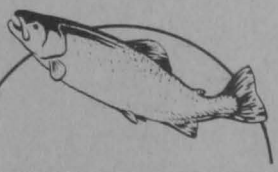
NUTRITIVE VALUE OF POLLOCK FISH SCALES AS DETERMINED BY RAT FEEDING TESTS



SPECIAL SCIENTIFIC REPORT–FISHERIES No. 260

UNITED STATES DEPARTMENT OF THE INTERI
FISH AND WILDLIFE SERVI

EXPLANATORY NOTE

The series embodies results of investigations, usually of restricted scope, intended to aid or direct management or utilization practices and as guides for administrative or legislative action. It is issued in limited quantities for official use of Federal, State or cooperating agencies and in processed form for economy and to avoid delay in publication.

United States Department of the Interior, Fred A. Seaton, Secretary

Fish and Wildlife Service, Arnie J. Suomela, Commissioner

NUTRITIVE VALUE OF POLLOCK FISH SCALES AS DETERMINED BY RAT FEEDING TESTS

by

Donald G. Snyder
Biochemist

and

Hugo W. Nilson
Supervisory Chemist

Special Scientific Report--Fisheries No. 260

The Library of Congress has cataloged this publication as follows:

Snyder, Donald Graeff, 1926–

Nutritive value of pollock fish scales as determined by rat feeding tests, by Donald G. Snyder and Hugo W. Nilson. [Washington, U. S. Dept. of the Interior, Fish and Wildlife Service, 1959]

11 p. diagr. 27 cm. (U. S. Fish and Wildlife Service. Special scientific report: fisheries no. 260)

Bibliography: p. 9.

1. Scales (Fishes) I. Nilson, Hugo Waldemar, 1901– joint author. II. U. S. Fish and Wildlife Service. III. Title. (Series)

SH11.A335 no. 260 636.087 59–60216

Library of Congress

The Fish and Wildlife Service series, Special Scientific Report--Fisheries, is cataloged as follows:

U. S. *Fish and Wildlife Service.*

Special scientific report: fisheries. no. 1–

[Washington] 1949–

no. illus., maps, diagrs. 27 cm.

Supersedes in part the Service's Special scientific report.

1. Fisheries—Research.

SH11.A335 639.2072 59–60217

Library of Congress

ABSTRACT

Rat feeding studies have indicated that pollock fish scale protein is as well digested but about 30 percent less assimilated than a protein supplement consisting of 3 parts casein and 1 part lactalbumin. Fish scales as the only source of 9 percent protein in the diet are incapable of supporting growth of young rats, but the scales can be utilized as a limited source of protein when supplemented with casein-lactalbumin protein. Increased utilization of scale protein in combination with stepwise higher levels of casein-lactalbumin indicate that no toxic substances per se are present in scales for growing rats.

CONTENTS

	Page
Introduction	1
Material and analysis	1
Experimental	2
Results and interpretations	3
Discussion	6
Summary and conclusions	7
Acknowledgements	8
Literature cited	9
Table 1	10
Table 2	11
Figure 1	13

INTRODUCTION

During the past few years, the fishing industry has been confronted with the increasingly difficult problem of annually disposing of thousands of tons of fish scales, as fillets have replaced unprocessed fresh fish in sales volume. Harbors nearby to plants normally allow for an inexpensive area of scale disposal, but unless tidal flows are strong, pollution may result.

Attempts to use the scales as fertilizer on local farms have been unsuccessful, and the incineration of scales would be costly. Chemical engineers and pharmaceutical houses have been asked for suggestions for utilizing the scales, but this approach, too, has been unsuccessful. In order to determine whether the scales may have value as a source of protein in farm-animal diets, the nutritive value and some factors affecting the nutritive value of the nitrogen compounds in whole scales are being studied.

Part of this work--rat-feeding studies to compare the nutritive values for growth, the biological values for maintenance, and the digestibilities of pollock fish scale and of casein-lactalbumin protein--is reported herein. The nitrogen content of pollock scales is equivalent to about 60 percent crude protein (N x 6.25).

