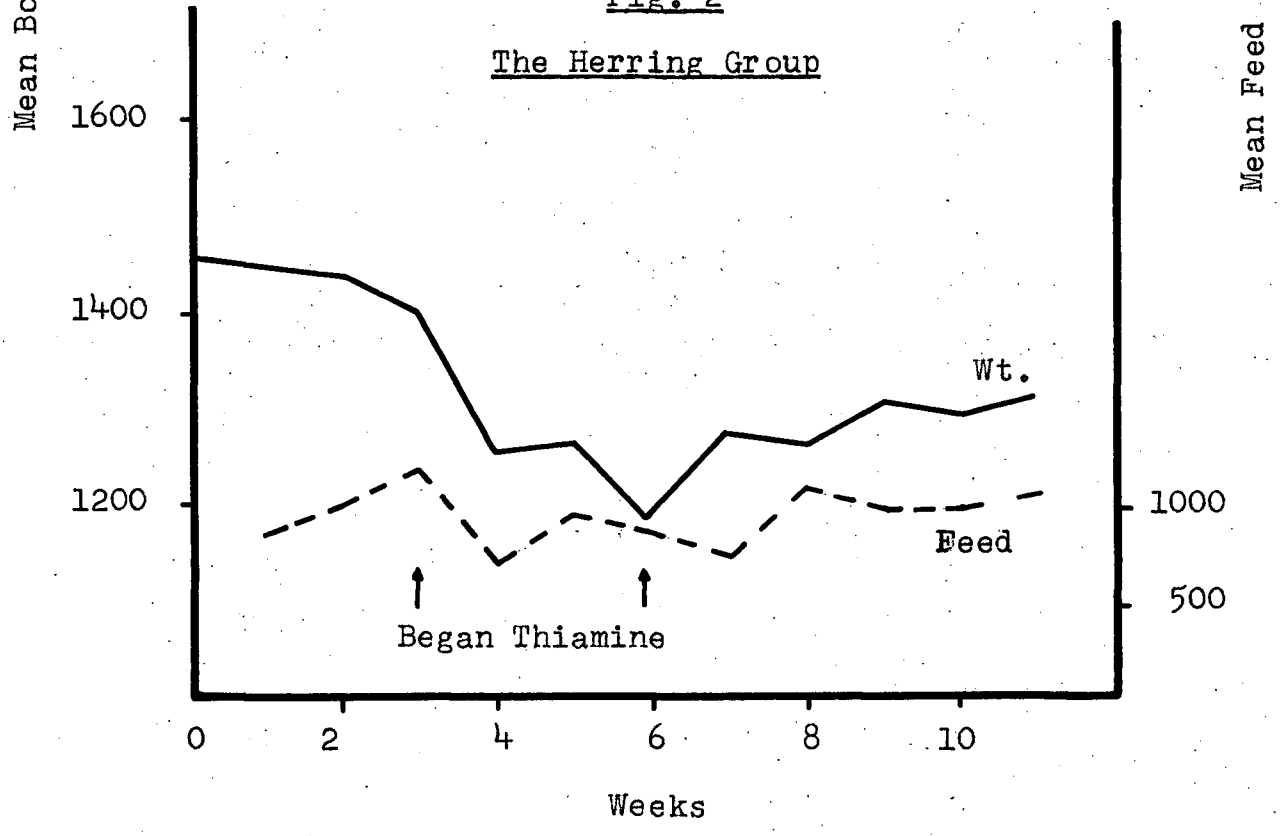


Fig. 2

The Herring Group



Appendix D

Reproduction and Weaning Data for the Experimental Groups

Ration	Total Females	Females Littered	Kits Born	Kits Weaned	Kits Born per Female	Kits Weaned per Female
HAF-1	9	4	8	2	0.9	0.2
HAC-2	10	2	?	0	0.0	0.0
HF-3	9	6	13	7	1.4	0.8
HC-4	8	0	0	0	0.0	0.0
WAF-5	10	2	5	5	0.5	0.5
WAC-6	9	1	1	1	0.1	0.1
WF-7	9	6	39	30	4.3	3.3
WC-8	10	0	0	0	0.0	0.0
CAAF-9	10	7	22	18	2.2	1.8
CAAC-10	8	0	0	0	0.0	0.0
CAF-11	9	4	12	6	1.3	0.7
CAC-12	7	0	0	0	0.0	0.0
CBAF-13	8	5	30	27	3.8	3.4
CBAC-14	8	0	0	0	0.0	0.0
CBF-15	8	5	28	27	3.5	3.4
CBC-16	7	0	0	0	0.0	0.0
FAF-17	8	3	13	2	1.6	0.3
FAC-18	8	4	11	4	1.4	0.5
FF-19	8	4	24	24	2.8	3.0
FC-20	4	1	?	0	0.0	0.0
Total or Ave.	167	54	206	[5]153	1.2	0.9

[5] 7 Kits were used for enzyme studies

Appendix E

Growth Data for the Experimental Groups

Table 27

Average Weights[6] of the Mink During Growth

Females

Ration	No.	June 28	July 14	July 27	Aug. 17	Sept. 28	Nov. 11	Total Gain
HAF-1	6	394	540	640	698	840	880	531
HF-3	6	340	492	607	687	933	953	613
WAF-5	6	333	497	627	740	933	1003	670
WF-7	6	347	537	663	790	1100	1120	773
CAAF-9	6	351	518	620	710	973	1007	656
CAF-11	5	340	494	620	697	1012	1024	684
CBAF-13	6	340	542	647	723	927	847	507
CBF-15	6	372	567	653	730	940	943	571
FAF-17	5	448	568	644	730	968	928	480
FF-19	6	368	502	583	627	823	853	485
Ave.	58	359	526	630	713	945	956	597

Males

HAF-1	6	403	693	920	1180	1593	1697	1294
HF-3	5	530	856	1088	1306	1832	1952	1422
WAF-5	6	382	602	793	997	1397	1447	1065
WF-7	6	416	685	943	1230	1587	1760	1344
CAAF-9	6	446	727	900	1126	1533	1690	1244
CAF-11	6	401	638	853	1067	1487	1607	1206
CBAF-13	6	430	733	900	1023	1460	1450	1020
CBF-15	6	457	728	913	1124	1413	1503	1046
FAF-17	6	450	723	890	1085	1537	1417	967
FF-19	3	347	527	707	807	1280	1333	987
Ave.	56	426	691	891	1095	1512	1585	1159

