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Studies on Edible Yeasts¹

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I. Food Yeast, Brewers' Yeast, and Skimmed Milk as Supplements to a Deficient Diet

PREVIOUS investigations carried out in this laboratory² point to the fact that the diets consumed by the low-income level groups among the Puerto Rican population are always deficient, more or less, in several nutritional factors, principally vitamin A, essential amino acids, calcium, and, possibly, some of the B vitamins. With the intention of alleviating this situation somewhat, several investigators³ studied the possibility of correcting such deficiencies by supplementing the diet with cheap and, at the same time, easily available foodstuffs, such as soybeans, chickpeas, and skimmed milk.

The present study is an attempt to determine the nature and extent of the supplementary action exhibited by dried food yeast (*Torula utilis* No. 3), brewers' dried yeast,⁴ and dried skimmed milk, when these foods are incorporated into a modified Puerto Rican rural diet in amounts corresponding to 25 percent of its total protein content.

EXPERIMENTAL METHODS

Diets. The basic diet utilized in this investigation was devised in accordance with the data collected by Descartes, Díaz Pacheco, and Noguerras⁵ on the food consumption habits of Puerto Rican rural families. Its composition was as follows: rice, 37.5 percent; white flour, 5 percent; corn-meal, 5 percent; yellow sweet potatoes, 10 percent; plantains, 6.3 percent; tanniers, 6.2 percent; red kidney beans, 10 percent; sugar, 12.5 percent; and codfish, 7.5 percent.

As the main interest of the present investigation was to study the

1. Received for publication January 13, 1948.

2. D. H. Cook and T. Rivera, Rice and beans as an adequate diet. Porto Rico Rev. Pub. Health & Trop. Med., 5:3-7, 1929.

D. H. Cook, J. H. Axtmayer, and L. M. Dalmau, A comparative study of the nutritive value of three diets of frequent use in Puerto Rico. Puerto Rico J. Pub. Health & Trop. Med., 16:3-13, 1940.

3. J. H. Axtmayer, Nutritional values of mixtures of polished rice, red kidney beans, chickpeas, and soybeans. Puerto Rico J. Pub. Health & Trop. Med. Med., 21:274-279, 1946.

4. Fleischmann, Type 2019.

5. S. L. Descartes, S. Díaz Pacheco, and J. R. Noguerras, Food consumption studies in Puerto Rico. Bull. 59, Agric. Exper. Sta., Río Piedras, 1941.

supplementary action of yeast on a protein- and vitamin B-deficient rural diet, it was necessary to rule out all other deficiencies, such as vitamin A and calcium. To supply vitamin A, a colored variety of sweet potatoes, very rich in carotene,⁶ was used instead of the white variety regularly consumed by local country folk; to furnish an adequate amount of minerals, 2 percent of a salt mixture⁷ was incorporated into the diet. Thus fortified with vitamin A and minerals, the rural diet will be referred to in this paper as the "modified rural diet." Its protein content was 11.72 percent on a wet basis. With this basic diet, seven different experimental rations were prepared, each of which contained about 11 percent protein:

Ration 1: The nitrogen in this ration was derived entirely from the modified rural diet without any supplement.

Ration 2: Seventy-five percent of the nitrogen in this ration came from the modified rural diet, while 25 percent was from dried *Torula* yeast.

Ration 3: Seventy-five percent of its nitrogen was derived from the modified rural diet with 25 percent from dried brewers' yeast.

Ration 4: Of the nitrogen in this ration, 75 percent came from the modified rural diet and 25 percent from dried skimmed milk.

Ration 5: This ration was exactly the same as Ration 1 as to nitrogen source, but synthetic B vitamins (thiamine, riboflavin, nicotinic acid, pyridoxine, and pantothenic acid) were added in an amount⁸ equal to that supplied by the brewers' yeast in Ration 3.

Ration 6: This was the same as Ration 1 with thiamine alone added as supplement in an amount equal to that supplied by the brewers' yeast in Ration 3.

Ration 7: This ration was the same as Ration 2 as to nitrogen source, but synthetic B vitamins (thiamine, riboflavin, nicotinic acid, pyridoxine, and pantothenic acid) were added in an amount equal to that supplied by the brewers' yeast in Ration 3.