Very little research has been conducted with fish scales in the past, and none has been conducted that would promise a solution to the proposed problem. For many years, however, investigators have been studying the nutritive value of similar biological materials. Meunier et al. (1927), Routh and Lewis (1938), and Routh (1942a, 1942b) have investigated the nutritive possibilities of wool, and Wagner and Elvehjem (1942, 1943) and Newell and Elvehjem (1947) have investigated the nutritive quality of some waste keratins. The nutritive values of meal from untreated and treated chicken feathers have been studied by Routh (1942b), Binkley and Vasak (1950), Wilder et al. (1955), Feedstuffs (1956), as well as others.

In general, these studies have indicated that scleroproteins are nutritionally inadequate but can be used as a partial source of protein with proper supplementation. As far as is known, no studies have been conducted to determine the feed value of fish scales, the scleroprotein now being considered.

MATERIAL AND ANALYSIS

The fish scales used in this study were kindly furnished by the staff of the Fishery Technological Laboratory, U. S. Bureau of Commercial Fisheries, East Boston, Massachusetts. Pollock (Pollachius virens) were scaled by hand. The scales were washed with water, drained, spread in pans, and dried in an oven at 100° C. They were shipped to this laboratory, where they were ground as finely as possible

in a coffee grinder. An analysis of the particle size of a typical batch of ground scales showed that 3 percent were caught on a 20-mesh U. S. Standard screen, 19 percent on a 40-mesh, 36 percent on a 60-mesh, 18 percent on a 80-mesh, and 7 percent on a 100-mesh, and that 17 percent passed through a 100-mesh screen.

The moisture, protein (N x 6.25), fat, and ash of representative samples of scales from the various lots showed no great variation. The mean and ranges of moisture content of 4 lots of scales were 4.4 percent and 1.0-6.9 percent, respectively; of protein content of 11 lots, 59.5 percent and 56.6-62.5 percent; of fat content of 4 lots, 0.004 percent and 0.0-1.0 percent; and of ash content of 5 lots, 38.9 percent and 36.1-43.1 percent. Association of Official Agricultural Chemists (1955) methods of analyses were used.

No significant differences in the nutritive value of the scale protein were found among lots. The small differences in moisture, protein, fat, and ash contents of the scales probably can be attributed to the nutritional status and age of the fish from which the scales were collected (1954).

EXPERIMENTAL

Postweaning rats were fed diets containing a total of 9 percent protein from pollock fish scales (PFS) or a protein supplement of 3 parts casein and 1 part lactalbumin (CL)in stepwise substitution, on an equal nitrogen basis, of the PFS protein, namely, 2.25, 4.50, and 6.75 percent. In addition, the CL was also fed alone at these three levels of protein, as well as at the 9 percent level.

The basal diet had the following composition: lard, 8; cod-liver oil, 2; salt mixture U.S.P. XIV, No. 2, for vitamin-A bioassay, 4; and dextrin, 86 parts by weight. To every 100 g. of the basal diet were added 100 mg. of choline chloride, 15 mg. of alpha tocopherol, and 75 mg. of the following vitamin mixture: thiamine HCl, 1; riboflavin, 2; pyridoxine, 1; Ca pantothenate, 10; nicotinamide, 10; inositol, 5; para-aminobenzoic acid, 30; biotin, 0.05; folic acid, 0.2; menadione, 14.2; and ascorbic acid, 2 parts by weight. Vitamin B_{12} was supplied to the rats by adding 0.2 ml. of a 0.005 percent aqueous solution to individual 250 ml. water bottles every time they were refilled. The ground PFS and CL were incorporated into this basal diet at the expense of dextrin.

Four highly inbred black-hooded rats, two males and two females, were allotted to each group. The males were allotted at initial live weights of 49 to 53 and the females at 48 to 50 g. Litter-mates were randomly distributed among the various groups, but never more than one to any single group.

The rats were housed individually in wire-screen cages fitted on wire-mesh floors. The temperature of the room was maintained at 80° F. The rats were supplied with food and water ad libitum, and weekly records were taken of live weight and food consumption. The feeding study lasted 10 weeks. The apparent digestibilities of the protein consumed by individual rats were determined in aliquots of the ground, debris-free feces, collected during the fourth and fifth week of the feeding test.

