

FORTIFIED FOODS FOR VIETNAMESE BABIES

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Do - Quang - Oanh

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FORTIFIED FOODS FOR VIETNAMESE BABIES

By

Do-Quang-Oanh

AN ABSTRACT OF A THESIS

Submitted to
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ABSTRACT

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The vietnamese baby diet mainly consists of rice gruel, poor in nutritive value. Nutritional diseases in early childhood are widespread and high infant mortality is the result. It is important to raise the nutritional quality of the baby's food to make optimal development possible.

Several enriched diets, based on rice, were formulated from other native products such as soy, pork, fish, and egg to bring the total protein level up to 15%. Other supplements included sweet potato as a source of vitamin A, yeast as a source of vitamin B complex, and calcium carbonate.

The test diets, as formulated, were a dry flour-like product. In animal tests they were fed dry, but for human consumption, water would be added and the mixture cooked into a gruel similar to the native product.

Weanling rats were maintained on the experimental diets for 24 days in two feeding trials. The three diets containing pork, cod, and egg as the main protein source gave

similar weight gains of 6.50, 5.65, and 6.15 g/da, respectively, and protein efficiencies of 2.6, 2.6, and 2.4 g gain/g protein consumed, respectively. The two all-vegetable-protein diets gave weight gains and protein efficiencies significantly lower than the animal protein diets. The soy diet without vitamins and calcium supplement produced a low weight gain of 1.65 g/da and a protein efficiency of 1.2. Rats on the rice diet realized the lowest growth, 0.29-0.44 g/da, and poor protein efficiency, 0.54-0.67. During the second set of feeding trials, three out of six rats on the all-vegetable protein diet showed marked hair loss. The plant and animal protein mixture (soybean flour and cod) was the best diet tested. Rats on this diet showed a growth rate of 6.75 g/da and a protein efficiency of 2.7.

Tests of storage stability are now being made on three of the diets.

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INTRODUCTION

Hunger obsesses the human being from birth till death. In the first years of his life he is weakest, most helpless. He is unable to provide, prepare, choose his food or even feed himself. He can only cry when hungry and wait for his mother to nurse him. The quantity and quality of his food depend largely on the financial position and the cultural and religious background of his parents. His experiences during the first years, however, have far-reaching consequences upon his health and behavior in his adult life.

Many studies have been done in the Western countries on children's dietetics and diseases. Children's societies and associations, public and private, have been created for the studies of different problems in pediatrics. Considerable knowledge of essential nutrients and their biological and physiological reactions in the human body has been collected. The standard nutrient requirements for different types of healthy persons have been worked out. Investigations on food compositions have been done. Studies on nutritional

disorders lead to the cure of many diseases such as kwashiorkor, rickets, pellagra, and beriberi.

By contrast, very few nutritional studies have been done in the underdeveloped countries where millions of people are underfed because of lack of knowledge, and lack of food. Among them, children suffer the most. Nutritional diseases in early childhood are widespread. Caloric deficiency and an unbalanced diet cause a lot of sorrow. The death rate in childhood is very high indeed.

PROBLEM OF BABY NUTRITION IN VIET-NAM

As in most of the underdeveloped countries, the nutritional problems of babies are still overlooked in Viet-Nam. Many factors account for the lack of study of children's health and the lack of a program of nutritional education for mothers. Other social problems contribute to the deterioration of the child's health.

Lack of Medical and Scientific Technicians

In the Republic of Viet-Nam there are a medical college, a pharmacological college and a faculty of sciences in Saigon, two other faculties of sciences, one in Dalat and one in Hue. There were about 60 physicians, 50 pharmacists, 80 midwives, 80 nurses and 80 bachelors of science graduated this year. The science graduates generally enter teaching. The number of scientific technicians graduated from colleges is increasing yearly, but still is very small for a population of 13 millions (Population densities are shown in Fig. 1 and Table 1; Table 2 contrasts numbers of medically trained people in Viet-Nam with those in the U.S.). Moreover,

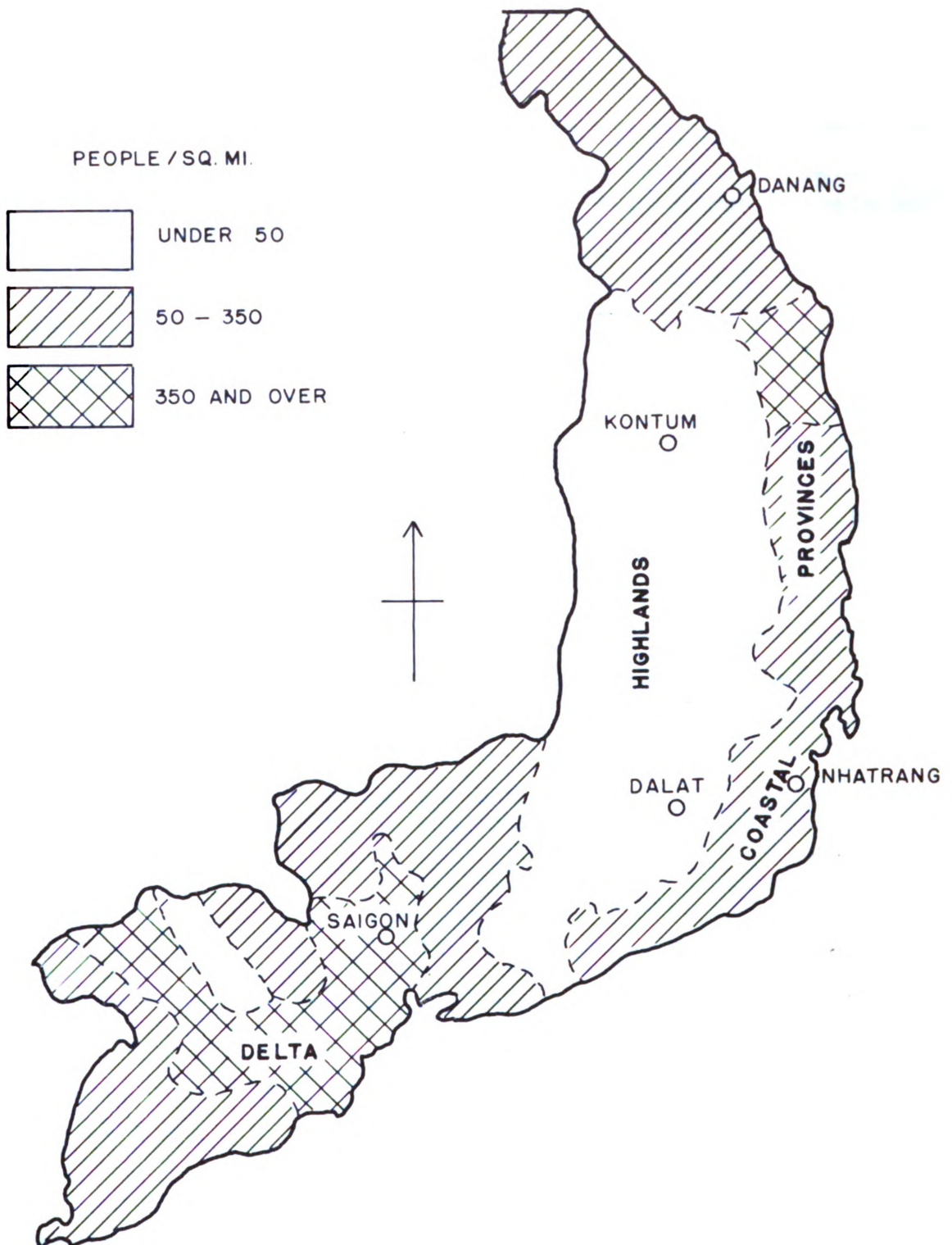


Fig. 1.--Population Density, Republic of Viet-Nam. (Source: National Institute of Statistics, Republic of Viet-Nam.)

Table 1.--Population of Viet-Nam.

Region	Area Km ²	Population	Urban Population	Density People/Km ²
South Viet-Nam	67,873	8,908,400	1,823,400	131
Lowland of Central Viet-Nam	55,647	4,296,500	408,100	77
Highland of Central Viet-Nam	48,145	584,400	100,100	12
TOTAL	171,665	13,789,300	2,331,600	80
Total cultivated area	28,628			

Source: Republic of Viet-Nam, National Institute of Statistics, July, 1959.

Table 2.--Approximate ratio of the number of medically trained people to the total population.

Profession	Number	Percent in Saigon	Republic of Viet-Nam (1958)	United States (1959)
Physician	398	64	1:33,000	1:790
Dentist	58	84	1:224,000	1:1694
Midwife	184	57	1:71,000	Not com- parable
Pharmacist	213	75	1:61,000	1:3270

Interdept. Comm. Nutri. Nat. Def., Nutrition Survey in V.N., (1959).

the scientists are concentrated in the capital, Saigon, which has about 1.5 million inhabitants, one tenth of the population.

The physicians, midwives, and nurses devote all their time to practicing curative medicine rather than preventive medicine. There are no organizations for the study of public or children's health and diseases, no journals or reviews of nutrition.

Financial Situation

The Vietnamese have a very low income. National income in 1956 was 59,496 millions piastres or 1,000 millions U. S. dollars. Each inhabitant has a yearly income of about \$77 (U. S.).

As there is a great difference in wealth among the people, the mass of the population has a much lower income than the above figure. They generally have big families, an average of 4 or 5 children per family. They sometimes have to feed old people, their parents. In most cases the men are the bread-winners while the women do the cooking and look after the children. The minimum wage set up by the Government is \$20 (U. S.) a month. On the average they earn about \$25-30 (U. S.) a month for a family of six or seven people. Wages are much lower in the country. They cannot buy expensive imported foodstuffs for their children.

The Interdepartmental Committee on Nutrition for National Defense (U. S.) in a Nutrition Survey in Viet-Nam (Oct.-Dec. 1959) interviewed 175 maternity patient families and found that their daily expense for food was \$11.8 (V. N.) per person with a range of \$4 (V. N.) to \$30 (V. N.) (\$60 V. N., official rate or \$75 V. N., market rate = \$1 U. S.). The Committee reported "Primary foods purchased in descending order of frequency were rice, vegetables, river and sea fish, nuoc-mam (fish sauce), pork, bananas, eggs, citrus fruit, and chicken. Rice was of the milled white variety in all instances, and was preferred because of better flavor and availability. Fifteen per cent of the families interviewed has meat less than once a week; 10 per cent, fruit; 5 per cent, fish; and 3 per cent, vegetables less than once a week."

These interviews were done in Saigon. In the country the daily expense of the peasants would not reach the above figures.

Cultural Factors

The Vietnamese public is not informed about the relationship between nutrition and health as there is no magazine or paper discussing nutrition topics. The women are bound by the customs of their society. The feeding of their

infants follows age-old practices of their grandmothers and mothers. Religion, custom, obligation, and love direct the mothers to visit daily or to come and stay for some time with their daughters when the latter are having babies. These "grandmothers" advise their daughters in the old-fashioned way of feeding and nursing the babies. They think that rice alone can bring good health and advise their daughters to eat a lot of rice instead of nourishing foods. That is how infant-feeding practices and prejudices against certain foods during pregnancy and lactation are maintained from generation to generation.