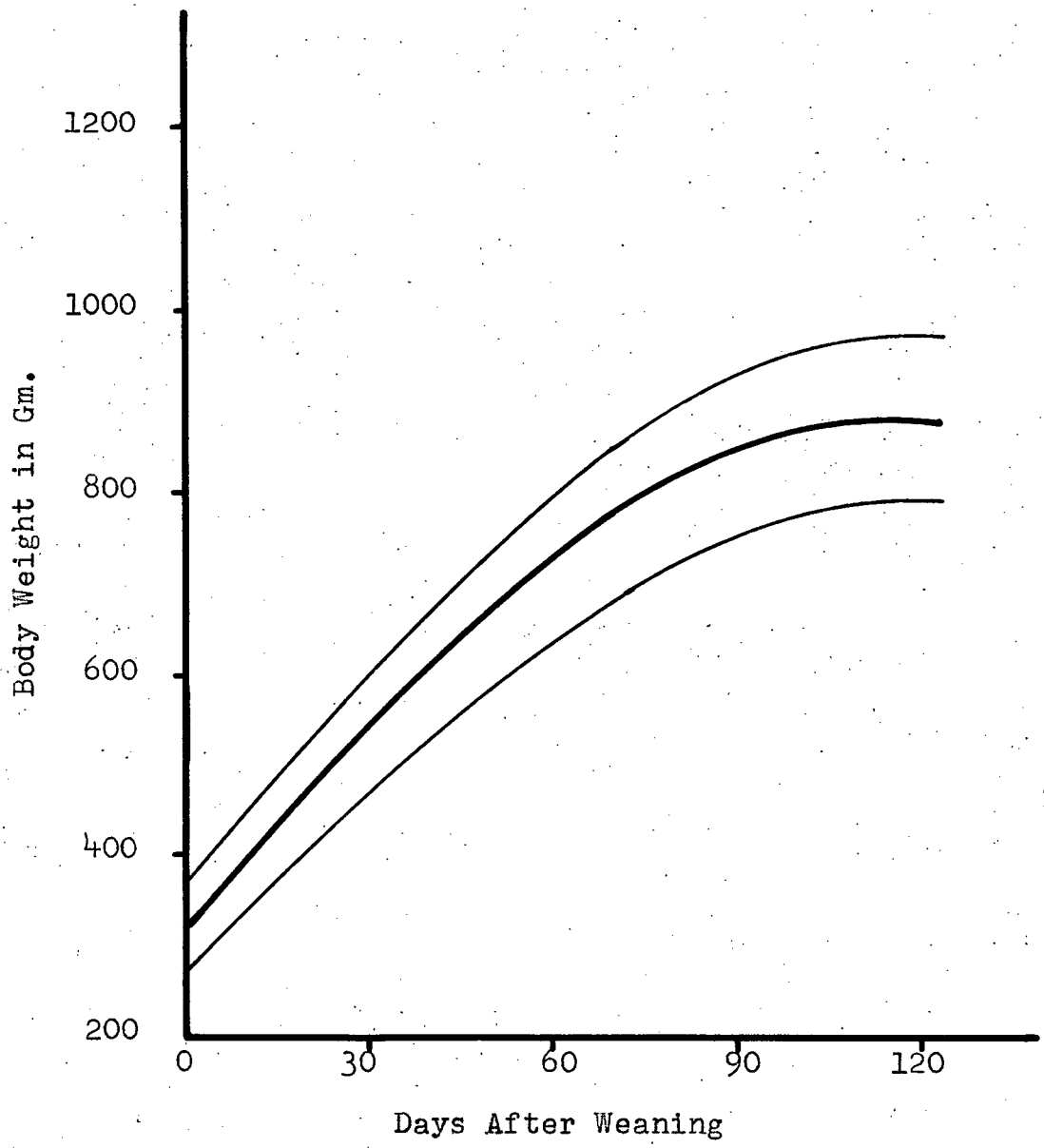
6 All weights are in grams

Appendix E

Growth Data for the Experimental Groups

Fig. 3

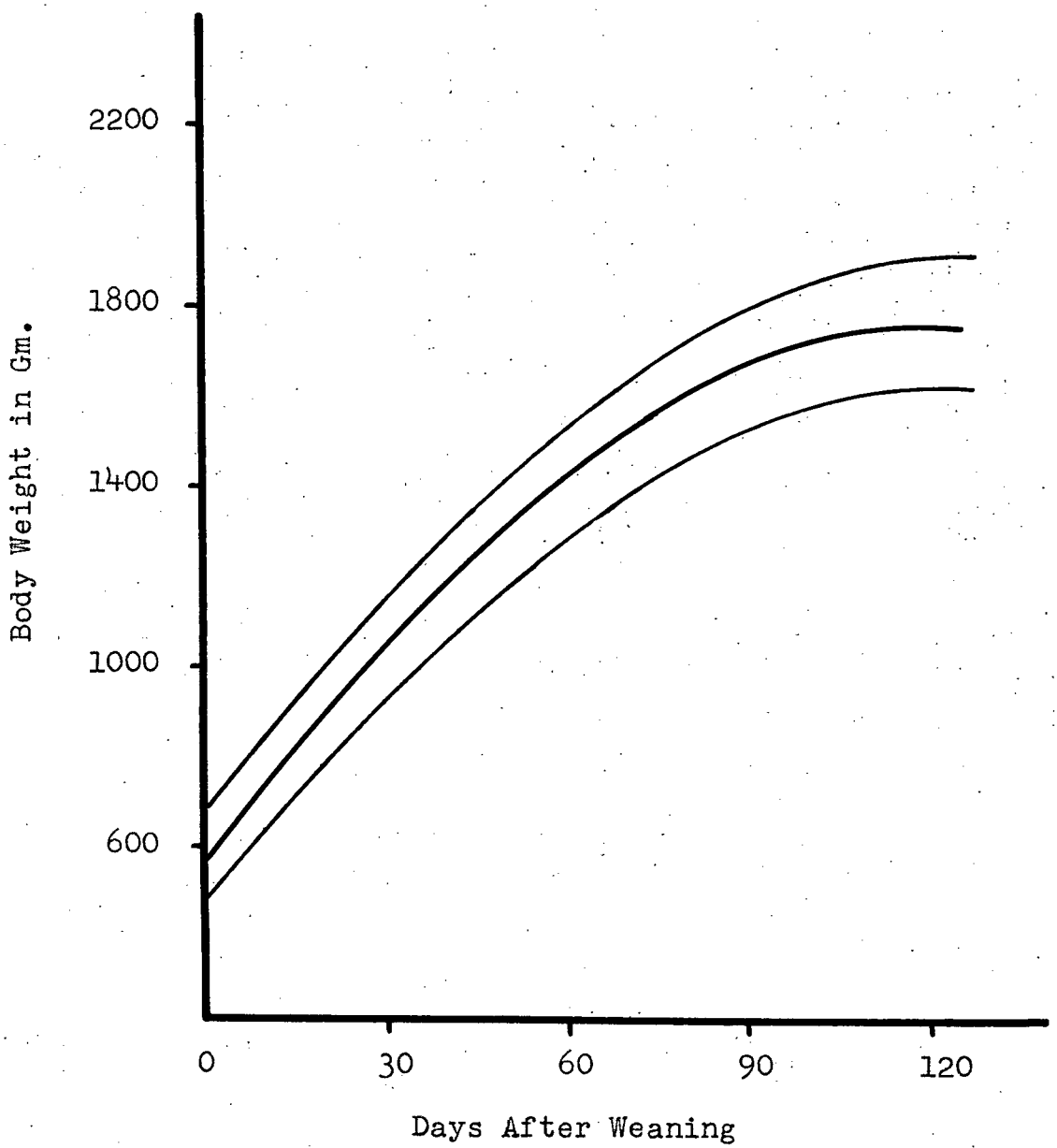
Average Growth Curve of the Female Kits



Appendix E

Growth Data for the Experimental Groups

Average Growth Curve of the Male Kits



Appendix F

Test For Significance Within a Group

1. Proteins

The analysis of variance for growth has indicated that there are differences in gain due to the different supplementary protein sources. The significance of the differences may be calculated by a modification of the "t" test. Now "t" is equal to:

"t" $= \frac{D}{SE}$ where D is the difference between two means and SE is the Standard Error between the two means. The Standard Error between two means in a variance analysis is:

the square root of $\frac{\text{Error Variance (2)}}{\text{No. of variates entering one mean}}$

Now the minimum difference between any two variate means required for significance = M.S.D. which is equal to:

M.S.D. = $\frac{\text{Error Variance (2)}}{\text{No. of variates entering one mean}}$ times "t" @ $p = .05$ of error degrees of freedom. This expression is derived from the "t" expression where D = (t) SE. Any difference between means which is larger than the calculated M.S.D. will then have significance at the $p = .05$ level. This calculation is applicable to any level of p.

For the proteins, M.S.D. is equal to:

M.S.D. = sq. root of $\frac{2242\ (2)}{4}$ times "t" @ $p = .01$ for 4 d.f.

$= 33.5\ (4.6)$

$= 154$ gm.

The mean gains for the five supplementary protein groups were:

Horse	965 gm.
Whale	963
Chicken A	948
Chicken B	786
Herring	729

The mean difference between Horse and Whale is 2 gm.; Horse and Chicken A is 17 gm.; Horse and Chicken B is 179 gm.; and Horse and Herring is 236 gm.; Whale and Chicken B is 177 gm.; Whale and Herring is 234 gm. The mean difference between Chicken A and Chicken B is 162 gm.; Chicken A and Herring is 219 gm. The mean difference between Chicken B and Herring is 57 gm. A mean difference greater than 154 gm. indicates significance between the means at p = .01.

2. Interaction between Sex and Proteins

The M.S.D. for the interaction between Sex and Proteins at p = .05 was calculated to be 132 gm. The mean gains for the females and males receiving different supplementary protein sources were:

	Females	Males
Horse	572 gm.	1358 gm.
Whale	722	1205
Chicken A	670	1225
Chicken B	539	1033
Herring	483	977

For the females the mean difference between Horse and Whale is 150 gm.; Horse and Chicken A is 98 gm.; Horse and Chicken B is 63 gm.; Horse and Herring is 89 gm. The mean difference