Procedure. During seven weeks, groups of six twenty-eight-day-old male Wistar albino rats, weighing between 55 and 68 g., were fed on each of the rations described above; measurements were taken of the food intake, the weight of the feces passed, and of their nitrogen content, during the fourth and fifth weeks of the experiment. The rate of weight increase of the litter mates was also compared.

From the results obtained, the coefficient of apparent digestibility

6. 500 I. U. of vitamin A per gram.

7. P. B. Hawk and B. L. Oser, Modified Osborne Mendel salt mixture. *Science*, 74:369, 1931.

8. R. E. Johnson, The effect of a diet deficient in part of the vitamin B complex upon men doing manual labor. *J. Nutrition*, 24:585-596, 1942.

TABLE 1
Coefficient of Apparent Digestibility and Growth-promoting Value of the Protein in the Different Diets Given

Diet	Protein Content of Diet	No. of Rats	Mean Body Weight G.		Amount Per Rat—Average Weekly						Growth-promoting Value of Protein	Increase in Weight per G. of Food Eaten	
			Initial	Final	Weight Increase G.	Food Intake G.	Nitrogen Intake G.	Weight of Dry Feces G.	Nitrogen in Feces G.	Nitrogen absorbed G.			Coefficient of Apparent Digestibility
1. Modified rural diet	11.72	6	57	166	17.3	73.9	1.389	3.56	0.214	1.171	84.5	2.00	0.23
2. <i>Torula</i> yeast-supplemented diet	11.47	6	60	213	27.0	100.1	1.835	5.27	0.319	1.516	82.6	2.35	0.27
3. Brewers' yeast-supplemented diet	11.60	6	55	213	28.1	99.7	1.847	5.52	0.354	1.493	80.8	2.43	0.28
4. Dried skimmed milk-supplemented diet	11.41	6	56	195	25.5	87.2	1.589	4.20	0.262	1.327	83.5	2.57	0.29
5. B Vitamins-supplemented diet	11.38	3	68	215	26.3	101.0	1.836	5.64	0.343	1.493	81.3	2.29	0.26
6. Thiamine-supplemented diet	11.72	3	57	159	16.0	69.1	1.299	2.52	0.160	1.139	87.8	1.93	0.23
7. <i>Torula</i> yeast- and B vitamins-supplemented diet	11.56	6	55	213	27.6	98.1	1.815	4.41	0.295	1.520	83.7	2.43	0.28

of the protein in each diet, as well as the ratio of body weight increase to gram of protein eaten, was calculated. These results are shown in Table 1.

The animals were weighed once a week. The average total gain in weight per week for each of the seven groups is shown on Chart I.

RESULTS

At the end of the stipulated seven weeks' period, the rats receiving the modified rural diet (Ration 1) and the thiamine-supplemented diet (Ration 6) reached an average body weight of 166 and 159 g., respectively, while those fed the yeast supplemented diet and that supplemented by B vitamins (thiamine, riboflavin, nicotinic acid, pyridoxine, and pantothenic acid—Rations 2, 3, 5, and 7) attained an average body weight of 213, 213, 215, and 213 g., respectively. The animals on the skimmed milk-supplemented diet (Ration 4) were slightly heavier than those on either Rations 1 or 6, their body weight averaging 195 g. The rats receiving Rations 2, 3, 5, and 7 ate more of the food and made better use of it than those on the other rations, their increase in weight per gram of food eaten being 0.27, 0.28, 0.26, and 0.28 g., respectively. Those on Rations 1 and 6 had values of 0.23 for each of the two groups.

The nutritive or growth-promoting values of the protein in the diets, measured by the weight increase per gram of protein eaten, were found to be in the same corresponding order. Both yeast and the vitamin-supplemented rations gave slightly higher values than Ration 1, the figures being 2.35, 2.43, 2.29, 2.43, and 2, respectively. The value obtained for the skimmed milk-supplemented diet was the highest of all, 2.57. No significant differences were observed in the calculated coefficient of apparent digestibility for protein in the seven diets tested.

DISCUSSION

According to the results obtained, the addition of yeast in the proportion utilized in this study did not supplement, to any appreciable extent, the protein in the modified Puerto Rican rural diet. However, the study did point out that this diet was limited in some of the B-complex factors and that such deficiency could be easily corrected by the addition of small amounts of yeast.

The addition of dried skimmed milk in the level used showed a slight but definite improvement in the quality of the protein. Its growth-promoting value was the highest of all, even though the rate of growth of the animals was below that obtained with the yeast- and

vitamin-supplemented diets. This was due essentially to a lower food intake.