Expectant mothers believe that certain foods will bring illness to their infants. They think, for example, fish would cause a lot of saliva in their babies. Out of the 175 maternity patients interviewed, sixty avoided certain foods during pregnancy; 27 individuals mentioned spices; 13, salt; 8, fish; 6, chicken; and 3, fruit. Several declared that they avoided quantity of food as well as quality. They were afraid that, otherwise, the fetus would be too large and delivery difficult. This fear is prevalent among pregnant Vietnamese women.

One hundred and two out of 175 lactating mothers interviewed avoided certain foods thought to cause certain

illnesses to their nursing infants. They mentioned, in descending order of importance, fish (39), fried foods (22), vegetables (12), raw foods (30), spices (19), sour and acid food (13), shellfish, poultry, fruits, iced foods, eggs (See Table 3).

Table 3.--Foods avoided and preferred in the post-partum period

Group	Foods avoided	Foods preferred
Cereals and Tubers	Arrow-root Taro, Yam Manioc	Rice Potatoes Sweet potatoes
Meat, Fish, Eggs, Milk and Dry Beans	Sea food Snails Duck Duck eggs, Beef Buffalo, Goat	Fresh water fish Pork
Fats and Oils	All	
Fruits and Vegetables	Fresh vegetables in general	Water Convalvolus Cabbage (cooked)
Sauces and Condiments	--	<u>nuoc-mam</u>

Source: Laboratory of Nutrition (Saigon, Viet-Nam)

These expecting and lactating mothers living on a deficient diet would impair their infant's health. Their own health would suffer greatly. One fifth of them were significantly anemic. Ten per cent were deficient and 21 per

cent low in urinary thiamine; 74 per cent were low or deficient in urinary riboflavin. The infants are mostly breast fed. In town the trend is towards bottle feeding exclusively or mixed feeding (breast and bottle). In bottle feeding, they generally use condensed milk which has a very high carbohydrate content.

The Interdepartment Committee reported: "Ninety-nine per cent of the interviewed group breast fed their infants for a mean duration of 14 months. Supplementary feedings of rice or flour soup with sugar or salt were begun at a mean age of 7 months. By 3 years of age the child generally consumed the adult diet. Milk was almost never given after this time."

Breast feeding for a long period makes a significant contribution to the infant's health. But breast feeding alone does not provide enough calories or nutrients to a baby of over 5 or 6 months.

Hold (1957) presented data on milk production of healthy, well-nourished mothers. The amount secreted increases from 300-400 ml/da one week after the child's birth to 600-1000 ml/da 4 months later and thereafter slowly decreases.

The average protein content per 100 ml of breast milk is 1.2 gm. An infant of 4-5 months, weighing about 6 kg gets 700 ml of breast milk a day. His daily intake of protein is 8.4 gm or 1.4 gm per Kg weight which is a little low for his rapid growth. In the case of undernourished mothers the volume of their milk will be reduced. There is no doubt that after 6 months the amount of breast milk does not meet the infant's need for protein.

The Committee on Nutrition (American Academy of Pediatrics, 1958) concluded that "no nutritional advantage or disadvantage has yet been proven for supplementing adequate milk diets with solid foods in the first 3 or 4 months of age", but said that "no one today would question the nutritional inadequacy of a diet exclusively of milk throughout the first year, nor of the nutritional and other benefits to be derived from a varied diet at 4 to 6 months of age."

At the age of 7 months the Vietnamese child only gets supplementary feedings of rice flour which is cooked into a purée or gruel. Milled white rice, as is well-known, loses most of its thiamine. The child's food, chiefly composed of carbohydrates, lacks essential nutrients. After weaning, even the bottle or mixed feeding child generally does not get

cows milk. He has his rice gruel and sometimes chews a little cooked rice or vegetables at his parents' meal.

A great number of women become pregnant within a year of the birth of a previous child because birth control practices are prohibited by law or are unknown to them. Therefore, they must wean their infants much earlier, at the age of 5 or 6 months and give them cows milk instead. The poor families feed their babies with rice soup and nothing else.

The deficiencies of essential nutrients in the baby's diet affect his health adversely, hinder his growth, lower his resistance to infections and in many cases cause death. Many nutritional diseases are recorded such as enlarged thyroid, angular lesions, angular scars, etc. Anemia, prevalent in all groups of Vietnamese children, is commonly found in children suffering from severe protein malnutrition. It is indeed an important cause of mortality in many countries in South and East Asia. Nearly 40 per cent of the mortality in Viet-Nam falls within the 0-3 year age group. (See Fig. 2 for survival figures compared to the U. S.). The high death rate in the 0-3 year age group has perhaps a special significance, for it is during this period that the child suffering from protein malnutrition is most likely to die.

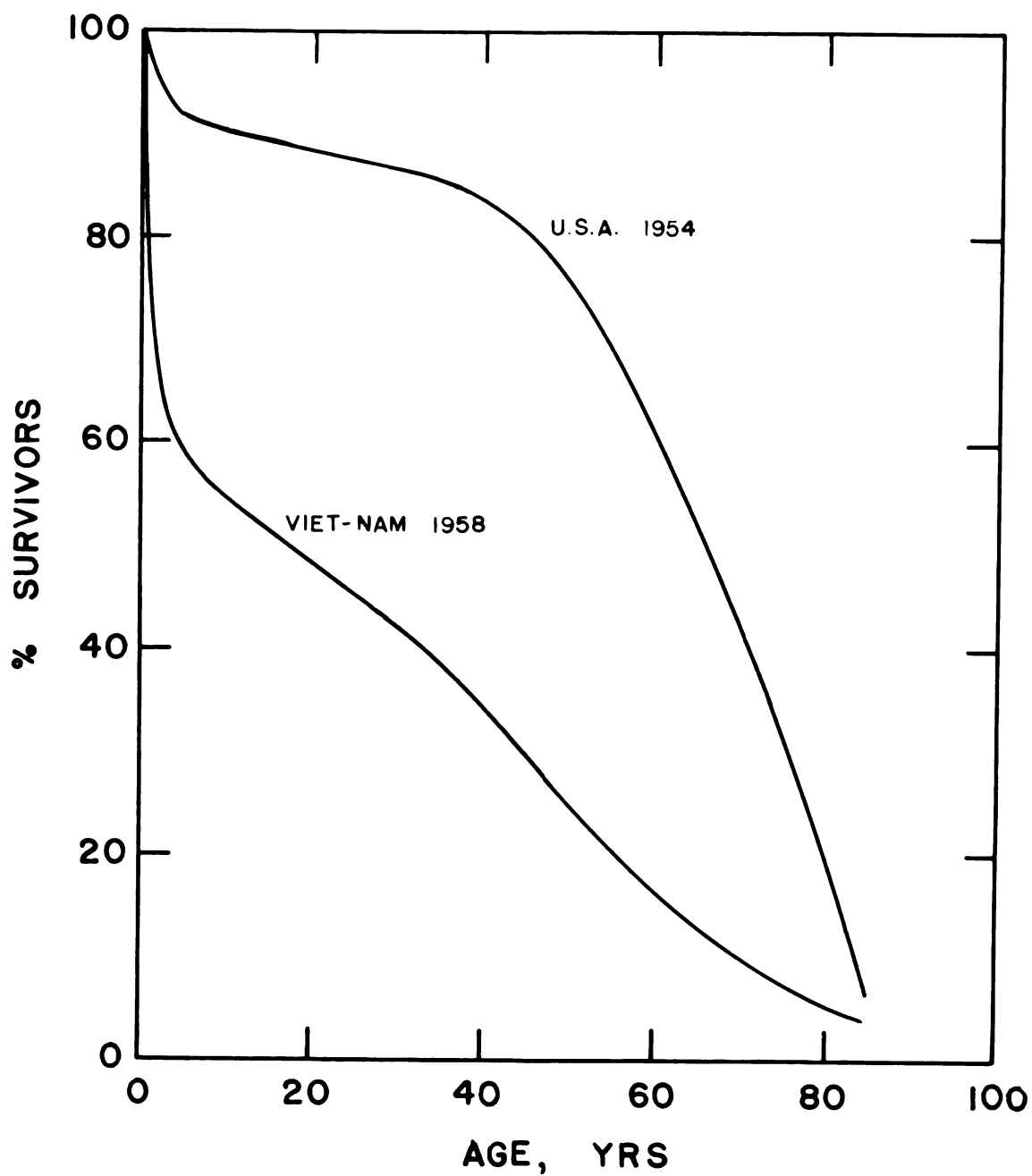


Fig. 2.--Survivor curves, United States and Viet-Nam.
(Source: U. S. Interdepartment Committee on
Nutrition Survey (Viet-Nam Oct. - Dec., 1959),
July 1960)

It seems clear that malnutrition plays an important role in the high death rate of the Vietnamese babies. Some cheap kind of nutritionally balanced food would surely curtail many deaths, prevent many of their diseases, and bring them health and strength.

The Vietnamese babies, especially those from poor families, get about a half of the U. S. or FAO recommended allowances of protein as shown in Table 4. Moreover, the rice protein has a rather low biological value, about 70. It is low in lysine and tryptophane. In this period of rapid growth a generous protein diet, in quantity and quality, must be given to the baby.

Their actual diet is low in iron and deficient in calcium. The baby requires a large amount of calcium for the formation of his bones. The diet lacks vitamin A and ascorbic acid. Its B vitamin content is very low. It is well known that milled white rice is deficient in B vitamins (FAO, 1948), still a nutritional problem for all countries having rice as a staple food. Obviously the diet does not provide the baby enough essential nutrients for his growth and general well being.

Table 4.--U.S. and FAO recommended daily allowances and probable Vietnamese baby diet.

Calculated Vietnamese Baby Diet (highest intake) ^{c,d}										
	U.S. Recommended Allowances ^a		FAO Recommended Allowances ^b		50g rice +500ml human milk		100g rice +300ml human milk		200g rice +50g sugar rice	
Age	7-12 mos.	1-3 yrs.	6-12 mos.	1-3 yrs.	7-12 mos.	7-12 mos.	7-12 mos.	7-12 mos.	1-3 yrs.	1-3 yrs.
Weight	9	12	-	-	6	6	6	6	9	9
Calories	100/kg	1300	110/kg	1235	90/kg	96/kg	96/kg	96/kg	924	905
Protein, g	-	40	3.5/kg	40	1.7/kg	1.9/kg	1.9/kg	1.9/kg	15.2	19
Calcium, g	0.8	1.0	0.9	1.0	0.172	0.126	0.126	0.126	0.048	0.060
Iron, mg	7	7	6	7	1.45	1.43	1.43	1.43	1.6	2.0
Vitamin A, I.U.	1500	2000	1340	2000	330	200	200	200	0	0
Thiamine, mg	0.5	0.7	0.5	0.6	0.11	0.12	0.12	0.12	0.14	0.17
Riboflavin, mg	0.8	1.0	0.8	1.0	0.25	0.17	0.17	0.17	0.06	0.09
Niacin, mg	7	8	5	6	1.66	2.4	2.4	2.4	3.2	4
Ascorbic acid, mg	30	35	20	25	22	13	13	13	0	0

^aU.S. Nat. Res. Counc., Recommended Dietary Allowances, 1958.