between Whale and Chicken A is 52 gm.; Whale and Chicken B is 183 gm.; Whale and Herring is 239 gm. The mean difference between Chicken A and Chicken B is 131 gm.; Chicken A and Herring is 187 gm. The mean difference between Chicken B and Herring is 56 gm.

For the males the mean difference between Horse and Whale is 153 gm.; Horse and Chicken A is 133 gm.; Horse and Chicken B is 325 gm.; Horse and Herring is 479 gm. The mean difference between Whale and Chicken A is 20 gm.; Whale and Chicken B is 172 gm.; Whale and Herring is 228 gm. The mean difference between Chicken A and Chicken B is 192 gm.; Chicken A and Herring is 248 gm. The mean difference between Chicken B and Herring is 56 gm.

For significance between the means of any two rations, the difference must be greater than 132 gms.

Appendix G

Procedure for Thiamine Determinations

There are many methods for the assay of thiamine which may be classified into animal, microbiological, chemical and physical methods. The animal methods are tedious and the results vary considerably. The microbiological methods are much more rapid and accurate but the main disadvantage is the tendency for substances other than thiamine to respond in the same way as the vitamin, and as a result the values may be high. Correction blanks have improved these methods considerably but the methods are not too applicable for routine determinations. Chemical analysis are considered to be more applicable to routine determinations than most other methods. The two main chemical methods are based on colourimetry and flourimetry. The thiochrome fluorimetric method is more widely applicable to foods and feed products than are the colormetric methods. Physical methods, based on spectrophotometers, are only used for relatively pure solutions of thiamine.

The method chosen for the assay of thiamine, as recommended by the Association of Vitamin Chemists, was the thiochrome procedure because of its adaptability to feed products and its ease for routine analyses which occurred from weekly to monthly time intervals.

The procedure followed was identical to that in the reference (14) except for the following modifications:

(1) Reagents

Isobutyl alcohol was purified in the following manner to remove any flourescense (51). Isobutyl alcohol was dried by shaking with anhydrous sodium sulfate. The mixture was then shaken for five minutes with 5 grams of Norit per 100 ml. of isobutyl alcohol. After filtration, the isobutyl alcohol was distilled in an all glass distilling apparatus and the fraction boiling between 105-108°C. was collected.