Four adult male albino rats, two each fed PFS and CL protein, were used to determine the biological value for maintenance, according to the method proposed by Mitchell (1924). The rats were housed in cages on wire screens, over funnels into which a small wire screen was inserted to catch feces and allow urine to pass. Urine was collected in 600 ml. beakers under toluene, and a few ml. of a 3-percent solution of H_2SO_4 was used as additional preservative. The feces were collected from the small screens and separated from food, hair, and other debris, and were ground. Nitrogen was determined in measured aliquots of the urine and feces.

The non-nitrogenous diet fed to the rats contained: dextrin, 60; sucrose, 21; salt mixture U.S.P. XIV, No. 2 for vitamin-A bioassay, 4; lard, 13; and cod-liver oil, 2 parts by weight. To each 100 g. of this basal diet was added 0.072 and 0.240 mg. of thiamine and riboflavin, respectively. The test protein was incorporated into a 4-percent agar - 10-percent sucrose gel mixture. A 3-day precollection and a 2-day collection nonprotein-feeding period; a 3-day precollection and a 3-day collection protein-feeding period; followed by a second 3-day precollection and a 2-day collection nonprotein-feeding period was used. The quantity of test protein fed daily during the protein-feeding period was equivalent to the nitrogen contained in the urine daily during the first collection period.

RESULTS AND INTERPRETATIONS

The data in table 1 and figure 1 indicate that the male and female rats fed the diet containing 9 percent protein from PFS alone lost considerable weight and died before the eighth week of the study. The rats fed the basal diet with no added protein lost about the same weight and died at about the same time. Evidently 9 percent PFS protein did not permit growth any better than no protein added to the basal diets.

The mean gain of 135.9 g. of the group of rats fed the diet containing 9 percent CL protein, indicates that CL was more efficiently utilized than the various other 9 percent protein combinations of PFS-CL contained in the diets (table 1). The mean gain of the group of rats fed this diet at the end of 10 weeks, however, was not statistically significantly different ($p \geq 0.05$) from that of the group fed the diet in which 2.25 percent PFS protein replaced a like amount of CL protein,

namely, 117.5 g. Therefore, inclusions of up to 25 percent protein from PFS in place of equal amounts of CL, as the sole source of protein, and at a total level of dietary protein of 9 percent, does not adversely affect the nutritive value of the protein for growing rats. It is interesting to note that the males grew better than the females when fed the diet containing 9 percent CL protein, which would be expected, but grew similarly when fed the diet containing 2.25 percent PFS and 6.75 percent CL protein, which would not be expected, since the mean gains of the two groups were not statistically significantly different. This observation might indicate that the males and females differ in their ability to utilize the PFS protein in this combination with CL protein.

The mean weights at 10 weeks of the group of rats fed the diets containing 6.75 percent PFS and 2.25 percent CL protein (15.3 g.) and 4.50 percent CL protein alone (23.8 g.) also were not statistically significantly different ($p \Rightarrow 0.05$). All of the mean weights of the groups of rats fed the other diets were statistically significantly different ($p \leq 0.01$) for the 10-week period. The coefficients of variation generally were smaller than usual for this type of a feeding study (9 - 12 percent).

Although the group of rats fed the diet containing 9 percent PFS protein alone died, the different groups of rats grew increasingly better when they were fed diets in which 2.25, 4.50, and 6.75 percent of this PFS protein was replaced by equal amounts of CL protein. This increased growth was apparently directly correlated with the stepwise higher levels of CL protein in relation to PFS protein in the diet. However, the rats fed diets containing these increasing levels of CL protein alone in the diet did not grow as well as rats fed these same diets containing, in addition, the decreasing levels of PFS protein to make a total of 9 percent protein. This added growth with PFS protein indicates that the PFS protein certainly was being utilized.

An increase of 31 g. in mean gain was obtained when the group of rats was fed the diet containing 6.75 percent PFS and 2.25 percent CL protein over that obtained for the group fed the diet containing 2.25 percent protein from CL alone. This increased growth cannot be the result of the added PFS protein or the low level of CL protein, since rats fed diets containing these levels alone did not gain weight. Apparently, the 2.25-percent level of CL protein was ample to balance the 6.75 percent PFS protein.