^bFAO, Nutrition et Alimentation Tropicales, Vol. 3, pp. 1837-1842 (Nutrition meeting Report No. 20), 1960.

^cU.S. Nat. Res. Counc., The Composition of Milks, 1953.

^dU.S. Dept. of Agriculture, Composition of Foods, Handbook No. 8, 1950.

REVIEW OF LITERATURE

Previous Programs

Malnutrition and undernutrition are not, of course, unique to Viet-Nam; nor are nutrition problems confined to children, although children are most affected for biological, cultural, and economic reasons. It is not surprising that several schemes to alleviate widespread nutritional deficiencies have been proposed and put into practice. It is not the purpose here to give an exhaustive review of the plans that have been proposed, but rather to note a few selected ones.

As is clear from what has already been said about Viet-Nam, malnutrition cannot be solved just by recommending an improved diet. There must be some assurance that the recommended changes are effected. Therefore, many factors, most of which have nothing to do with physical and biological science, must be considered in the design of a recommended diet.

Of special interest is INCAPARINA (INCAP, 1962), the generic name given to various mixtures of vegetable proteins

intended as dietary supplements, primarily for pre-school children. These mixtures were developed by INCAP (Institute of Nutrition of Central America and Panama) and are now in commercial production under license by INCAP. Scrimshaw et al. (1957) have outlined some of the problems, which are similar in many respects to those in Viet-Nam, faced by the developers of INCAPARINA in their 10 years of research; low cost, wide application, ease of transport and storage, local availability of ingredients, and acceptability to both children and parents are a few requirements mentioned. Table 5 shows the ingredients and composition of Formula 9B; note

Table 5.--INCAPARINA Formula 9B.

Ingredients, g/100g		Composition (per 100g)	
Ground whole corn	29	Protein	27.5g
		Fat	4.2g
Sorghum	29	Carbohydrate	53.8g
		Thiamine	2.3mg
Cottonseed flour	38	Riboflavin	1.1mg
		Niacin	7.8mg
Torula yeast	3	Vitamin A	4500 I.U.
		Calcium	656mg
CaCO ₃	1	Iron	8.4mg
		Phosphorus	698mg
Vitamin A	4500 I.U.	Sodium	3.7mEq
		Potassium	27.9mEq

Source: INCAP (1962)

that ascorbic acid is not supplied. This product can be prepared as an atole, a hot drink: 25 g of INCAPARINA plus 12 g of sugar dissolved in a glass of water and boiled for 10 to 15 minutes. The product sells for \$0.04 (U. S.) for a 75-gram bag. A one year's supply (75 g a day) for one child would cost \$14.60.

In many countries similar high protein mixtures have been prepared or tested for the supplementation of poor diets. In India the Institute of Food Technology at Mysore has produced a multipurpose food by blending 75 parts of peanut flour, 25 parts of Bengal gram (a pulse) flour fortified with vitamins and minerals (Subrahmanyam et al., 1957). The American "Food for millions" of California is based on soy-bean meal. In Uganda the British have tried a fortified buscuit made of peanut, cereal flour and sucrose (Dean, 1961). The French have tested a millet-peanut-fish mixture in Senegal (Senecal, 1961).

The addition of synthetic amino acids, lysine and threonine, to improve the nutritional value of cereals has been tested on rats by Osborne and Mendel (1914), Sure (1953), Hutchinson et al. (1956,1959), Deshpande et al. (1957), Bressani et al. (1961), and many others; but experiments on human beings have not been carried out to any large extent.

Ingredients of Proposed Diets

A review of the chemical and nutritive properties of the ingredients of the proposed diets is given below. The detailed description of the development and testing of the diets themselves is discussed in a later section.

Rice

Rice is one of the most important food crops and is the staple food of over half the world's population. Its cultivation is concentrated in Asia where 92 per cent of the world rice crop is produced.

The cultivated rice belongs to the species of Oryza sativa. There are over 8,000 varieties of rice in the world according to Ramiah and Rao (1953). The rice varieties are commercially based on the grain size and shape to be divided into short, medium, and long groups.

The removal of the outer layers of the rice grain is either by the traditional practice of home-pounding or by mechanical milling. Mechanization, industrialization and urbanization have proceeded at a quick pace in all under-developed countries. The small rice mills have nearly ruled out the laborious method of home-pounding. It is now almost

impossible to find home-pounded rice in the cities of South Viet-Nam.

People seem to prefer the highly milled rice which has better keeping qualities and a pleasant appearance. Under-milled rice becomes rancid quickly and is subject to infestation by insects. They don't know that highly milled rice has lost a great part of its trace components, especially the valuable thiamine (See Table 6).

Rice protein - Rice has a low protein content (7-8%). Its protein is of a good quality except for tryptophan and lysine (FAO, 1957).

Rice minerals - Rice is poor in iron and calcium. Its calcium-phosphorus ratio of about 1:7 is unfavorable. Kirk and Williams (1945) considered 1:2 the optimum ratio for calcium-phosphorus.

Rice vitamins - As mentioned above the milled rice has lost almost all its B vitamins, and there are many investigators who have traced the etiology of the associated disease, beriberi.

Rice enrichment in Bataan province, Philipines, a program to supply the missing thiamine, began in October 1948 and was still in effect at the time of a survey by FAO experts in 1952. Salcedo wrote (FAO, 1954) that "before enrichment

Table 6.---Percentage chemical composition of raw and milled rice.

Item	Water g	Pro- tein g	Fat g	Carbo- hydrate g	Minerals			Vitamins			
					Ca mg	P mg	Fe mg	A I.U.	Thia- mine mg	Ribo- flavin mg	Nia- cin mg
Brown, raw rice	12	8	2	78	39	303	2.0	0	0.32	.05	4.6
White or milled rice	12	7.6	trace	79	24	136	0.8	0	0.07	.03	1.6

Source: Watt B. K., and Merrill A. L. Composition of Foods - Raw, Processed and Prepared, U. S. Dept. of Agriculture, Handbook No. 8, 1950.

the annual mortality rates for beriberi were 263.57 and 149.29 per 100,000 population in the enriched rice area and for the control area respectively. Nine months after enrichment, the mortality rate was 116.7 per 100,000 in the enriched rice area, showing a reduction of 55.7 per cent while the mortality rate for the control area was 163.3 per 100,000 representing a rise of 9.84 per cent. This trend continued during the first 3 quarters of the fiscal year 1950. It dropped to zero in the experimental area after two years of enrichment."

Soy Beans

The Soy plant is botanically known as Glycine Mex.; there are some 1,500 varieties. Soybeans have been used for centuries in East Asia as human food: soy sauce, soy sprouts, soy curd, cake, and a variety of other fermented products and food preparations. They are one of the most thoroughly investigated agricultural products.

Soybean contains more protein (40-45%) than most of the legumes but less fat (16-20%) than most of the oilseeds.

Soybean is rich in phosphorus, iron, magnesium, and is a fair source of calcium. Soybean is also a fair source of some of the B-group vitamins, vitamins E and K but it does not contain vitamins A, D and B₁₂.

The figures of the average amino acids of soybean proteins given by Orr and Watt indicate that the amino acid content is quite good, except for methionine and cysteine.

Osborne and Mendel (1917) found that cooking greatly improved the biological value of soybean meal. Hayward, Steenbock and Bohstedt (1936) reported that the ordinary hydraulic process, in which the maximum temperature was about 82° C, and the low temperature expeller process, in which the oil was expelled at 105°C, produced meals of low value, but that the hydraulic process working at a higher temperature of from 130°C to 150°C gave meals of good value. Evans and McGinnis (1946), using chicks for testing, obtained the best product by autoclaving raw soybean oil meal at 100°, 110° or 120°C for 30 minutes. The effects of temperature on the nutritive value of soybeans were studied by Westfall and Hauge (1948). They autoclaved soybean flour at 108°C (5 psi) and at 120°C (15 psi) for different periods of time. They reported that autoclaving at 108°C for 15 minutes gave a protein having optimal quality. They noted some denaturation of soybean proteins at 120°C, which decreased their efficiency.

The investigators all agreed in that heat treatment improved the nutritive quality of soybean but disagreed in regard to the degree of the temperatures used.

Bowman (1948) found that soybean contained three anti-tryptic factors which could be separated by using different concentrations of ammonium sulfate. Borchers, Ackerson and Sandstedt (1947) removed nearly the whole of the inhibiting material from soybean by extraction with 0.05 N hydrochloric acid for 2 hours. The inhibiting power was destroyed by autoclaving suspensions in water for 60 minutes at 5 psi., 30 minutes at 10 psi, 20 minutes at 15 psi or 10 minutes at 20 psi, or by passing steam through the suspension for 90 minutes. They later (Borchers et al. 1951) noted that 5% of crude trypsin mixed with raw soybean oil meal completely counteracted the growth inhibition. Liener (1953) did not agree with the result and said that the restoration of growth by crude trypsin was not complete. He suggested that the growth inhibiting effect which was not completely counteracted by crude trypsin was due to the presence of an other toxic substance, the soyn in raw soybean. He attributed half the growth impairment to soyn and half to trypsin inhibitor. He later (Liener 1956, 1958) studied the soybean hemagglutinin, a leguminous plant protein which has the property of agglutinating red blood cells. He indicated that the maximum stability of soybean hemagglutinin against moderate heat was in the region of pH 6 to 7. Guanidine,

quadrivalent and trivalent metallic ions (Th^{4+} , Al^{3+} , Fe^{3+}) inhibited it. It was readily inactivated by pepsin, but it was resistant to the action of trypsin.

Everson et al. (1944) observed no appreciable varietal differences in nutritive value of soybean. They noted that there was an improvement by germination, and immature soybean seemed to supply a better protein than that of the mature bean, and heating improved the quality of the soybean protein.

The supplementary benefits of soybean to other cereals were demonstrated by Chick and Slack (1946). They indicated that a combination of 70 parts of malt extract, 10 parts of wheat flour and 16 parts of soy (on a solids basis) possessed "a growth promoting value for young rats about equal to that of the proteins of milk." This mixture contained sufficient essential fatty acids to prevent the scaliness of rat skin which had appeared with the diet containing protein only from cows milk. It also provided adequate amounts of vitamins B, so that the addition of yeast extract produced no significant benefit.

Cooper and Bryan (1951) used the American Multi-purpose Food, a fortified soybean product, as a supplement to school lunch. At the end of the school year the results

showed the benefit of the small supplement by the gain of 5 lbs per child in the group receiving 1 ounce of soybean grits per child per day as compared with the group without supplement.

Cahill et al. (1944) found that the average digestibility in adult subjects of the protein in cooked whole soybeans, cooked soy flour, and soy milk was 90.5%, 94%, and 89.6% respectively; and the average biological value of soybean protein for maintenance was 91.7%, 95.3% for cooked whole soybeans and cooked soyflour, respectively. The biological value of whole egg was 94.5%.