(2) Procedure

(a) Extraction

The fluorometric attachment for the Beckman, Model DU Spectrophotometer was used to measure the flourescence produced by thiochrome. It was established that the flourometric attachment was about one half as sensitive as the flourometers recommend for the procedure. As a consequence, the initial sample had to contain 20 to 60 mcg. of thiamine instead of 10 to 30 mcg. For routine thiamine analysis on the rations the sample size was 30 gm. because of the insensitivity of the flourometer and the low initial thiamine content of the rations. Because of the larger sample size, the acid hydrolysis was extended from 30 to 45 minutes in order to ensure complete solution of the various forms of thiamine. The enzymatic hydrolysis was an over-night hydrolysis. After enzymatic hydrolysis, the extracts were centrifuged in plastic centrifuge tubes and then filtered.

(b) Purification

No changes were made in the purification procedure but some techniques are worth noting. To avoid banding while filling the adsorption columns the following technique was found satisfactory. A small wad of glass wool and a small cork were inserted into the small end of the column. With the column filled to one inch from the top with distilled water, activated decalso was slowly added while the water-decalso mixture in the column was stirred vigorously with a stirring rod. The continuous stirring enabled the decalso to settle at a uniform rate and thus avoid banding.

It was established that purification by the columns was necessary. The test thiamine solution contained 5×10^{-7}gm. thiamine hydrochloride per ml. and was prepared by diluting to 100 cc. with distilled water, 10.0 ml. of intermediate thiamine solution plus 75 ml. of 0. 1N H_2SO_4 and 5 ml. of Sodium Acetate solution.

Small, funnels, made from test tubes, were used to collect the KCl elutate from the columns into the 25 ml. volumetric flasks. The use of these funnels prevented the formation of air traps in the volumetric flasks.

(c) Conversion to Thiochrome

Ten ml. of the acid KCl elutate and 5 ml. of alkaline ferricyanide were used for the formation of thiochrome. The higher relative concentration of ferricyanide was used in order

to ensure the complete conversion of thiamine to thiochrome. The amount of thiochrome to be oxidized was larger because of the insensitivity of the flourometer.

(d) Measurement of Thiochrome

Thiochrome was measured by the flouorometric attachment of the Beckman, Model DU Spectrophotometer utilizing the proper filters and the sensitivity knob set at 0.1.

(3) Sample Preparation

Approximately 500 gm. of sample were taken randomly from a block of feed in the feed storage refrigerator. The sample was cut into small pieces with a knife and then ground in a Hobart meat grinder from which the assay samples were taken. All samples were analyzed in duplicate and the assays were usually within 5 per cent of one another.

Appendix H

Digestibility Studies on the Frozen Rations

(1) Methods and Materials

Five adult female mink from each of the ten frozen rations (Table 7) were fed for a 3 day period (July 22-25, 1956) their regular rations which contained approximately 40 mg. of chromic oxide per gm. of dry ration. The chromic oxide was mixed into a 3 day feed supply by means of a Hobart mixer until a uniform green colour resulted. The feed was kept chilled in the refrigerator between feeding and just prior to feeding, the feed was re-mixed to ensure uniform distribution of the indicator. The faeces were collected on wax paper which was placed below the mink cages. At the end of the collection period, samples of feed and faeces were collected, dried at 100°C. for 48 hours and then ground in a Wiley Mill, using the 40 mesh screen. The samples were stored in tightly stoppered bottles until analyzed.

(2) Chemical Procedures

Both Worthen (65) and Madramootoo (42) have shown that the chromic oxide method is satisfactory for measuring the digestibility of mink rations and that the method is comparable to the total collection method. Chromic oxide in the feed and faeces was determined by the method as described by Bolin (10). This method has been used successfully in the Animal Nutrition Laboratory, University of British Columbia, in studying the

digestibility of a wheat-herring ration when fed to mink (65). The Coleman spectrophotometer was used in preparing a standard curve between the concentration of 0.2 to 0.14 mg. of chromic oxide per ml. of solution. A straight line relationship existed between the logarithim of the per cent transmission and the concentration of chromic oxide.

Nitrogen in the feed and faeces were determined by a standard method (46). Fat in the feed and faeces was determined by the method described by Bailey (4). The per cent total digestibility, nitrogen digestibility, fat digestibility and "carbohydrate" digestibility for each mink appears in Table 28.

The total digestibility may be calculated from the data so obtained. For convenience the following formula was derived.

$$\text{Per Cent Total Digestibility} = 100\left(1 - \frac{\text{per cent } Cr_2O_3 \text{ in feed}}{\text{per cent } Cr_2O_3 \text{ in faeces}}\right)$$

The individual nitrogen digestibilities for each mink can be calculated as follows: A sample calculation is given for mink No. 1 (Table 28).

1 gm. feed has 0.0465 gm. N $\equiv$ 0.041 gm. Cr_2O_3

1 gm. faeces has 0.0352 gm. N $\equiv$ 0.0138 gm. Cr_2O_3

For 1 gm. Cr_2O_3 in the feed there is $\frac{0.0465}{0.041}$ = 11.34 gm. N

For 1 gm. Cr_2O_3 in faeces there is $\frac{0.0352}{0.0138}$ = 2.55 gm. N

Nitrogen Absorbed = 11.34-2.55 = 8.79 gm. N

Per cent nitrogen digested = $\frac{8.79(100)}{11.34}$ = 77.5 per cent.

The following derived formula can be used to give directly the nitrogen digested.

$$\text{Per cent nitrogen digested} = \left(1 - \frac{\%\text{N in faeces}}{\%\ Cr_2O_3\text{ in faeces}} \cdot \frac{\%\ Cr_2O_3\text{ in feed}}{\%\text{ N in feed}}\right)100$$

The fat digestibility was calculated in the same manner as the nitrogen digestibility.