The addition of both dried *Torula* yeast and synthetic B vitamins (Ration 7) did not produce any marked beneficial effect on the growth-promoting value of the protein, when compared to the effect produced by adding any one of these supplements only.

Thiamine alone did not improve the nutritive value of the diet; this vitamin was apparently not a limiting factor. In an attempt to throw more light on this aspect, all diets were analyzed for thiamine and riboflavin. The thiamine content of the modified rural diet was found to be 1.24 μg . per gram. Considering that the animals fed on this diet consumed about 10 g. daily, this amount supplied enough thiamine for optimum growth, which explains why supplementing thiamine alone did not improve the nutritive value of said diet.

On the other hand, the riboflavin content of the diet was found to be 0.75 μg . per gram; that is, the daily intake of 10 g. of diet supplied only about 7.5 μg . of riboflavin. As the requirement of the rat for optimum growth is about 40 μg . daily, a definite deficiency of this vitamin was evident and suggested that riboflavin was at least one of the B-complex factors in which the rural diet was deficient.

The values for thiamine and riboflavin of the dried skimmed milk-supplemented diet were 1.50 and 1.53 μg . per gram, respectively. As the animals ate an average of 12 g. per day, this amount furnished enough thiamine for optimum growth but supplied somewhat less than half the riboflavin requirement.

As the animals on the yeast- and vitamins-supplemented rations consumed an average of 14 g. daily, they received an excess of the minimum requirement of thiamine and riboflavin necessary for optimum growth.

Thiamine and riboflavin contents of both the unsupplemented and supplemented rations are reported in Table 2. The thiamine was determined by the thiocrome method and the riboflavin, micro-biologically.

TABLE 2
Thiamine and Riboflavin Contents of Rations

<i>Ration</i>	<i>Thiamine</i> $\mu\text{g.}/\text{G.}$	<i>Riboflavin</i> $\mu\text{g.}/\text{G.}$
1. Rural diet (unsupplemented)	1.24	0.75
2. Rural diet + food yeast	2.49	2.85
3. Rural diet + brewers' yeast	2.44	4.17
4. Rural diet + dried skimmed milk	1.50	1.53
5. Rural diet + vitamins	2.21	3.53

SUMMARY

When 25 percent of the crude protein ($N \times 6.25$) of a Puerto Rican modified rural diet was replaced by an equivalent amount of crude protein of either food or brewers' yeasts, no appreciable improvement could be observed in the nutritive value of the former. However, the same amount of either one of these two yeasts corrected an evident deficiency in some of the factors belonging to the B group of vitamins.

So far as the rat requirements were concerned, the rural modified diet was found to be adequate in its thiamine content but deficient in riboflavin. As fed (air-dried), this diet contained 1.24 μg . per gram of thiamine and only 0.75 μg . of riboflavin.

When dried skimmed milk was used in the same proportion in place of the yeasts, a slight but definite improvement in the quality of the protein was noted.

II. Growth and Reproduction Performance of Rats Fed on Yeast as Sole Source of Protein

Dried yeast, as a food, functions in two very important ways. One is by supplying all known B vitamins and the other, by acting as a source of protein when utilized in the proper proportions.

It is the purpose of the writers to discuss the adequacy of yeast protein for the maintenance of normal growth and reproduction in the rat. The literature dealing with the subject has been not only scarce but also somewhat contradictory; for this reason, the authors have thought it well to review it here.

In 1919 Osborne and Mendel⁹ fed rats on brewers' yeast as the sole source of protein at levels of 30 and 40 percent. The animals were maintained on these diets for more than a year without exhibiting any gross abnormality, those on the 40 percent level growing at a normal rate. The rats fed the 30 percent level did not do so well. In 1923 Nelson, Heller, and Fulmer¹⁰ utilized diets containing from 25 to 50 percent of dried brewers' yeast and observed normal growth and reproduction in the animals receiving 45 percent level. Three generations of rats were reared on these diets, the growth of the off-

9. T. B. Osborne and L. B. Mendel, The nutritive value of yeast protein. *J. Biol. Chem.*, 38:223-227, 1919.

10. V. E. Nelson, V. G. Heller, and E. I. Fulmer, Studies on yeast. VII. The dietary properties of yeast. *J. Biol. Chem.*, 57:415-424, 1923.