Soybeans used as substitutes for milk - Mackay (1940) mixed equal amounts of soyflour and full cream dried milk, added sugar and hot water and gave this mixture to 80 children from 1 to 5 months old. Their stools were more frequent and softer than those of a comparable group given cows milk without soyflour. Nineteen per cent of the soy group, but only 6 per cent of the cows milk group failed to gain weight and had gastro-intestinal upsets. The stools improved if the mixture was boiled before feeding. During the food shortage after World War II, from 1946 to 1949, Dean (1953) investigated the value of plant mixtures as additions to the diets of over 600 German children ranging from 6 months to 12 years

old. A mixture of malted barley, wheat and soyflour successfully replaced a great amount of cows milk in the diets of the 6 to 12 months old group. His trials were most successful with babies 1 to 2 years old. For the last group, the diet which had no cows milk or other animal protein proved to be very little inferior to the control diet, which was rich in milk. Barnes (1959) tested a fortified extract of soybean on 191 infants ranging from 1 to 16 months for the purpose of measuring the acceptance rate of this food. He found that it was readily accepted by 83% of them, no reactions occurred and the infants did not get tired of it. Milk substitutes like "Soyalac" and "Mullsoy" are being used in America for feeding infants and children who are allergic to cows milk.

Yellow Sweet Potato

The sweet potato, commonly used as a human and animal food, is botanically classified as Ipomaea batatas Lam. There are more than 400 varieties widely distributed over the world, chiefly in tropical countries (Thompson, 1929).

Sweet potato has less protein than potato or cereals. Its protein content is about 5-6 per cent of dry matter in comparison with 8-9 per cent in rice. But its protein is of

good quality with a protein score of 81 and compares favorably with rice protein, protein score 72.

It has a higher mineral content than rice, especially iron and calcium. It also has higher vitamin levels than rice, and is especially rich in active carotene.

Lease (1941) reported that carotene of sweet potato in a ration containing 10 per cent of fat was used, in curing ophthalmia, as efficiently by the rat as an equivalent amount of pure crystalline carotene dissolved in oil. He also noted that the dehydrated sweet potato flour still contained 90 per cent of the carotene present in the raw potato.

Consolazio et al. (1956) did a survey of the Chinese soldier diet in Formosa. They compared the amount of vitamin A in the blood serum of a control group with that of an enriched diet group. After the test they found that there was a small increase of vitamin A level from 32.2 ± 11.4 to 43.7 ± 13.0 /100 ml serum in the control group, but in the enriched diet group the concentration almost tripled from 28.7 ± 10.5 to 74.1 /100 ml serum. The addition of only 2 mg of carotene per day--from 3.77 mg of carotene to 5.98 mg--from yellow sweet potato gave this remarkable increase of vitamin A in the serum. They concluded that the carotene of yellow sweet potato was readily available for metabolic use by humans.

Pork

Pork, as other kinds of meat, is an excellent source of protein, of vitamins B complex and of some minerals. (See Table 7.)

Table 7.--Proximate composition of pork muscle. (per 100 gm)

Item	Water g	Pro- tein g	Fat g	Ash g	Thia- mine mg	Ribo- flavin mg	Nia- cin mg	Vita- min B6 mg
Fresh pork, loin (thin)	63.0	17.9	18	1.0	0.80	0.19	4.3	0.50
Cooked loin, chop (lean)	53.0	34.6	11.3	1.3	1.2	0.19	5.5	0.48

Quoted by Lushbough and Schweigert, 1960.

Leverton et al. (1952) studied the effect of the addition of meat in the diet of 2-month old infants. They found that meat addition increased the hemoglobin and red cell values. For children allergic to cows milk, if soy milk is not satisfactory, Glaser (1953) advised strained meats. He successfully used meat soups made from strained lamb or pork or beef with oil, sugar, starch, calcium carbonate, salt, and water as the sole milk substitute for newborn infants. Ziegler (1953) tested his mineral enriched meat

formula composed of rice, strained meat, calcium, phosphorus, and vitamins on rats, which thrived very well. He also tried his rice-meat soup with all necessary minerals and vitamins on infants and children suffering from a variety of clinical disorders admitted to the University of Minnesota Hospitals and remarked striking improvement in the patients' condition. A satisfactory gain in weight was also noted.

In their feeding experiments of raw pork and processed pork to rats Poling et al. (1944) concluded that a commercial canning process resulted in a small decrease in the nutritive value of the proteins of the cured pork. They found that dehydrated pork had a higher nutritive quality than canned cured pork and roast fresh pork. In a review of literature on dehydrated foods, Doty et al. (1953) mentioned that there was little loss of the chemical constituents of dehydrated meat if the cooking liquors were incorporated in the dried products. Biological value and digestibility of the protein were not affected by the dehydration of meat.

In a chapter on the nutritive value of meat and meat products Lushbough and Schweigert (1960) reported that the thiamine levels of fresh pork are many times higher than those of beef, veal and lamb. They also noted that the retention of vitamins in processed meat varies in relation to

their stability in processing. Thiamine is sensitive to the severity of the heat treatment. Cooked and processed meats retain about 75 per cent of their original thiamine content. The retention of riboflavin and niacin in cooked meats ranges from 60 to 85 per cent and that of vitamin B6 from 45 to 65 per cent.

Rice and Robinson (1944) revealed that the storage of dehydrated pork at 26.7°C over a period of 219 days showed a thiamine retention of 29 per cent while at temperatures above 37°C thiamine decreased rapidly. There was little or no loss of riboflavin or niacin of dehydrated pork stored at temperatures up to 37.2°C . In an earlier study Rice et al. (1944) found that mixtures of 67 per cent of dried pork and 33 per cent of dried pork and 33 per cent of cereals retained 75 to 97 per cent, six times as much as dried pork alone when they were stored for 7 days at 49°C . Niacin and riboflavin were shown to be stable for nearly a year at temperatures up to 37.7°C (100°F). High storage temperatures had a destructive effect on the thiamine of dehydrated pork. Mixtures of 86 per cent of dried pork and 14 per cent of soy flakes had a thiamine retention two or three times as much as dehydrated pork alone (Nymon and Gortner, 1947).

Lushbough and Schweigert (1960) pointed out that, unless the cooking liquor or the drip were thrown away, the processed meat might retain all its minerals.

Dried Egg Powder

Egg is a very popular food all over the world. It is a good source of proteins, minerals and vitamins. It has an excellent nutritive value.

The amino acids of egg are excellent, well-balanced, and have, by definition, the highest protein score of 100 (FAO, 1957).

An experiment done by Swenseid et al. (1959) showed that six out of seven subjects had nitrogen equilibrium with an intake of 6.5 g total nitrogen. All the essential amino acids in their diet were supplied by 100 g of cooked egg. In their review of literature on the nutritive value of egg Everson and Souders (1957) reported that the comparative analyses of McNall did not reveal any difference in the total nitrogen of dried eggs and fresh eggs as calculated on a dry basis. Prolonged storage at room temperature had no effect on the amino acids of dried egg powder.

Using the biological and spectrophotometric methods for investigation Denton et al. (1944) found, in accord

with previous investigators, that dehydration caused little loss in the vitamins A, D, and riboflavin in whole egg. Vitamin A content noticeably decreased when spray dried eggs were stored at high temperatures 80°-98°F, or exposed to sunlight. Vitamin D and riboflavin were stable during storage periods of from 4 to 6 months at temperatures up to 98°F. In sealed tin cans dried eggs retained 75 per cent of their vitamin A regardless of storage temperatures (Bohren and Hauge, 1946). Klose et al. (1943) reported no destruction of thiamine during spray drying. On the contrary Cruickshank et al. (1944) mentioned 30 per cent loss of this vitamin in spray-dried egg. Klose et al. (1943) also indicated that thiamin retention of dried egg powder during storage depended on the storage temperatures. There was no loss at 15°F for 9 months; little destruction during 6 months of storage at 70°F was noted, but, afterwards, the thiamine retention decreased to 50% at the end of 9 months; at 98.6°F, 26% was lost during the first month of storage and after 9 months 50% remained.

Fish

The FAO's Nutrition Division has considered edible fish meal as a cheap and good source of protein of high nutritive value to supplement the diet of poor people in

tropical regions. United Nations Children's Fund (UNICEF) is also interested in "the possibilities of using fish flour as an alternative to milk protein" because of its high content of proteins, minerals, and B vitamins (FAO Fisheries Bull., 1957).

It is generally agreed that fish protein is of very high nutritive value, but this value may be reduced by improper processing methods. Carpenter et al. (1957) used dehydrated fish meal as supplementary protein to cereals in the feeding trials of chicks. They found that drying at high temperature, 105°C decreased 20% in "gross protein value" of fish than drying at 55°C. They also reported that fish protein is more readily damaged at storage when it has a high moisture content. There was no significant reduction of the nutritive value of dried cod fillets, having 2-3% moisture, packed in nitrogen and held at 100°C for 31 hours; but there was a reduction of 28% in nutritive value of similar cod, having 11% moisture, held at 105°C for 36 hours. Sure (1957) reported that the addition of 1% defatted fish flour to whole yellow corn brought a 3.5-fold increase in rat weight and 119.7% increase in protein efficiency ratio. The addition of 3% fish flour resulted a 4.5-fold increase in rat weight and 131.1% increase in protein efficiency ratio. The addition

of 5% fish flour produced a 7-fold increase in rat weight and 177% increase in protein efficiency ratio. Good influence on growth and protein efficiency was also found when fish flour was supplemented to whole wheat, whole milled rye, grain sorghum and millet. Sure found that dehydrated fish products contained enough minerals for the good growth of albino rats. Simulated Indian diets containing 1% and 3% of fish flour were fed to rats which showed a much better growth than the basal diets (Metta, 1960).

Fish biscuit composed of 12% millet flour, 25% sugar, 20-24% groundnut press-cake flour, and 12-14% deodorized fish flour were tried on 47 Malayan children for a month. They were given 14 biscuits containing about 70 gm protein per week. The children receiving biscuit supplement showed a better weight gain than the control group (Thomson and Merry, 1962).

Yeast

Yeast used as food consists of dead, dry cells. There are several types of dried yeasts; but the two varieties the most used are Saccharomyces cerevisiae (brewer's dried yeast) and Torulopsis utilis (Torula). In the United States the only types admitted for food enrichment are brewers' dried yeasts which must contain not less than 40 per cent of

protein, and, in each gram, the equivalent of not less than 0.12 mg of thiamine hydrochloride, 0.04 mg of riboflavin, and 0.25 mg of niacin (U. S. Pharmacopeia 14, 1950).

Dried yeast has a high percentage of proteins which contain all the essential amino acids (Block and Bolling, 1943). It also contains non-protein nitrogenous constituents such as purines, pyrimidines, glucosamine, etc. which are included in the nitrogen determination and calculated as protein. There is an apparent lack of sulfur amino acids in yeast. In spite of these defects some believed that yeast can be used as a major source of protein if it is supplemented by some cheap source of sulfur-containing amino acids (Von Loesecke, 1946). Trials of yeast as a protein concentrate for the feeding of stock animals have all shown it to possess a high nutritive value.

It is well-known that yeast is an excellent source of B vitamins. Boiled yeast is a better source of thiamine and riboflavin than fresh, unboiled yeast (Parsons et al., 1945). Fresh yeast fed to human subjects was poorly absorbed as source of thiamine. There was even an decrease of urinary thiamine below the preceding basal period, and an increase of fecal thiamine. Living yeast cells were recovered in the feces. When boiled yeast was fed, there was a marked

improvement in thiamine absorption. There was also no absorption of riboflavin from the fresh, raw yeast. Riboflavin was greatly absorbed when the subjects were fed dead, dried yeast (Price et al., 1947).