The term "carbohydrate" used in this study requires some explanation. When a ration is viewed on the basis of its proximate composition, the main constituents are protein (Nitrogen 6.25), fat or ether extract, water, ash, fibre and nitrogen free extract. In this study the term "carbohydrate" refers to the fibre and nitrogen free extract terms of a proximate analysis since these two are chiefly composed of carbohydrates. Since this study was done on mature adult mink and the ash content of the ration was low, the per cent digestibility of the ash was assumed to be zero for the purpose of calculating the per cent "carbohydrate" digestibility. "Carbohydrate" digestibility can then be calculated from the per cent total digestibility, nitrogen digestibility, fat digestibility, per cent feed fat and the per cent feed nitrogen as described in the following formula:

"Carbohydrate" Digestibility =

$$\text{Per cent total digestibility} - \left[\frac{(\%\text{ feed fat})(\text{fat digestibility})}{100}\right.$$

$$\left.\text{plus } \frac{(\%\text{ Feed Nitrogen})(6.25)(\text{Nitrogen digestibility})}{100}\right]$$

A sample calculation for mink No. 1 is:

"Carbohydrate" Digestibility =

$$70.3 - \frac{(17.3)(92.2)}{100} \text{ plus } \frac{(4.7)(6.25)(77.5)}{100} = 31.5 \text{ per cent.}$$

Table 28

Total Digestibility, Nitrogen Digestibility, Fat Digestibility and "Carbohydrate Digestibility" for Individual Mink

Ration	Mink No.	Per Cent Faecal N	Per Cent Fat Faecal	Per Cent Feed Cr_2O_3	Per Cent Faeces Cr_2O_3	Per Cent Feed N	Per Cent Feed Fat	Per Cent Total Digestibility	Nitrogen Digestibility	Fat Digestibility	Calculated "CHO" Digestibility
HAF-1	1	3,5	4.5	4.1	13.8	4.7	17.3	70.3	77.5	92.2	31.5
	2	3.2	4.0	4.1	13.7	4.7	17.3	70.1	79.0	93.1	30.8
	3	4.5	4.8	4.1	11.1	4.7	17.3	63.1	64.0	89.7	28.8
	4	3.4	2.9	4.1	10.1	4.7	17.3	59.4	70.0	93.2	22.7
	5	4.7	4.5	4.1	10.2	4.7	17.3	59.8	78.5	89.5	21.2
HF-3	25	4.2	3.5	4.2	10.6	5.3	19.5	60.4	68.6	92.9	19.6
	26	4.1	5.2	4.2	17.5	5.3	19.5	76.0	82.8	93.6	30.3
	27	5.3	3.3	4.2	13.9	5.3	19.5	69.8	69.6	94.9	28.2
	28	4.0	2.8	4.2	18.6	5.3	19.5	77.4	83.0	96.7	31.3
	30	3.1	4.2	4.2	15.8	5.3	19.5	73.4	84.5	94.3	27.0
WAF-5	49	4.5	4.3	4.9	13.9	4.9	15.0	64.5	67.7	89.9	30.3
	50	3.3	5.8	4.9	10.9	4.9	15.0	65.0	69.5	82.6	31.3
	51	3.3	7.2	4.9	13.1	4.9	15.0	62.6	75.1	82.0	27.3
	52	2.7	4.1	4.9	12.6	4.9	15.0	61.1	78.5	89.4	23.7
	53	2.2	3.3	4.9	13.8	4.9	15.0	64.5	84.2	92.0	24.9
WF-7	73	4.1	4.5	4.0	11.8	5.2	11.2	66.1	73.2	86.4	32.6
	74	4.6	4.9	4.0	8.8	5.2	11.2	54.5	59.6	80.2	26.1
	75	3.6	3.8	4.0	10.3	5.2	11.2	61.2	72.6	86.8	27.9
	77	3.8	4.5	4.0	9.0	5.2	11.2	55.6	71.2	82.1	23.3
	79	2.8	5.5	4.0	10.7	5.2	11.2	62.6	79.5	81.6	27.7
CAAF-9	97	3.0	3.5	4.4	14.6	5.2	9.7	69.9	82.5	89.1	34.5
	98	2.9	4.8	4.4	9.6	5.2	9.7	54.2	74.3	77.3	22.6
	99	4.6	4.8	4.4	11.4	5.2	9.7	61.4	65.6	81.9	32.3
	100	5.3	1.8	4.4	11.6	5.2	9.7	62.1	61.0	93.0	33.3
	101	4.5	4.5	4.4	10.3	5.2	9.7	57.3	63.2	80.2	29.0
CAF-11	120	3.8	2.5	4.1	9.6	5.0	10.0	57.3	67.4	89.3	27.3
	121	3.9	2.5	4.1	11.9	5.0	10.0	65.5	73.0	92.3	33.5
	122	4.5	3.5	4.1	12.6	5.0	10.0	67.5	70.2	88.6	36.7
	124	3.7	3.0	4.1	9.3	5.0	10.0	55.9	67.2	86.8	26.2
	125	4.0	4.2	4.1	9.8	5.0	10.0	58.2	66.2	82.4	29.3
CBAF-13	145	3.8	3.8	4.7	12.4	5.3	10.0	62.1	72.6	85.6	29.4
	146	5.3	4.1	4.7	11.6	5.3	10.0	59.5	59.2	83.3	31.6
	147	4.3	5.1	4.7	12.1	5.3	10.0	61.2	68.2	80.2	30.6
	148	4.6	5.0	4.7	12.6	5.3	10.0	62.7	67.7	81.3	32.2
	149	4.6	3.2	4.7	14.1	5.3	10.0	66.7	71.7	89.4	34.0
CBF-15	169	4.5	3.2	4.6	10.9	5.7	12.7	57.8	66.6	89.4	22.7
	170	3.8	3.2	4.6	12.1	5.7	12.7	62.0	74.6	90.4	23.9
	171	4.5	3.3	4.6	11.5	5.7	12.7	60.0	68.9	89.6	24.1
	172	3.9	3.9	4.6	11.0	5.7	12.7	58.2	71.6	87.2	21.6
	173	4.1	4.8	4.6	9.9	5.7	12.7	53.5	66.5	82.4	19.3
FAF-17	196	4.7	3.3	4.5	8.6	4.6	14.5	47.7	46.3	88.1	21.6
	198	4.9	3.7	4.5	9.7	4.6	14.5	53.6	49.9	88.6	26.5
	199	2.9	6.0	4.5	10.9	4.6	14.5	58.7	74.1	82.9	25.4
	200	3.9	3.9	4.5	10.0	4.6	14.5	55.0	61.5	87.9	24.6
	201	2.7	4.2	4.5	8.3	4.6	14.5	46.0	68.3	84.3	14.2
FF-19	217	3.0	4.3	4.2	11.6	4.4	19.8	63.8	75.0	92.1	25.0
	218	2.5	3.5	4.2	13.2	4.4	19.8	68.2	81.8	94.4	27.0
	219	2.6	3.5	4.2	13.4	4.4	19.8	68.7	81.6	94.5	27.6
	220	4.3	4.3	4.2	7.7	4.4	19.8	45.4	47.1	88.1	15.0
	221	2.6	5.3	4.2	9.7	4.4	19.8	56.7	74.4	88.4	18.7