An increase of 73 g. in mean gain was obtained when the group of rats was fed the diet containing 4.50 percent PFS and 4.50 percent CL protein over that obtained from the group fed the diet containing 4.50 percent CL protein alone. This greater growth of rats fed a diet containing an even lower level of PFS protein suggests that the PFS

protein was more completely balanced by the additional CL. In this case, CL protein could be used for growth when fed alone in the diet at this low level and the PFS protein might be envisioned as contributing additional nutrients.

An increase of 34 g. in mean gain was obtained when the group of rats was fed the diet containing 2.25 percent PFS and 6.75 percent CL protein over that obtained for the group fed the diet containing 6.75 percent CL protein alone. This increased growth of 34 g. is not as great as when the previous pair of diets are compared, namely, 73 g. This result is to be expected, inasmuch as there was less PFS protein in the diet and the nutritively superior CL protein was present in sufficient quantity to permit greater utilization. It is interesting to note that there was a relation of nearly 1:2:1 (31, 73, 34 g.) for the increased growth when the three pairs of diets were fed. This might indicate that the balancing values of the two proteins vary when different levels of each were included in the diets.

The data in table 1 indicate, in general, that as the rats grew better less protein was required per unit gain in weight. This decreasing requirement for protein suggests that increasing amounts of CL protein permit the rats to utilize the PFS protein more efficiently. When this index, as well as the previously compared growth rates, is used as the criteria for PFS protein utilization, trends of data indicate that no toxic substances per se are present in the fish scales.

The data in table 2 indicate that the mean apparent digestibility of PFS protein was about 80 percent when fed at the 9-percent level in the diet. The data also indicate that this level of digestibility was not statistically significantly increased ($p \Rightarrow 0.05$) when the rats were fed diets in which the CL protein was substituted in part or in whole for 9 percent PFS.

The increase in digestibility of diets containing the smaller to greater levels of CL protein alone is interesting. It may be that as the rats ingested the stepwise higher levels of food nitrogen, the residual metabolic products eliminated in the feces, as well as the digestibility of the CL protein remained equal. Hence, the ratios of ingested food nitrogen divided by the unabsorbed food nitrogen plus residual metabolic nitrogen would increase. There would be an error in calculating the digestibility of the protein for rats fed a diet containing a small amount of protein which would restrict food intake and limit growth compared with those obtained for rats fed a diet containing an adequate level of protein which would result in increased food intake.and greater growth.

Biological values for maintenance of 61.2 and 61.1 were gotten for PFS protein with two sets and, similarly, 85.3 and 89.4 were gotten for CL protein. The CL protein appears to be utilized about 30 percent

more than PFS protein. True digestibilities determined in this test indicated that both PFS and CL protein were completely digested. Whereas the apparent and true digestibility of the two proteins by rats are similar, quite different assimilation is indicated by the biological values.

DISCUSSION

Investigators studying the nutritive value of other waste scleroproteins have concluded that they are nutritionally inadequate, but can be used as a partial source of protein with proper supplementation. This conclusion is also true with pollock fish scale protein.

Rats fed a diet containing 9 percent protein from PFS alone lost considerable weight and died in about 8 weeks. Rats fed diets containing PFS in combination with stepwise higher levels of CL protein, for a total dietary protein level of 9 percent, however, utilized the diet for growth increasingly better. These rats also grew better than those fed the stepwise higher levels of CL protein alone in the diet. In general, as the rats grew better they utilized the food for growth more efficiently.

Routh (1942a, 1942b), Wagner and Elvehjem (1942, 1943), Newell and Elvehjem (1947), Wilder et al. (1955), and others showed that the various nutritively deficient scleroproteins could be utilized or balanced with proper supplementation, but in general they did not indicate that this utilization or balance could be improved when greater levels were included in the diets. In most cases the supplementations consisted only of empirical amounts of amino acids added to the diet in order to find out which acids alone or in combination permitted better utilization of the scleroprotein.