Hughes (1946) carried out yeast feeding trials in Lagos prison where a high percentage of prisoners had recognizable lesions of riboflavin deficiency. He suggested that "4 grams of food yeast was not quite sufficient to supply the deficiency. A supplement of 9 grams, however, produced cure in all cases." He recommended 10 grams of yeast for the prevention of the local malnutrition. Von Loesecke (1946) summarized two different opinions: Wintz, Salomon, Funk, Dirr, etc. reported an increased output of uric acid with intake of yeast; on the contrary Pierce, Still and Kock did not notice such an increase of uric acid excretion when yeast was added to a low protein diet.

In France sweet biscuits containing 8-12% yeast and non-sweet biscuits containing 20-25% yeast are well-accepted by the consumers. In 1955 the Sub-Committee on Food Yeast Research of the French National Center of Nutrition Research recommended a daily distribution to school children of 10 grams of dried yeast. It proclaimed that this intake would bring an appreciable amount of vitamins B complex to the

children and would not give them any undesirable side-effects (Gounelle and Cofman, 1956).

Calcium

Calcium is one of the chief elements for bone development. Although calcium is needed at all ages, requirements are greatest during infancy, at the time when the child develops rapidly.

In many countries the inclusions of nutritional ingredients are either enforced by law or done voluntarily by the bakers or millers. The level of calcium prescribed in the United States is 300-800 mg per pound of enriched bread, with an average of 400 mg (NRC, 1958). In Sao Paulo, Brazil the level of calcium supplement per kilogram of flour is 2,500 mg as $\text{Ca}_3(\text{PO}_4)_2$; in Canada 1,250 mg per kilogram of flour as CaCO_3 or as edible bone-meal; in Denmark 2,000 mg as CaCO_3 ; in England 1,200 mg (Menden and Cremer, 1959).

PROPOSED BABY FOOD MIXTURES

All the foods reviewed which have a very high protein content (20% for the African biscuits, 25% for INCAPARINA, 42% for the American and Indian multipurpose foods) are intended as a treatment for kwashiorkor or as a supplement to the ordinary diet of the poor.

It is the purpose of this study to formulate a number of enriched foods which will approach the FAO or U. S. recommended allowances. The proposal here is to take rice as a basic ingredient and enrich it with other native products, preferably those already produced in abundance (See Tables 8, 9, and 10.). The addition to rice of soy, fish, pork, or egg in appropriate quantity can bring the protein content of the mixture up to 15%, which is equivalent to 15-16% calories, slightly higher than the 14% level recommended by the British Medical Association (Davidson et al., 1961). Other ingredients must be added to supply certain vitamins and minerals: sweet potato for vitamin A, yeast for vitamin B complex, and calcium carbonate for calcium.

Table 8.--Selected crops in Viet-Nam.

Main Food Crops	Area, ha	Production metric tons
Paddy	2,503,460	5,311,250
Sweet Potatoes	41,033	203,245
Manioc	34,059	180,878
Maize	26,952	26,315
Beans	2,114	964
Soy beans	2,219	1,416
Mung beans	10,340	5,993
Vegetables	10,046	105,040
Pineapple	5,749	40,915
Other fruits	37,156	249,988
<u>Oleaginous Crops</u>		
Coconuts (1,000)	34,450	117,765
Peanuts	23,379	18,816
Sesame	2,141	530
<u>Other Crops</u>		
Tea	8,964	4,138
Coffee	5,606	3,340
Tobacco	6,521	5,211
Black pepper	374	344
Sugar cane	29,202	823,750

Source: Department of Agriculture of Viet-Nam, 1959.

Table 9.--Livestock and poultry production in Viet-Nam, 1960.

Animal	Number
Buffaloes	707,000
Cattle	1,030,000
Pigs	3,500,000
Chickens	16,322,000
Ducks	9,887,000

Table 10.--Vietnamese exports.

Items	Quantity, Metric Tons
A. Vegetable products:	
Rice and broken rice	154,451
Rich by-products	1,702
Oil cake	2,952
Peanuts	1,697
Sesame	199
Dry beans	377
Starchy roots and tubers	504
Fresh vegetables	210
Edible fruits	290
Coconut oil	2,825
Peanut oil	346
Maize	1,117
B. Animal products:	
Hogs	5,293
Eggs	1,194
Egg yolk	234
Meat	191
Bones	1,980
C. Fishery products:	
Fish	392
Crustaceans	241

Source: Department of Agriculture of Viet-Nam, 1961.

Table 11 shows five fortified diets and contrasts some of their nutritional components with a diet of rice only. Note that 20-30% substitution with supplementary ingredients is required to increase the level of essential

Table 11.--Diet ingredients and proximate composition^a per 100 g.

Diet code	RICE	SOY	VEG	COD	PORK	EGG
Ingredients, per cent	Rice 99 NaCl 1	Rice 75 Soy flour 24 NaCl 1	Rice 69.5 Soy flour 19 Sweet potato 6 Yeast 4 CaCO ₃ 0.5 NaCl 1	Rice 81.5 Cod 7 Sweet potato 6 Yeast 4 CaCO ₃ 0.5 NaCl 1	Rice 79.5 Pork 9 Sweet potato 6 Yeast 4 CaCO ₃ 0.5 NaCl 1	Rice 73.5 Egg 15 Sweet potato 6 Yeast 4 CaCO ₃ 0.5 NaCl 1
Composition:						
Protein, g	7.5	15.1	15.0	14.7	14.9	15.0
Calcium, g	0.024	0.060	0.26	0.23	0.22	0.24
Iron, mg	0.8	3.4	3.4	2.0	2.2	2.5
Vitamin A, I.U.	0	28	1200	1200	1200	1400
Thiamine, mg	0.07	0.20	0.42	0.37	0.40	0.38
Riboflavin, mg	0.03	0.08	0.28	0.25	0.25	0.35
Niacin, mg	1.6	1.7	3.2	4.0	4.0	3.0

^aWatt, B. K., and A. L. Merrill, Composition of Foods--Raw, Processed, Prepared, USDA Handbook No. 8, 1950, except protein, which was measured for this study.

nutrients substantially. Except for the SOY diet, the proposed mixtures will provide, in contrast to rice only, a 2-fold increase in protein and niacin and 3-fold increase in iron, 5-fold in thiamine, 10-fold in riboflavin and calcium, and from 0 to 1200 I.U. of vitamin A. A hundred grams of fortified foods would provide over half the U. S. recommended allowance of vitamin A and thiamine, about half the protein and niacin, and about a third of the recommended allowances of riboflavin, calcium, and iron. (Compare Tables 4 and 11.) The addition of the protein-rich ingredients will also improve the quality of the protein in the mixture, as calculated by protein score (Table 12). With the exceptions of calcium carbonate and salt, all fortification is with agricultural products.

The mixtures will be a dry powder, which has certain advantages: they look like the commonly used rice flour; they can be easily processed; they need only moisture-proof packaging, and should have good keeping qualities without refrigeration. Provided the cost of raw materials, processing, packaging, storage, and transportation can be kept low, they should make a cheap food. They are prepared by cooking with water into a puree the texture and consistency of which is not greatly different from the traditional rice gruel. As

Table 12.--FAO provisional pattern of essential amino acids^a contrasted with the calculated^b pattern in the proposed diets. (mg amino acid/g N)

Diet code:		RICE	SOY	VEG	COD	PORK	EGG
Amino Acids	Protein source, %	Rice 100	Rice 37 Soy 63	Rice Soy	35 Rice 50 Cod	43 Rice 42 Pork	41 Rice 44 Egg
	(total protein in diet equals 100%):			Yeast Sweet potato	12 Yeast Sweet 3 potato	12 Yeast Sweet 3 potato	12 Yeast Sweet 3 potato
Iso-leucine	270	322	329	326	318	310	371
Leucine	306	535	503	492	496	485	531
Lysine	270	236	334	342	330	388	338
Phenylalanine	180	307	308	303	306	273	331
Tyrosine	180	269	226	234	221	248	270
Sulfur							
containing:							
Total	270	222	206	205	234	222*	274
Methionine	144	142	107*	110*	153	144	183
Threonine	180	241	245	256	270	275	285
Tryptophan	90	65*	78	81	69*	77	89*
Valine	270	415	360	368	374	378	424
Protein score	100	72	74	76	77	82	99

^aFAO, Protein Requirements, Nutritional Studies No. 16, 1957.

^bOrr, M. L., and B. K. Watt, Amino Acid Content of Foods, U. S. Dept. of Agri., Home Economics Research, Report No. 4, 1957.

* Limiting amino acid.

basically formed from rice, 70-80%, their color, odor, and taste should be acceptable.

These mixtures are intended to be used as staple foods for weanling babies. An amount of 150-200 g dry powder will give a child of 1-2 years of age, weighing 6-9 kg, all the necessary nutrients for good development (Table 11) with the exception of ascorbic acid.

The experimental tests of the proposed mixtures consisted of two series of feeding trials on weanling rats and the initiation of storage stability studies. In the first test series, the six diets shown in Table 11 and a positive control, commercial dog chow, were fed. In the second series, the RICE, VEG, and COD diets (Table 11) and the dog chow control were repeated and two new diets (described below) were added. The storage stability studies were started on three of the more promising diets.

PREPARATION OF THE INGREDIENTS

Rice Flour

For the first experiment white milled rice was ground in a Wiley mill through a 20-mesh screen. In the second experiment commercial rice flour of unknown history was used.

Soy Flour

For both feeding tests, soy beans obtained from a local elevator were soaked in water over night, dehulled by hand, and washed. They were then autoclaved at 110°C for 30 minutes. The autoclaved beans were dried at $64-66^{\circ}\text{C}$ for 24 hours in a small tunnel dryer. The dried beans were ground in a Wiley mill through a No. 20 sieve.

Cod

In both feeding tests, frozen cod fillets were cut in large pieces, put in a pan, covered with water, brought to a boil, and cooked for 20 minutes with occasional stirring. The cooked fish was drained through cheese cloth and slightly

pressed. It was then dried in a forced air dryer at 82-85°C for one hour, then at 71-73°C for three hours. The dried fish was ground in a Wiley mill through a No. 20 sieve. Five pounds of cod yielded 12-13 ounces of fish flour.

Pork

Pork loin was trimmed of fat and chopped into small pieces. The pork pieces were cooked in boiling water for 20-25 minutes with occasional stirring. The cooked pork was drained, partially dried, and then ground through 3/8-in. holes of an electric meat grinder. The fat on the surface of the gravy was drawn off. The gravy was evaporated to about one-tenth of its former volume and added back to the ground meat. The ground pork was dried at 77-80°C for one hour, then at 71-73°C for 5 hours in a forced air drier. The dried pork was hand-pounded in a mortar. Five pounds of pork gave about 1 lb. 4 oz. of dried, ground meat.

Whole Egg Powder

A commercial product, of unknown history, was used.

Yeast

It was a commercial strain, possibly Anhauser-Busch chipped yeast, strain G.