Appendix I

The Net Protein Utilization of the Supplementary Proteins

(1.) Preparation of the Samples

About 1000 gm. of each protein sample were dried at 60°C. in a tunnel drier. Each sample was then broken into fine pieces, wrapped in filter paper and cheese cloth and extracted ten times with ethylene dichloride in the large fat extractor (12 hours). The fat was removed to ensure a uniform sample since the proteins varied in their fat content. The samples were air dried and then ground into a fine powder in a Wiley hammer mill. Duplicate samples of each protein source were analyzed for protein (N x 6.25)) by a standard method (46). The average results of the per cent protein of the fat extracted supplementary protein sources are presented in Table 29.

Table 29

Average Per Cent Protein of the Fat Extracted Protein Sources

Protein Source	Per Cent Protein
Horse Meat	82.2
Whale Meat	86.6
Chicken Waste A	67.1
Chicken Waste B	59.4
Herring	79.7
Casein (Vitamin Free)	91.0

(2.) Method of Assay

The method of assaying the net protein utilization of the supplementary protein sources is essentially the method of Miller and Bender (47) as modified by Robertson (52) for the Wistar Rat. Details of the method are described under Experimental.

(3.) Experimental

(a) Animals

The rats used were of the Wistar strain which were weaned at 20 days and fed for a week prior to the experiment on a stock ration. Each experimental group consisted of 3 females whose initial weights were between 65-70 gm. and 3 males whose initial weights were between 65-70 gm. The total initial weights of all the groups were balanced with respect to one another. There were 7 experimental groups of rats; one group for each of the five supplementary proteins to be tested, one group on a known protein, casein, to serve as a control and one group on a non-protein diet. The animals were housed in individual pans and were provided with fresh water daily.

(b) Rations

The Miller and Bender asaay method (47) requires the test proteins to be fed at the 10 per cent level in a common non-protein basal ration which is fed to the non-protein group. The composition of the non-protein basal ration is given in Table 30.

Table 30

Composition of the Non-Protein Basal Ration

Lard	22.3
Corn Starch	61.2
Sucrose	7.5
Vitamin Mix (NBC)	1.5
Salts (U.S.P. No. 2)	7.5
	100.0 gm.

Each experimental diet, with the exception of the non-protein diet, was adjusted to the 10 per cent protein level with the respective protein source.

(c) Procedure

Each experimental group was fed their respective diet for seven days. Weight gained or lost and total feed consumption were recorded for each rat. At the end of the experimental period, the rats were killed with chloroform and the abdominal, thoracic and cranial cavities were opened by incision with scissors. The rats were then dried at 105 deg. C. for 48 hours in the tunnel drier in order to determine body water content. The nitrogen content of the rations were determined by a standard method (46). Final weight, initial weight, dry weight, body nitrogen, feed consumption, per cent nitrogen of the rations and group totals are given in Table 31.

(d) Calculations

Body nitrogen was calculated from the body water content of the rats. Miller and Bender (47) have shown a high correlation between body nitrogen and body water for rats of the hooded strain which were from 33-57 days old. Robertson (52) has verified this relationship in the Wistar rat by determining the nitrogen to water ratio in 33 rats whose weights ranged from 40-100 gm. and developed the following equation for the calculation of body nitrogen from body water.

$y = 27.89\ x^{1.077}$ y = mg. body nitrogen

$+ S_r = 3.74\%$ x = gm. body water

$- S_r = 3.61\%$

From the above equation the body nitrogen of each rat was calculated. The net protein utilization (N.P.U.) was determined from the following equation as presented by Miller and Bender (47).

$$N.P.U. = \frac{B - (B_k - I_k)}{I}$$

B and B_k are the total body nitrogen of the animals on the test and non-protein diets respectively, and I and I_k are the nitrogen intake of the two groups.

Table 31

Data For Net Protein Utilization Calculations

Protein Source	Rat No.	Final Wt. Gm.	Initial Wt. Gm.	Dry Wt. Gm.	Body Nitrogen mg.	Feed Consumption Gm.
Horse Meat %N = 1.65	1f[7]	69	65	22	1758	35
	2 f	82	63	28	2037	52
	3 f	75	62	23	1959	46
	4 m[88]	77	71	23	2037	49
	5 m	87	70	28	2244	54
	6 m	86	68	28	2203	58
Total			399		12238	294
Whale Meat %N = 1.60	1 f	76	63	24	1959	43
	2 f	80	67	27	2000	48
	3 f	83	65	27	2123	54
	4 m	83	69	26	2163	50
	5 m	77	67	25	1959	49
	6 m	81	70	25	2123	59
Total			401		12327	303
Chicken Waste A %N = 1.50	1 f	74	65	24	1880	58
	2 f	75	62	25	1880	49
	3 f	67	62	20	1758	53
	4 m	76	69	23	2000	43
	5 m	77	65	23	2037	49
	6 m	86	73	28	2123	54
Total			396		11678	306
Chicken Waste B %N = 1.75	1 f	74	65	26	1758	58
	2 f	75	62	24	1923	49
	3 f	67	62	25	1553	53
	4 m	76	69	27	1837	43
	5 m	77	65	23	2037	49
	6 m	86	73	24	2366	54
Total			396		11477	306

7 f = female

88 m = male

Table 31 (Continued)