The rats fed the diet containing a 9-percent level of protein from CL alone grew better than rats fed any other 9 percent protein combination of PFS and CL protein. At this level of protein in the diet, at least, CL protein must be better balanced in nitrogen nutrients than the protein from any of the combinations. Thus, the growth of rats fed diets containing increasing levels of PFS protein in relation to fixed levels of CL protein was less and less. Toxic factors, per se, in the scales must be ruled out, because there then would be no variation in the utilization of PFS protein by the rats. Furthermore, there were no visible symptoms of toxicosis.

The protein of the fish scales is digested sufficiently well so this cannot be an important factor in explaining the effects noted. The results showed that, at the 9-percent level in the diet, both PFS protein and CL protein were about 80 percent digested when fed to rats. This high level of digestion indicates that the incomplete, probably imbalanced, PFS protein had been absorbed to a considerable extent and was available for metabolism. The utilization after

absorption, as shown by the biological values, is quite dissimilar, however. This difference in utilization may suggest that better assimilation of this protein, that is utilization, is responsible, at least in part, for the greater nutritive value of PFS in combination with the stepwise higher levels of CL protein. Apparently the PFS protein is more completely utilized when the higher levels of CL are fed because the protein furnished by CL supplies more and more of the lacking amino acids or other nitrogen compounds.

The problem remains as to the value of more commonly available sources of protein to supplement PFS protein as feed, and the ability of other species of animals to metabolize this protein.

SUMMARY AND CONCLUSIONS

Postweaning rats were fed diets containing pollock fish scales (PFS) or a protein supplement of 3 parts casein and 1 part lactalbumin (CL). The latter also was fed at three levels of protein in stepwise substitution of, as well as in place of, the pollock scales. Apparent digestibilities of protein by individual rats were determined in diets fed during this feeding study. The biological value for maintenance and true digestibility was determined for PFS and CL protein.

The data indicate:

1. The mean apparent digestibility is 80 percent when PFS protein is fed at a 9-percent level in the diet to male and female rats. This value of digestibility is not significantly increased ($p \Rightarrow 0.05$) when rats are fed diets in which the CL protein is substituted in part or in whole for the 9 percent PFS. PFS and CL protein is completely digested, as indicated by true digestibility values, when only enough is fed to equal metabolic nitrogen.

2. A level of 9 percent PFS protein as the only source of protein in an otherwise adequate diet is incapable of supporting growth in young rats. This nutritional inadequacy of PFS protein is likely due to a deficiency and/or imbalance of specific nitrogen nutrients.

3. PFS protein can be utilized by rats as a limited source of protein for growth when supplemented with CL protein in the diet. Decreasing ratios of PFS to CL protein in the diet permit progressively better utilization of the PFS protein.

4. No toxic substances *per se* are present in pollock fish scales for growing rats.

5. The biological value of PFS protein for maintaining rats is about 60 percent; which is about 30 percent less than for CL protein.

ACKNOWLEDGEMENTS

The authors wish to thank Mr. Robert Kifer for his assistance in caring for the animals, and Mrs. Sue Nealis for her assistance in the statistical analysis of the data.

ASSOCIATION OF OFFICIAL AGRICULTURAL CHEMISTS
1955. Official methods of analysis, 8th ed., Washington, D. C.

BINKLEY, CHARLES H., and OTTO R. VASAK
1950. Production of a friable meal from feathers. AIC - 274, Bureau of Agricultural and Ind. Chem., Agr. Res. Admin., U.S.D.A.

FEEDSTUFFS
1956. Feathermeal used as a source of protein for lambs, vol. 28, p. 21.

MEUNIER, L., P. CHAMBARD, and H. COMTE
1927. Sur la digestion pancreatique de la laine. Compt. rend. acad., vol. 184, pp. 1208-1210.

MITCHELL, H. H.
1924. A method of determining the biological value of a protein. J. Biol. Chem., vol. 58, pp. 873-903.

NEWELL, G. W., and C. A. ELVEHJEM
1947. Nutritive value of keratin. III. Effect of source, particle size, and method of grinding. J. Nutrition, vol. 33, pp. 673-683.