Dried Sweet Potato

The Southern Regional Research Laboratory provided their dehydrated sweet potato flakes. The potatoes had been blanched 6 minutes with steam at 212°F, sprayed 2 minutes with a sulfate solution containing 0.4% Na₂SO₃ and 0.1% NaHSO₃, then drained for 10 minutes and dried at 175°F for 20 hours (Molaison et al., 1962). The dehydrated sweet potatoes were packed in tin cans and sealed in an atmosphere of nitrogen containing less than 2% oxygen on November 28, 1961. Before use in the feeding tests, the flakes were ground in a Wiley mill through a 20-mesh screen.

Dog Chow

A commercial dog food, Purina chow, was ground in a hand operated meat grinder.

Calcium Carbonate

A laboratory grade was used.

Salt

Diamond crystal salt was used.

PROXIMATE ANALYSIS OF INGREDIENTS

A proximate analysis of materials is necessary in order to know the major food constituents, and especially to formulate the protein level of the test diets for biological assay. The methods are given below; results are given in Table 13.

Moisture Determination

The raw or cooked materials, except the cooked pork were ground in a Wiley mill through a mesh No. 20. The cooked pork had too high a fat content for grinding and was hand-pounded in a mortar. Duplicate 10 mg samples of all ingredients were weighed and dried in an electric oven at 103-105°C for 5 hours. They were cooled in a dessicator and weighed. The loss in weight was reported as per cent moisture, wet basis.

Protein Determination

Nitrogen was determined on triplicate samples by the Kjeldahl method. The quantity of nitrogen found was multiplied by the factor 6.25 for conversion into protein.

Fat Determination

The ground, oven-dried materials were weighed in duplicate samples of 5-20 g, depending on the expected fat content. They were extracted by ether in a Soxhlet apparatus for 16 hours, then the ether was evaporated at low heat. The receiving flask was dried at 100°C for 1 hour, cooled in a dessicator and weighed. The percentage of fat was calculated on the powder weight before oven drying.

Ash Determination

Duplicate 2-gram samples were weighed in tared, porcelain crucibles. Then they were incinerated at 600°C for 2 hours, cooled in a dessicator and weighed. The percentage of ash was also calculated on the powder weight before oven drying.

Table 13.--Proximate composition of the ingredients.
(per 100 grams)

Item	Water g	Protein g	Fat g	Ash g
White rice	11.8	7.5	0.2	0.5
Rice flour	8.7	7.8	0.2	0.5
Sweet potato	3.6	7.5	0.6	2.6
Soy flour	6.7	39.8	19.0	5.1
Cod powder	5.0	88.1	0.9	6.3
Pork powder	1.8	72.6	22.7	5.6
Egg powder	3.3	47.5	23.2	4.1
Yeast	4.9	48.9	2.0	6.2
Dog chow	6.5	24.5	7.1	7.9

BIOLOGICAL ASSAY

First Feeding Experiment

A growth study was conducted on weanling Holtzman albino male rats. At arrival they were all put on the dog chow diet for a day; then they were divided at random into seven groups of six rats each, and placed in the cages in a random manner. One control group was fed with the commercial dog chow, a second group with the RICE diet, and the five other groups with the five proposed mixtures shown in Table 11. The initial weights at 0 days (average 58.8 g, range 54-64 g) were not significantly different among groups. The rats were kept in individual cages with raised screen bottoms and received food and water ad libitum. Food consumption was recorded every two days (the amount of scattered food was estimated) and each rat was weighed, to the nearest gram, every four days.

The growth response of the rats was tested over a 24-day period; calculations were based on the last 20 days. Several rats which showed hair loss were maintained on that diet for a longer period. The rats on the RICE diet were

switched, at the end of the 24-day test period, to a mixture of the remaining COD, EGG, and PORK diets. The remaining rats were killed at the end of the 24th day.

Results of the First Feeding Experiment

RICE diet: The rats in this group grew very little and gained only 6 g on the average for the whole feeding period. Nevertheless, they remained healthy, but they were not lively and slept most of the time in a corner of the cage. They ate little, only about 7 g/da; their coats were not shiny. Because of lack of flesh, their skin became wrinkled and the skeletal structure was very apparent. What a difference at the end of the test period when they were given a mixture of the COD, EGG, and PORK diets. Their weight shot up from 68 g to 141 g in 8 days, with an average weight gain of 9 g/da. They were lively now, ran around, and ate a lot, about 14 g/da. They looked fat and had a fine coat.

SOY diet: The rats on this diet grew fast in the first 8 days, then more slowly, and stopped gaining weight after 20 days. One of them lost considerable hair under the stomach and at the hind legs and died after 42 days.

VEG diet: The rats on this diet also grew well in the first 8 days, 5 g/da, then at a slower rate, 1-2 g/da.

Three out of six lost some hair, as shown in Fig. 3a, but they gained weight. One of the rats which had lost its hair was given daily about 5 mg of inositol for 15 days after the 24-day test period and partly recovered its hair. One other rat was given daily about 5 mg of calcium pantothenate and also showed some recovery of hair.

COD, EGG, and PORK diets: The rats in these groups grew admirably well and gained, on the average, 6 g/da for the last 20 days of the feeding period. No deficiency was noted in any of these rats. (Fig. 3b compares one of these rats with a rat on the RICE diet.)

Dog chow: The rats on this control diet showed the highest gain in weight, 7 g/da.

Nearly half the rats in this first feeding study showed symptoms of respiratory infection, which became evident after about two weeks. The number showing obvious symptoms was about the same for each diet, and, although, in each diet, the rat showing the lowest gain also showed symptoms of respiratory infection, other rats on the same diet showed symptoms but had average or better-than-average gains. No adjustments to the data have been made because of infection, but it is worth noting that the standard deviation of the weights at 24 days (all diets except RICE) was higher in the



Fig. 3a.--Hair loss on the hind leg of a rat on the VEG diet after 24 days of feeding test.



Fig. 3b.--Comparative growth at the end of 24 days of a rat on the RICE diet (left) and a rat on the EGG diet (right).

first feeding test than that in the second feeding test, 15.8 g vs. 11.2 g.

Performance is summarized in Table 14, and the average weights are shown in Fig. 4. The plotted points in Fig. 4 do not necessarily show significant differences. According to an analysis of variance of weights, by the fourth day, the rats on the RICE diet were significantly lower in weight and remained so for the rest of the test. After the fourth day, the weights of rats on the RICE diet were not included in any of the statistical analyses because the variance was significantly lower than for the other diets. After 24 days, the average weights of rats on SOY and VEG diets were significantly different from each other and from all other diets; average weights of rats on COD and EGG were significantly different from the average of the rats on dog chow, but not PORK; average weights of rats on PORK and dog chow were not significantly different. These differences were analyzed by t-test, for which the protection levels may be too small.

Second Experiment

In the light of the first experiment, a second experiment was carried out with six groups of weanling Holtzman

Table 14.--Average weight gain, food consumption, and protein efficiency during the last 20 days of a 24-day feeding period.

Diet Code	Weight gain g/day	Food consumption g/day	Protein intake g/day	Food efficiency g gained/g consumed	Protein efficiency g gained/g prot. cons.
RICE	0.29	7.2	0.54	0.04	0.53
SOY	1.7	9.0	1.36	0.19	1.22
VEG	3.1	12	1.80	0.26	1.70
COD	5.7	15	2.20	0.38	2.60
EGG	6.1	17	2.55	0.36	2.40
PORK	6.5	17	2.50	0.39	2.60
Dog chow	7.2	17	4.16	0.42	1.70

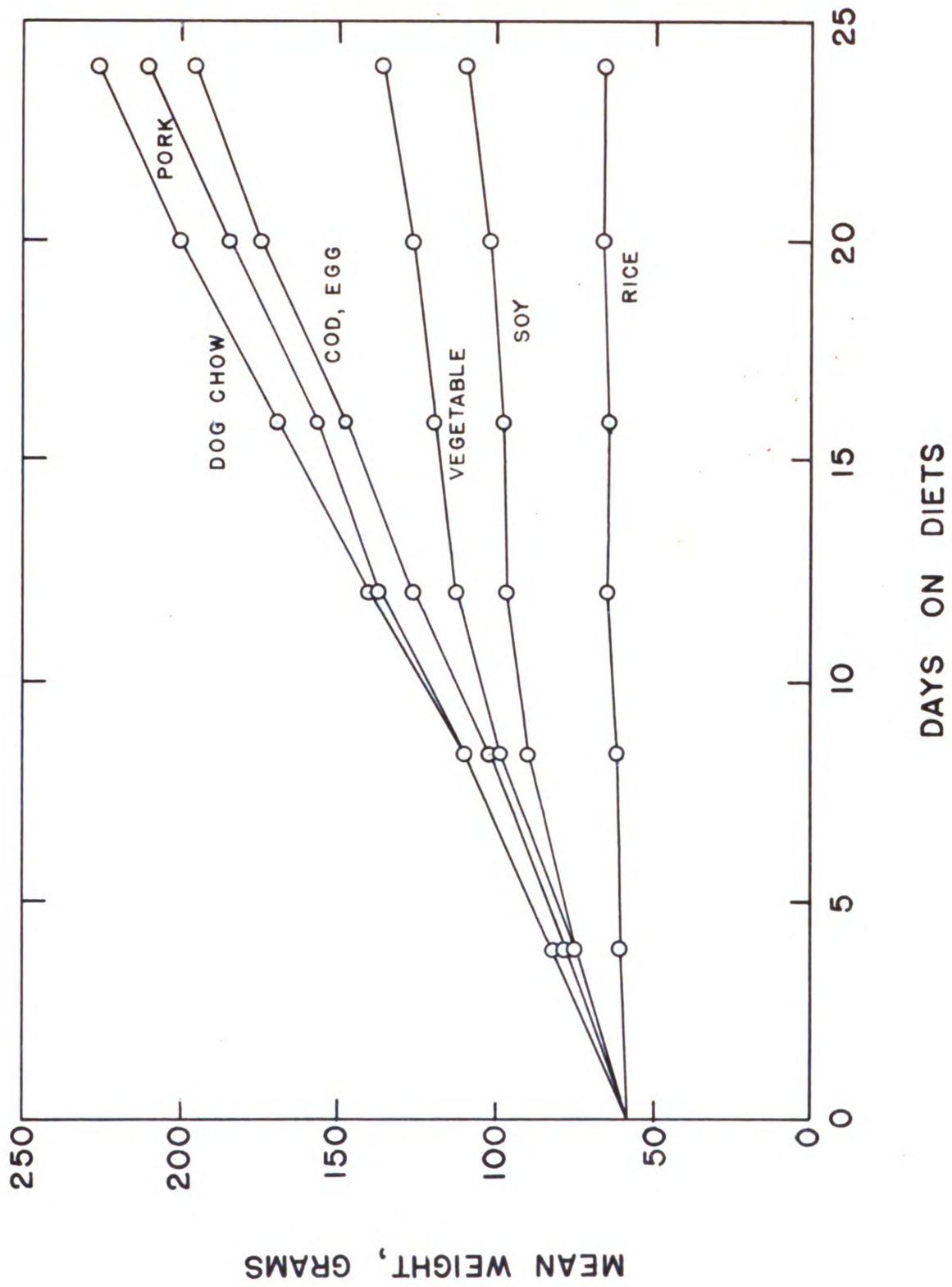


Fig. 4.--Average weights of male weanling rats, First Feeding Test.

albino rats to clarify some points and verify the results of the first test. Upon arrival, the rats were put on dog chow for a day, then assigned randomly in individual cages as described in the first feeding test.