Protein Source	Rat No.	Final Wt. Gm.	Initial Wt. Gm.	Dry Wt. Gm.	Body Nitrogen	Feed Consumption Gm.
Herring	1 f	80	65	23	2163	49
%N = 1.60	2 f	84	66	27	2163	48
	3 f	74	60	24	1880	47
	4 m	85	71	28	2163	54
	5 m	77	62	23	2037	46
	6 m	92	72	28	2449	56
Total			396		12855	300
Casein	1 f	69	60	20	1837	46
%N = 1.60	2 f	83	65	27	2123	56
	3 f	80	63	26	2037	48
	4 m	86	65	27	2244	60
	5 m	81	63	26	2080	52
	6 m	88	77	26	2366	42
Total			393			304
Non-	1 f	54	61	16	1400	43
Protein	2 f	59	66	17	1556	23
%N = 0.00	3 f	56	63	16	1469	25
	4 m	59	67	16	1596	31
	4 m	62	70	19	1596	36
	4 m	64	71	19	2101	38
Total			398		9718	196

Appendix J

The Relationship Between Feed Consumption and Body Weight

Table 32

National Research Council Daily Dry Matter Intake for Mink on a Maintenance Diet (11)

	Wt. lbs.	Food lbs.	Wt. lbs.	Food lbs.
Under-lbs.	2.0	.18	1.5	.15
lbs.	2.0 - 3.0	.19	1.5 - 2.0	.16
Over lbs.	3.0	.20	2.0	.17

Table 32 was recalculated to give an average relationship between body weight and dry matter intake and this is represented in Table 33.

Table 33

Computed National Research Council Relationship Between Body Weight and Feed Intake

Wt. lbs.	Food lbs.
1.5	.15
1.75	.16
2.0	.175
2.5	.19
3.0	.20

Table 34

Weekly Feed Consumption of Mink on the Canned Rations

Week	Horse Meat (23)	Whale (24)	Chicken Waste A (19)	Chicken Waste B (19)	Herring (17)
1	30 cans	38	34	32	27
2	35	40	36	40	25
3	36	51	38	52	33
4	33	41	41	52	32
5	34	32	40	42	30
6	34	45	40	42	27
Ave.	37.7 ± 2.05	41.7±6.40	3.82±2.73	43.3±7.66	29.0±2.23

The average weight of one can of feed was 548 gm. The numbers in brackets are the number of mink per group.

VIII. Bibliography

1. Allison, J.B., Biological Evaluation of Proteins, Physiol. Revs., 35, 664-700, 1955.

2. Ashton, W.M., Elements of Animal Nutrition, Griffen, London, 120, 1950.

3. Bacharad, A.L., and Rendle, T., ed., The Nation's Food, London, 1946.

4. Bailey, C.B., Studies in Animal Growth, Master's Thesis, The Library, University of British Columbia, 1956.

5. Banks, A., The Development of Rancidity in Cold-Stored Herrings: The Influence of Some Antioxidants, J. Sci. Food Agric., 3, 1952.

6. Bate-Smith, E.C., and Morris, T.N., ed., Food Science, Cambridge, 1952.

7. Bernard R., Smith, S.E., and Maynard, L.A., The Digestion of Cereals by Minks and Foxes with Special Reference to Starch and Crude Fibre, Cornell Vet., 32, 29-36, 1942.

8. Blanck, F.C., ed., Handbook of Food and Agriculture, Reinhold, 1955.

9. Blaxter, K.L., and Brown, F., Vitamin E in the Nutrition of Farm Animals, Nutr. Abst. Revs., 22, 1-12, 1952-1953.

10. Bolin, D.W., King, R.P., and Klosterman, E.W., A Simplified Method for the Determination of Chromic Oxide (Cr_2O_3) when Used as an Index Substance, Science, No. 3023, 116, 634, 1952.

11. Brody, S., Bioenergetics and Growth, Reinhold, New York, 1945.

12. Day, R.M., Ranch Observations, American Fur Breeder, Aug., 1953.

13. Diqaud, A., and Fournier, P., Queleques aspects du probleme de la conservation de la vitamin B_1, Bull. Soc. sc. Hyg. Aliment., 35, 116-122, 1947.

14. Ed., Association of Vitamin Chemists Inc., Methods of Vitamin Assay, 2nd ed., Interscience, New York, 107-127, 1951.

15. Enders, R.K., Reproduction in the Mink (Mustela Vision), Proc. American Philosophical Soc., 96, 692-755, 1952.

16. Goulden, C.H., Methods of Statistical Analysis, John Wiley and Sons, New York, 1939.

17. Gunn, C.K., A Study of Whale Meat as a Substitute for Horse Meat in Mink Growing and Furring Rations, Fur Trade J. of Canada, March, 1956.

18. Hanning, F., Rice, L.M., Shands, J.A., Batterman, W., and Bray, R., Antioxidants in the Home Preservation of Foods, J. Home Economics, 45, 660, 1953.

19. Hartman, L., and White, M.D.L., The Significance of Peroxide Determination in Fats by the Dichlorophenol-Indophenol Method, J. Sci. Food Agric., 3, 112, 1952.

20. Hogl, Von. O., and Wenger, F., Antioxydantien in Fetten und Olen, Lebensmitteluntersuchung and Hygiene, 45,335-363, 1954.

21. Howell, R.E., and Gunn, W.S., Protein in the Diet of Mink, National Fur News, 27, No. 6, 1955.

22. Kandutsch, A.A., and Baumann, C.A., Factors Affecting the Stability of Thiamine in a Typical Laboratory Diet, J. Nutrition, 49, 209, 1953.

23. Kansky, L.L., The Influence of the Winter Plane of Nutrition on the Growth Rate and Subsequent Parturient Behaviour of Beef Heifers, Master's Thesis, The Library, University of British Columbia, 1955.

24. Kifer, P.E., and Schaible, P.J., Dry Diets for Ranch Raised Mink 1. Suitability for Late Growth and Maintenance, Michigan Quarterly Bulletin, 37, 592-598, 1955.

25. Kifer, P.E., and Schaible, P.J., Dry Diets for Ranch Raised Mink 11. Effect on Breeding, Whelping and Early Growth of Kits, Michigan Quarterly Bulletin, 38, 129-138, 1955.

26. Klocke, J.F., Tack, P., Ohlson, M., Nitchals, R., Leffler, E., and Henry, N., Nutritive Value of Fish From Michigan Waters. 11. Thiamine of Lake Herring, Carp, Common Sucker, Burbot and Smelt, Food Research, 12, 36-43, 1947.