NISHIHARA, HATASU
1954. Calcium oxalate content of fish scales. Science Rept., Saitama University Ser. B, vol. I, pp. 159-164.

ROUTH, JOSEPH I.
1940. Chemical studies on powdered wool. J. Biol. Chem., vol. 135, pp. 175-181.

ROUTH, JOSEPH I.
1942a. Nutritional studies on powdered wool. J. Nutrition, vol. 23, pp. 125-130.

ROUTH, JOSEPH I.
1942b. Nutritional studies on powdered chicken feathers. J. Nutrition, vol. 24, pp. 399-404.

ROUTH, JOSEPH I., and HOWARD B. LEWIS
1938. The enzymatic digestion of wool. J. Biol. Chem., vol. 124, pp. 725-732.

WAGNER, JOSEPH R., and C. A. ELVEHJEM
1942. Nutritive value of keratins. I. Powdered swine hoofs. Proc. Soc. Exp. Biol. Med., vol. 51, pp. 394-396.

WAGNER, JOSEPH R., and C. A. ELVEHJEM
1943. Nutritive value of keratins. II. Powdered swine hoofs in poultry rations. Poultry Science, vol. 22, pp. 275-276.

WILDER, O. H. M., PAUL C. OSTBY, and BARBARA R. GREGORY
1955. The use of chicken feather meal in feeds. Poultry Science, vol. 34, pp. 518-524.

Table 1

Mean grams gain in weight, mean grams of diet consumed, and mean grams gain per gram of protein ingested for rats fed diets containing pollock fish scale protein and casein-lactalbumin protein

Diet designation	Mean gain in weight in grams			S.E.[1/]	Mean grams of diet consumed			Mean grams gain per gram of protein ingested		
	male	female	male and female		male	female	male and female	male	female	male and female
PFS, 9.00%	-17.5	-15.0	-16.3	0.50	111.0	109.0	110.0	-1.76	-1.51	-1.64
PFS, 6.75%-CL, 2.25%	22.5	8.0	15.3	5.38	347.0	292.0	322.0	0.72	0.29	0.51
PFS, 4.50%-CL, 4.50%	98.5	94.5	96.5	3.52	563.5	595.5	579.5	1.95	1.76	1.86
PFS, 2.25%-CL, 6.75%	119.5	115.5	117.5	2.86	530.5	545.5	538.0	2.50	2.36	2.43
CL, 9.00%	152.7	121.5	135.9	9.17	654.3	592.8	619.1	2.59	2.27	2.40
PFS, 0.00%-CL, 0.00%	-20.5	-22.5	-21.5	1.73	96.0	104.0	100.0	-	-	-
CL, 2.25%	-15.0	-15.5	-15.3	2.69	180.0	166.0	173.0	-3.88	-4.37	-4.13
CL, 4.50%	21.5	26.0	23.8	1.65	308.5	314.0	311.3	1.56	1.84	1.70
CL, 6.75%	73.5	74.0	73.8	5.20	448.0	432.5	440.3	2.41	2.53	2.47

[1/] Standard error of the mean for gain in weight at the concluding week of the feeding study.

Table 2

Mean apparent digestibility of the proteins in the diets fed to rats to compare pollock fish scale and casein-lactalbumin protein

Diet designation	Mean % apparent digestibility				
	males	females	males and females	S.E.1/	C.V.2/
PFS, 9.00%	83.7	75.3	79.5	2.82	7
PFS, 6.75%-CL, 2.25%	76.3	76.8	76.5	1.34	4
PFS, 4.50%-CL, 4.50%	80.6	80.8	80.6	1.25	3
PFS, 2.25%-CL, 6.75%	79.7	81.3	80.4	1.50	4
CL, 9.00%	84.3	83.1	83.8	0.89	3
PFS, 0.00%-CL, 0.00%	-	-	-	-	-
CL, 2.25%	62.9	47.4	55.2	5.56	20
CL, 4.50%	70.6	75.8	73.2	1.55	4
CL, 6.75%	80.0	78.6	79.2	4.16	11

1/ Standard error of the mean for gain in weight for the group of male and female rats.
2/ Coefficient of variation in % for the group of male and female rats.