Except for the RICE diet, all the other diets were brought up to about 15% protein. the RICE, VEG, and COD diets were the same as in the first experiment. Two new diets were introduced: VIP, which is the same as the VEG diet with the addition of two B vitamins, inositol and calcium pantothenate; and PAP, a plant and animal protein diet, a mixture of soy and cod meals. Ingredients are given in Table 15.

Table 15.--Diet ingredients, for second feeding tests g/100 g.

Ingredients	Diet code				
	RICE	VEG	VIP	PAP	COD
Rice	99	69.5	69.5	75.5	81.5
Soy flour		19	19	9	
Cod				4	7
Sweet potato		6	6	6	6
Yeast		4	4	4	4
CaCO ₃		0.5	0.5	0.5	0.5
NaCl	1	1	1	1	1
Inositol			0.010		
Ca. pantothenate			0.010		

Results of the Second Feeding Experiment

RICE diet: As in the first experiment, the rats of this group gained very little weight, an average of 0.40 g/da. They were very thin and three began to show some hair loss after two weeks on test.

VEG diet: These rats grew quite well during the first 12 days, then they gained little weight. One of them lost some hair during the last week of the experiment.

VIP diet: The rats grew quite well, but one lost some hair during the last week.

COD diet: These rats exhibited good growth; no sign of deficiency was remarked in any of them.

PAP diet: The rats in this group were very healthy and grew admirably; they gained on the average 6.8 g/da.

Dog chow: The rats in this control group grew as well as did those on dog chow in the first feeding test.

No signs of respiratory infection were noticed throughout the test period. All rats were killed after the 24th day.

Performance based on the last 20 days of the test period is shown in Table 16. With the exception of the RICE diet, the males showed higher weight gains and higher protein efficiencies than the females.

Table 16.--Average weight gain, food consumption, and protein efficiency during the last 20 days of a 24-day feeding period. (Test 2)

Diet code	No. of rats	Weight gain g/ day	Food consumption g/ day	Protein consumption g/ day	Food efficiency g gained/ g consumed	Protein efficiency g gained/ g prot. cons.
RICE	Male: 5	0.44	8.8	0.68	0.05	0.67
	Female: 1	0.95	11.1	0.86	0.09	1.1
VEG	Male: 4	4.35	14.6	2.29	0.30	2.0
	Female: 2	2.60	11.6	1.74	0.22	1.5
VIP	Male: 3	5.75	16.5	2.47	0.35	2.3
	Female: 3	3.28	12.4	1.86	0.26	1.8
COD	Male: 2	6.12	15.3	2.24	0.40	2.7
	Female: 4	4.60	14.4	2.11	0.32	2.2
PAP	Male: 6	6.76	17.0	2.55	0.40	2.7
Dog chow	Male: 4	6.81	15.8	3.87	0.43	1.7
	Female: 2	4.78	14.7	3.60	0.32	1.3

The average weights of the males are plotted in Fig. 5. It happened that the average initial weight of the rats on the RICE diet, 59.0 g, was significantly higher than the average weight of all the other rats, average 54.2 g, range 51 to 60 g. On the fourth day, and thereafter to the end of the test, the average weight of the rats on the RICE diet was significantly lower than the average weights of rats on all other diets. As in the first test, rats on the RICE diet were not included in any statistical analyses because of the significantly lower variance of the weights. An effort has been made in Fig. 5 to show significant differences, 5% level, with one exception noted below, as determined from t-tests. At 16 days, rats on the VEG diet were significantly lower in weight than those on the PAP and dog chow diets. At the end of the test, rats on the VEG diet were significantly lower in weight than rats fed on all other test diets; rats on VIP were lower in weight than those fed on PAP and dog chow; average weights of the rats fed on COD were only slightly higher than those fed on VIP and significantly lower than those fed on PAP and dog chow at the 10% level.

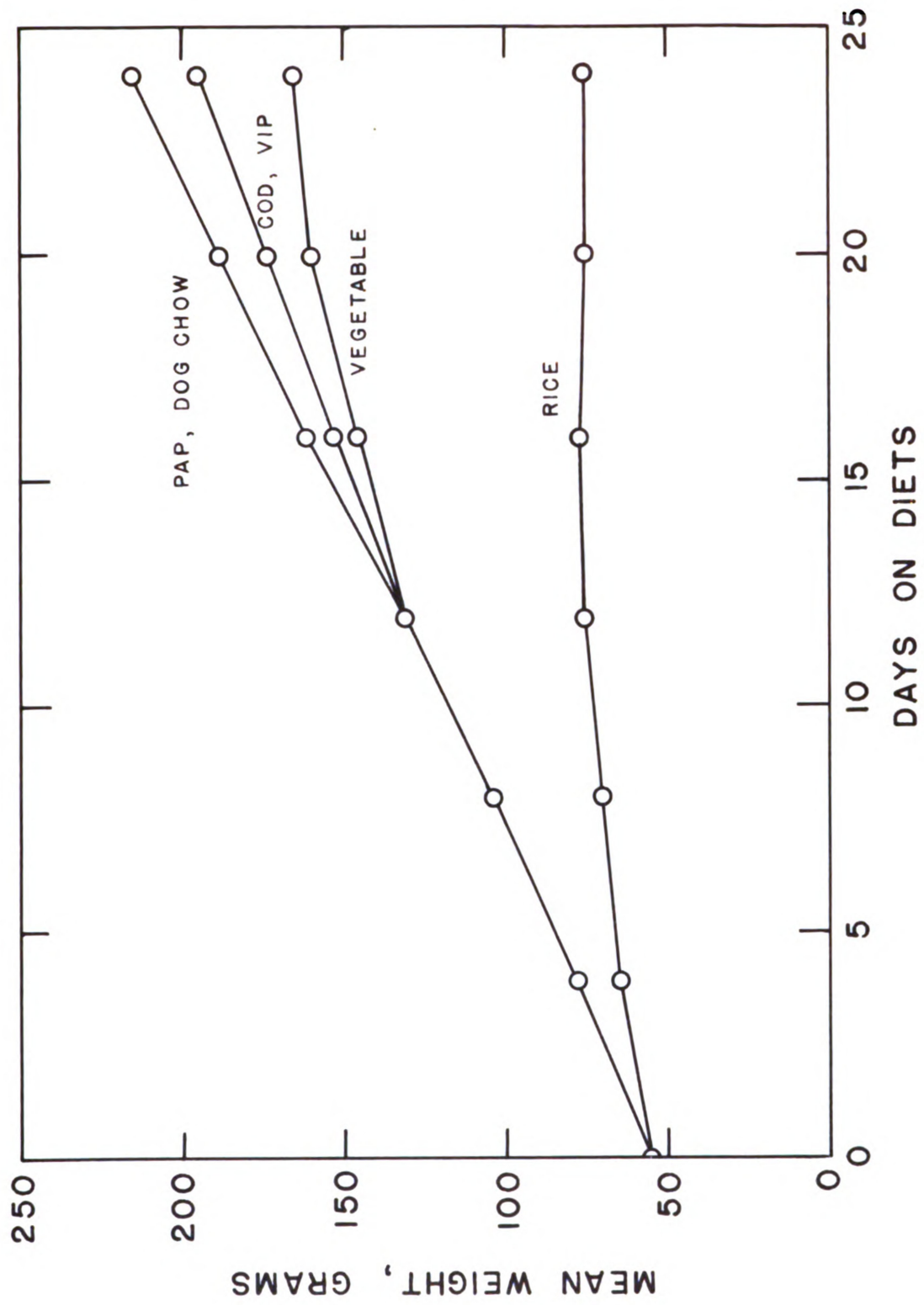


Fig. 5.--Average weights of male weanling rats, Second Feeding Test.

STORAGE AND PALATABILITY

Studies of storage effects on the COD, EGG, and PAP diets are being carried. For this purpose, the diets were freshly prepared; some samples of each diet were air-packed in unsealed Kraft paper bags, some heat sealed in 1 mil polyethylene bags, and some air-/and nitrogen-packed in 211 x 400 cans. A complete set of packaging treatments (all diets and containers) is stored at 35°C, and 3 cans of each diet, air-packed, are stored at 5°C.

Before storage, determinations of moisture, protein, fat, color, rancidity, carotene, and palatability were made on either dry or cooked food, as appropriate. The same determinations, except protein and fat, will be made periodically during a one-year storage period.

Ingredients

Rice: a commercial rice flour of unknown history was used.

Egg powder: stabilized, dry whole egg solids, Anhauser-Busch, Inc.

Cod: prepared, as for the feeding tests, from frozen cod fillets.

Soy flour: prepared, as for the feeding tests, from beans from the 1962 harvest, probably Chippewa variety.

Yeast: chipped yeast, Anhauser-Busch, Inc., strain G.

CaCO₃: a laboratory grade.

NaCl: Diamond crystal salt.

Analytical Procedures

The procedures for moisture, protein, and fat determinations are described in the chapter "Proximate Analysis of Ingredients."

Thiobarbituric acid test (TBA)

Two methods were used: a distillation method and a direct method.

Distillation method (Tarladgis, et al., 1960) - Ten grams of sample were transferred into a 500 ml Kjeldahl flask with 97.5 ml of distilled water. To this was added 2.5 ml 4N HCl (1 + 2 HCl) and a small amount of Dow antifoam A. The distillation was performed at the highest temperature setting and 50 ml of distillate were collected. Active foaming was evidenced. Five ml of distillate were pipetted into a glass stoppered tube and 5 ml of 0.02M TBA solution (TBA dissolved in 90% glacial acetic acid) were added and mixed. The tubes were immersed in a boiling water bath for

35 minutes and then cooled. The absorption measurements were done with a Beckman DU spectrophotometer at 538 $m\mu$.

Direct method (Yu et al. 1957) - One gram of sample was accurately weighed and transferred into a 250 ml boiling flask. To this was added 5 ml each of TBA reagent, distilled water, 0.6N HCl, and 10 ml trichloroacetic acid. The flask was put in a boiling water bath and refluxed for 30 minutes, then 35 ml 0.6N HCl and 30 ml distilled water were added, and the solution refluxed for 10 minutes more. It was then cooled and centrifuged for 10 minutes at 3,000-3,500 rpm. The optical density was determined with a Beckman DU spectrophotometer at 535 $m\mu$. The result is expressed as "TBA number" which indicates milligrams of malonaldehyde per 1,000 gm of sample.