27. Koch, R.B., Mechanisms of Fat Oxidation, Bakers Digest, April, 1956.

28. Kraybill, H.R., and Dugan, L.R., Antioxidants: New Developments for Food Use, J. Ag. Food Chem., 2, 81-84, 1954.

29. Lang, J.B., and Shaw, J.N., Chastek Paralysis Produced in Oregon Mink and Foxes by Feeding Fresh Frozen Smelt, North Amer. Vet., 24, 234-237, 1943.

30. Lea , C.H., Recent Developments in the Study of the Oxidative Deterioration of Lipids, Chem. and Indust., 1303, 1953.

31. Lea, C.H., Methods for Determination of Lipids, J. Sci. Food Agric., 3, 1952.

32. Lea, C.H., Rancidity in Edible Fats, Food Investigation Special Report No. 46, London, 1938.

33. Lee, F.L., Brooks, R.F., Pearson, A.M., Miller, J.J., and Wanderstock, J.J., Effect of Rate of Freezing on Pork Quality, American Dietetic Assn. J., 30, 351, 1954.

34. Lehman, A.J., Fritzhugh, O.G., Nelson, A.A., and Woodland, G., The Pharmacological Evaluation of Antioxidants, Advances in Food Research, 3, 197-208, 1951.

35. Lehrer, W.P., Wiese, A.C., Harvey, W.R., and Moore, P.R., The Stability of Thiamine, Riboflavin and Nicotinic Acid of Lamb Chops During Frozen Storage and Subsequent Cooking, Food Research, 17, 24, 1952.

36. Lehrer, W.P., Wiese, A.C., Harvey, W.R., and Moore, P.R., Effect of Frozen Storage and Subsequent Cooking on the Thiamine, Riboflavin and Nictonic Acid Content of Pork Chops, Food Research, 16, 485, 1951.

37. Leoschke, W.L., Madison, Wisconsin, Personal Communication, 1956.

38. Leoschke, W.L., Nutrition of the Mink, Transactions Am. Assoc. Cereal Chem., 11, 212-216, 1953.

39. Lieck, H., and Agren, G., On the Occurrence of a Thiamine Inactivating Factor In Some Species of Swedish Fish, Acta Physiologica Scandinavica, 8, 203-214, 1944.

40. Lips, A., Chapman, R.A., and McFarlane W.S., The Application of the Ferric Thiocyanate Method to the Determination of Incipit Rancidity in Fats and Oils, Oil and Soap, 20, 240, 1943.

41. Lundberg, W.O., A Survey of Present Knowledge, Researches and Practices in the United States Concerning the Stabilization of Fats, The Hormel Institute of the University of Minnesota, Publication No. 20, 1947.

42. Madramootoo, H., Studies in Mink Nutrition, Undergraduate Thesis, Dept. of Animal Husbandry, University of British Columbia, 1952.

43. Mapson, L.W., Effect of Processing on the Vitamin Content of Foods, British Med. Bull., 12, 73-77, 1956.

44. Maynard, L.A., Animal Nutrition, 3rd, ed., McGraw Hill, New York, 1951.

45. McCay, C.M., Nutrition of the Dog, 2nd ed., Comstock, New York, 1949.

46. Methods of Analysis, 7th ed., Association of Official Agricultural Chemists, Washington, 1950.

47. Miller, D.S., and Bender, A.E., The Determination of the Net Utilization of Proteins by a Shortened Method, Brit. J. Nutr., 9, 382-388, 1955.

48. Morrison, F.B., Feeds and Feeding, 12th ed., Morrison Publishing Co., New York, 1946.

49. Nutrient Requirements for Domestic Animals No. VII., Nutrient Requirements for Foxes and Minks, National Research Council, Washington, 1953.

50. Patton, S., and Kurtz, G.W., 2-Thiobarbituric Acid as a Reagent for Detecting Milk Fat Oxidation, J. Dairy Sci., 34, 669, 1951.

51. Poder, N., Recovery of Solvents Used in the Chemical Determination of Thiamine, Ind. Eng. Chem.-Anal. Ed., 15, 25, 1943.

52. Robertson, M.C., Master's Thesis, The Library, University of British Columbia, 1957.

53. Sebrell, W.H., and Harris, R.S., ed., The Vitamins, Academic Press, New York, Vol. III, 1954.

54. Smith, P.C., and Proutt, L.M., Development of Thiamine Deficiency in the Cat on a Diet of Raw Fish, Proc. Soc. Expt. Bio. and Med., 56, 1-3, 1944.

55. Soodak, M., and Cerecedo, L.R., Studies on Oxythiamine, J. Am. Chem. Soc. 66, 1998-1999, 1944.

56. Spector, H., Peterson, M.S., and Friedman, T.E., ed., Methods for Evaluation of Nutritional Adequacy, Advisory Board on Quartermaster Research and Development, Dept. of the Army Office of the Quartermaster General, Washington, Dec. 1954.

57. Spector, H., and Peterson, M.S., ed., Nutrition Under Climatic Stress, Advisory Board on Quartermaster Research and Development Committee of Foods, National Research Council, Washington, May, 1954.

58. Stephenson, A.B., Preliminary Studies on Growth, Nutrition and Blood Chemistry of Beavers, Master's Thesis, The Library, University of British Columbia, 1956.

59. Sumner, J.B., and Myrback, K., The Enzymes, Academic Press, New York, Vol. I, Part 2, 1186-1206, 1951.

60. The National Provisioner, Research Throws New Light on Fats as Animal Feeds Supplement, May 23, 1953.

61. Thiamine Losses and Salt Mixtures, Nutrition Revs., 13, 14, 1955.

62. Westock, S., Making Whale Meat into Mink Food, National Fur News, 27, No. 3, 1955.

63. Wood, A.J., Extension Research Memo No. 13, Canada Mink Breeders, 1957.

64. Worthen, L.P., A Study of the Factors Affecting Thiamine Inactivation, B.S.A. Thesis, Dept. of Animal Husbandry, University of British Columbia, 1953.

65. Worthen, L.P., A Study of Wheat-Herring Meal and Chastek Paralysis, Report, Animal Nutrition Laboratory, University of British Columbia, 1952.

66. Yudkin, W.H., Thiaminase, The Chastek-Paralysis Factor, Physiol. Revs., 29, 389-402, 1949.