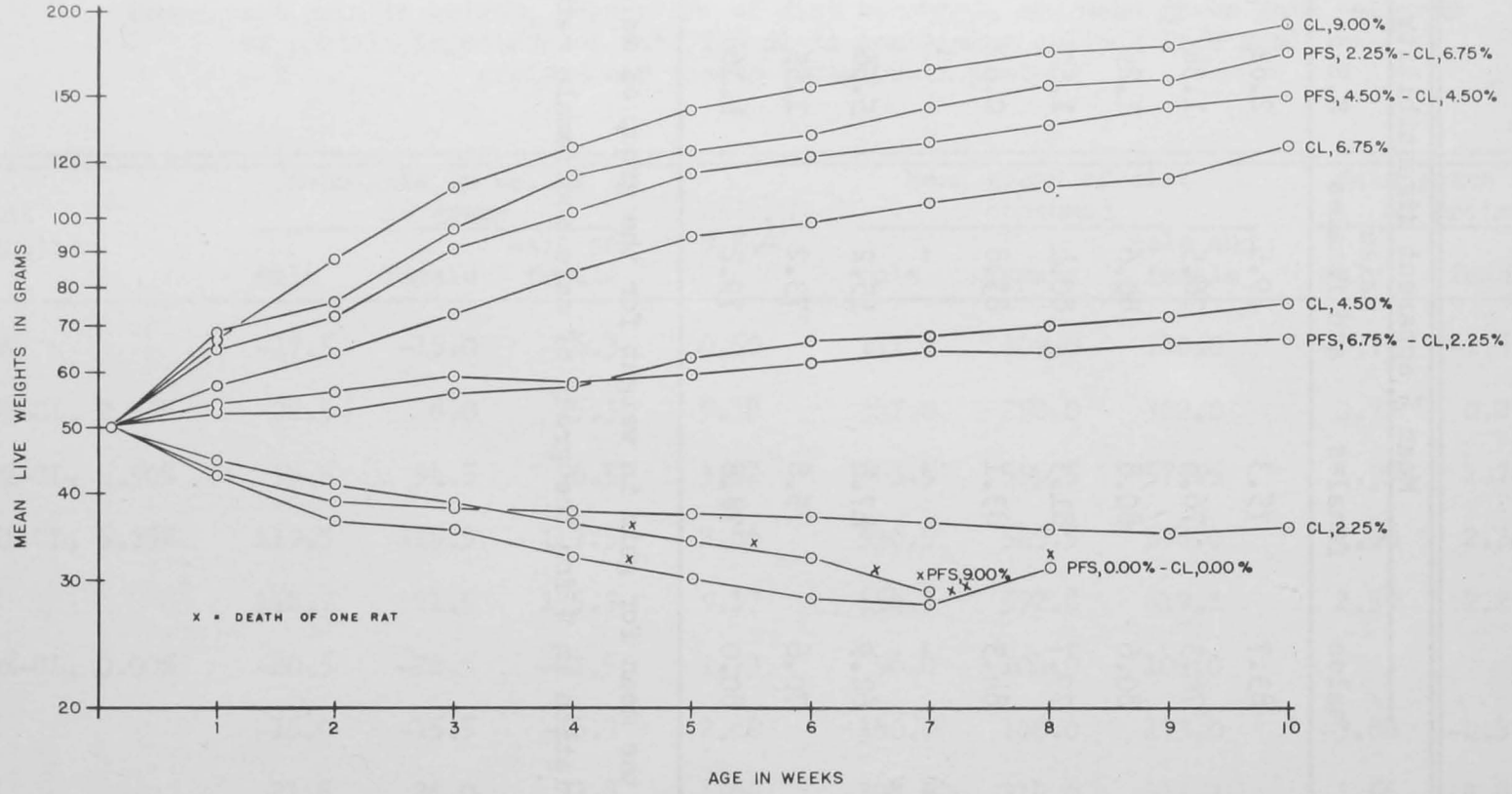


FIGURE 1

MEAN WEEKLY WEIGHTS OF GROUPS OF RATS FED DIETS
TO COMPARE POLLACK FISH SCALES AND CASEIN-LACTALBUMIN PROTEIN