Peroxide number

About 100-110 gm of sample were dried in a oven at 103-105°C for 5 hours. The dried matter was soaked with ether and placed in a cold room for 24 hours or longer. It was then filtered and washed with portions of ether. The ether of the filtrate was evaporated at low heat in a tared, dried 250 ml Erlenmeyer flask, then put in a dessiccator for 24 hours. The flask was weighed to determine the exact weight of the extracted oil. The peroxide number was

determined on this oil, which was dissolved in 25 ml of a glacial acetic acid-chloroform mixture (3:2, by volume). One ml of saturated KI solution was added. The contents of the flasks were swirled for exactly 2 minutes and 50 ml water added. The liberated iodine was titrated with 0.1N thio-sulfate. Starch solution was used as indicator. The peroxide number was calculated as milliequivalents of peroxide per kilogram of oil.

Color determination

The evaluation of color was done by two methods: transmittance and reflectance measurements.

Transmittance measurement - Five grams of sample were soaked with 10 ml acetone CP with occasional stirring for exactly 5 minutes. It was filtered through Whatman No 2 filter paper. The residue was also poured into the filter. It was washed three times with 1-2 ml acetone. Exactly 10 ml filtrate and washing were collected. The transmittance was determined with a Bausch & Lomb Spectronic 20 colorimeter from 400 $m\mu$ to 540 $m\mu$.

Reflectance measurement - The measurements were made of both dry and cooked foods in a Gardner Color Difference Meter. The standard plate used was white ($L = 90.9$; $a = -2.0$; $b = +2.5$) Dry foods were measured without further preparation.

Cooked foods were prepared before measurement as follows.

Ten grams of sample and 100 ml water were put in a 400 ml beaker. A hot plate was turned on "full". When it was hot the beakers containing samples were placed on it for 12 minutes. Occasional stirring was made during cooking. The cooked foods were transferred to a glass petri dish for color measurement.

Carotene

Ten grams of sample were weighed out and 100 ml of an acetone-Skellysolve B mixture (3:7, by volume) were added. It was shaken and let stand for 15 hours or longer in a stoppered flask in the dark. It was then filtered through a Buchner funnel and the residue was washed with two 20 ml portions of Skellysolve B. The filtrate was transferred into a separatory funnel. The acetone was washed from the extract with two or three 100 ml portions of distilled water. The Skellysolve layer was transferred into an Erlenmeyer flask, filtered through anhydrous Na_2SO_4 and evaporated to about 25 ml over a boiling water bath. The solvent was passed through an absorption column containing a mixture of 3 parts Hyflo Supercel and 1 part activated magnesia. The carotene elute was measured with an Evelyn colorimeter. The carotene content was determined from a standard curve made with B carotene.

Palatability

Ten grams of each diet (RICE, VEG, COD, PAP) were cooked with 100 ml water for 12 minutes. A panel of three people tasted the hot cooked foods and gave their opinions.

Results of Analysis and Tasting

The results of the analyses are summarized in Tables 17 and 18.

Table 17.--Physical and chemical determinations of COD, EGG, and PAP diets before storage.

Diet code:		COD	EGG	PAP
Dry food	Moisture, %	9.3	8.1	8.7
	Protein, %	14.8	15.1	15.1
	Fat, %	0.4	4.0	2.2
	Color ^a			
	L	81.9	82.2	81.9
	a	+ 0.3	+ 0.7	+ 0.8
	b	+11.3	+17.2	+13.5
	TBA methods:			
	Distillation	-	0	-
	Direct	0	0	0
Cooked food	Peroxide number, Me/100 g	0	12.9	7.4
	Carotene, mg/100 g	0.870	1.09	0.957
	Color ^a			
	L	59.5	59.7	58.0
	a	- 0.2	- 0.5	- 0.2
	b	+17.0	+18.1	+18.3

^aGardner Color Difference Meter

Table 18.--Per cent transmittance of the acetone extract of
COD, EGG, and PAP diets.

Wavelength, m u.	Diet code		
	COD	EGG	PAP
400	73	52.5	67.5
420	62	36.5	55
430	57.5	32.5	51
440	54.5	27.5	45.5
445	53.5	27	44
450	52.5	26.5	43
455	52	27	43.5
460	52	27.5	44
470	54.5	29	47
480	57	33.5	53.5
500	76	67	76
520	96	96.5	98

Palatability

All the panel members agreed that the texture and consistency of the cooked COD, EGG, and PAP diets were very similar to the rice gruel alone. They also noted the coarse granulation of cod present in the COD and PAP diets.

The diet flavor was that of the principal supplementary component, cod, egg, or soy, but it was not considered to be objectionable by any of the panel members.

The Eastern panel member felt that, although all diets were not salty enough, they were acceptable; moreover, the COD diet had no strong fishy odor, to which there might be some objection. He was very pleased to find no noticeable

fishy odor in the PAP diet. In the opinion of two panel members with Western palates, all diets were very bland and had no off-flavor. However, one member found a slight bitter aftertaste in all diets and one noted a slight, undesirable custard-like taste in the EGG diet.

DISCUSSION OF THE RESULTS

The protein content of the test diets was raised to 15%, which corresponds to about 15-16 per cent calories, and is slightly higher than the level recommended by the British Medical Association and the U. S. allowances. This 15% level has been set to balance the low digestibility of vegetable protein, which forms the biggest part of the protein content of the diet. This protein level seems to be a very good one because it has produced a weight gain equal to that induced by the 24.5% protein level of the control diet.

There is no doubt that the quantity of protein in a food is important. The rats fed on the rice diet having 7.5% protein gained very little weight, and were generally inactive. Three out of six rats on the rice diet showed signs of nutritional deficiency evidenced by hair loss. On the contrary, the rats fed on diets of 15% protein reached a final weight 3 to 4 times their initial weight after a 24-day feeding period.

It is noted that protein alone at a high level is not sufficient. The rats fed on the SOY diet, made up of rice and

soy flour to a 15% protein level, did not grow well. They were better off during the first week. When their internal reserve of nutrients was exhausted their weight gains dropped sharply and they showed signs of nutritional deficiency as evidenced by hair loss and the premature death of one of them which was kept on the diet beyond the 24-day test period. Other groups of rats getting supplements in addition to protein (vitamin A in sweet potato, vitamins B complex in yeast, calcium in calcium carbonate) were much healthier and gained steadily in weight all during the experiment, except the group on the VEG diet, which did not grow as much during the last week of the tests. From this study one must be aware of the limitation in just increasing the quantity of protein in a diet. Other nutrients play an equally important role as protein in the building of a good health.

The protein score calculated by the FAO method (1957) provides a quick insight into the protein value of the mixtures; but it is not accurate because of the involvement of many other factors, either identified or unidentified, as is noted above. A presumably good diet (calculated from chemical score) must be followed by a bioassay. The protein score of rice is 72; that of rice + soy flour is 76, rice + cod 77, rice + pork 82, and rice + egg 99. (Table 12) By

contrast, the bioassay showed that rice + cod and rice + pork had a better protein efficiency, 2.6, than rice + egg, 2.3.

It is found that, when other components were equivalent, soy flour was less nutritious than animal products. The VEG diet produced a weight gain of 3-4 gm a day and a protein efficiency of 1.7-2.0 while rats on the COD, PORK, or EGG diets realized a weight gain of over 6 g/da and a protein efficiency of about 2.5. Hair loss in three rats fed on the VEG diet was noted, whereas no sign of deficiency was shown in any rat fed on animal protein.

In the second test the addition to the VEG diet of inositol and calcium pantothenate (diet code VIP), at the level of 10 mg per 100 g food seemed to favor the growth of rats. The rats on VIP diet obtained a weight gain of 5.75 g/da in comparison with 5.35 g/da for rats on unmodified VEG. Moreover, three out of six rats fed on the VEG diet began to lose weight during the last week of the experiment, while the rats on the VIP diet still gained some weight. High temperature during soy bean processing, 110°C for 30 minutes, or too long a drying time might destroy some B vitamins. This point has to be elucidated in order to improve soy and soy meal for the feeding of man, animal, or poultry.

In both feeding tests the COD and dog chow diets gave the same results; but the results of the RICE and VEG diets were better in the second test than in the first. In particular, the VEG diet showed an appreciable difference: a weight gain of 3.1 g/da and a protein efficiency of 1.7 in the first test in comparison with a weight gain of 4.3 g/da and a protein efficiency of 2.0 in the second.

The plant and animal protein mixture, code PAP, seems to be the best diet tested for a number of reasons. All 6 rats grew just as well as the control group. As expected, the protein efficiency of PAP is better than that of the control diet because of the high protein level of dog chow, 24.5% in comparison with 15% in PAP.

Panel members agreed that the texture and consistency of the cooked COD, EGG, PAP diets were similar to the rice gruel alone. The flavor of fish in the COD diet, of egg in the EGG diet, and of soy bean in the PAP diet was noticeable but not objectionable. The soy flavor masked the fish flavor in the PAP diet. The cod meal, perhaps, should be ground more finely because its coarse granulation was felt during tasting and might be difficult for a child to swallow.

It is concluded that plant and animal products supplement one another very well. The PAP diet is the most

successful and should be considered as a possible baby food. The percentage of fish in it is very small, 4%, and consequently, the PAP diet is expected to be the least expensive of any of the diets having some animal protein.

All the proposed baby foods lack ascorbic acid, which probably must be supplied from fresh fruit. Generally speaking, the stability of ascorbic acid is incompatible with the expected storage conditions and final preparation of these diets. Some consideration might possibly be given to tomato flakes, but it is anticipated that the color and flavor of the resulting mixture would be appreciably affected.

The nutritive value and acceptability of these foods can now best be studied by conducting feeding tests of the more promising diets on weanling infants under the supervision of qualified people, perhaps in a hospital.

Additional careful consideration must be given to the following aspects of the over-all problem before any large scale production is attempted, assuming, of course, that the diets chosen are proven to be nutritionally sound.

- Processes and machines, especially in respect to technical skills and availability of equipment;
- Product availability and production costs;
- Chemical specifications for quality control of the finished product;

-An educational campaign explaining the purpose and use of these foods for the health of the children.

SUMMARY

A number of enriched diets to alleviate nutritional deficiencies in the present diet of Vietnamese babies have been tested. Some of the enriching ingredients were processed; some were procured from commercial sources. The proposed diets use rice, 70-80% of the diet, as the basic ingredient; the protein content was brought up to a 15% level by the addition of soy, cod, egg, pork, or a combination of soy and cod; the addition of sweet potato provided vitamin A, yeast provided vitamin B complex, and calcium carbonate provided calcium.

Two rat feeding assays were carried out. COD, EGG, and PORK diets gave close results in weight gain as well as protein efficiency. The results of the VEG diet were better in the second test than in the first; but they still were much lower than those of the animal protein diets. The addition of inositol and calcium pantothenate seemed to improve the VEG diet. The SOY diet was only a little better than the RICE diet. The RICE diet produced comparable, lowest results

in the two experiments, a very poor weight gain of 0.29-0.44 gm a day and a low protein efficiency of 0.54-0.67.

The plant and animal protein mixture, code PAP, which is considered the best, induced an excellent weight gain of 6.76 gm a day and a protein efficiency of 2.7.

The cooked COD, EGG, and PAP diets had a texture and consistency similar to those of the rice gruel alone. Their flavor was not objectionable.

It is concluded that these enriched foods provide the essential nutrients for the good development of rats and are clearly superior to the rice alone. They might be used as fortified foods for Vietnamese babies with benefit.

Storage stability of the COD, EGG, and PAP diets is currently under investigation.

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