exception of tryptophan. The quantities of amino acids supplied by frankfurters and bologna were usually rated fair; however, bologna proved to be a somewhat better source than frankfurters.

Dark rye flour contains three of the essential amino acids (histidine, isoleucine, and valine) in excellent quantities, and was usually rated as a good source of others. This is in marked contrast to light rye flour, which appeared to be a poor to fair source of the amino acids.

On the basis of 100 grams of the food and food protein, macaroni, noodles, wheat flakes, and shredded wheat were similar in contents of all amino acids, except lysine, which was considerably lower in wheat flakes. Lima beans provided a better source of cystine and valine than either canned baked beans or pork and beans, both on the basis of fresh weight and protein.

Fresh lima beans, baked beans, and pork and beans are similar in their contents of amino acids, being usually classified as poor or fair. This was also true of cornmeal, rice, and cornflakes, though cornmeal rated somewhat higher than the other two in this group.

The need for such evaluations of foods becomes apparent when one attempts to assess the protein quality of foods. When menus cannot be based on protein of excellent quality, planning of dietaries to include, simultaneously, two or more foods—which are classified as good or fair sources of the amino acids—may enable supplementary relationships to play an important role in the maintenance of good nutrition in the individual.

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Received for review July 26, 1957. Accepted October 23, 1957. Research supported in part by a U. S. Department of Agriculture contract, sponsored by the Human Nutrition Research Division, Agricultural Research Service.

NUTRITIVE VALUE OF MILK PRODUCTS

Growth and Reproduction of Rats on Diets of Evaporated Milks and a Vegetable Fat Milk Product

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Growth and reproduction studies were carried out with rats to compare the nutritive value of an experimental vegetable fat milk product with a conventionally processed and a high-temperature, short-time processed evaporated milk. Weanling rats maintained on the milk diets with minerals added grew well for 12 weeks. Second and third generations were successfully reared on the milk diets. Judged by the criteria of weight gain and reproduction performance, the milk product made from nonfat milk solids, vegetable fats, and water, and fortified with vitamins, was somewhat better nutritionally than the other two milks. The poorest performance was shown by the rats fed the conventional evaporated milk, which appeared to be inadequate to meet the needs of female rats during repeated gestation and lactation.

MILK is recognized as an important component of the diet, and its nutritional adequacy in both raw and processed forms has been investigated in several animal species. Raw milk supplemented with iron, manganese, and copper has been demonstrated to be adequate for normal growth and reproduction in rats (*3, 6*) and in dogs (*2*). Results reported by Bixby and coworkers (*3*) indicate that homogenized milk heated for 30 minutes at 165° F., dried whole milk, and a canned whole milk sterilized by a high-temperature, short-time process suffered little if any change in nutritive value.

Conflicting findings have been reported regarding the effects of the usual commercial processes of evaporation, sterilization, and spray-drying of milk. Cook and coworkers (*4*) found by rat growth methods that the nutritive value of milk proteins was slightly reduced by commercial methods of producing evaporated milk. Schroeder, Iacobellis, and Smith (*12*) used a nitrogen balance study in dogs to show that there was no decrease in digestibility or biological value due to the evaporation process. Nitrogen balance studies in rats, carried out by Whitnah (*13*), indicated very small differences in nutritive value between fresh, experimental evaporated, and commercial evaporated milks.

Similarly, Hodson (*8*) found no significant lowering of protein nutritive value in the sterilization of evaporated milk as measured by the rat repletion method, but later (*9*) reported slight differences when rat growth methods were used. In contrast, loss of some protein efficiency was incurred during preheating prior to the spray-drying proc-

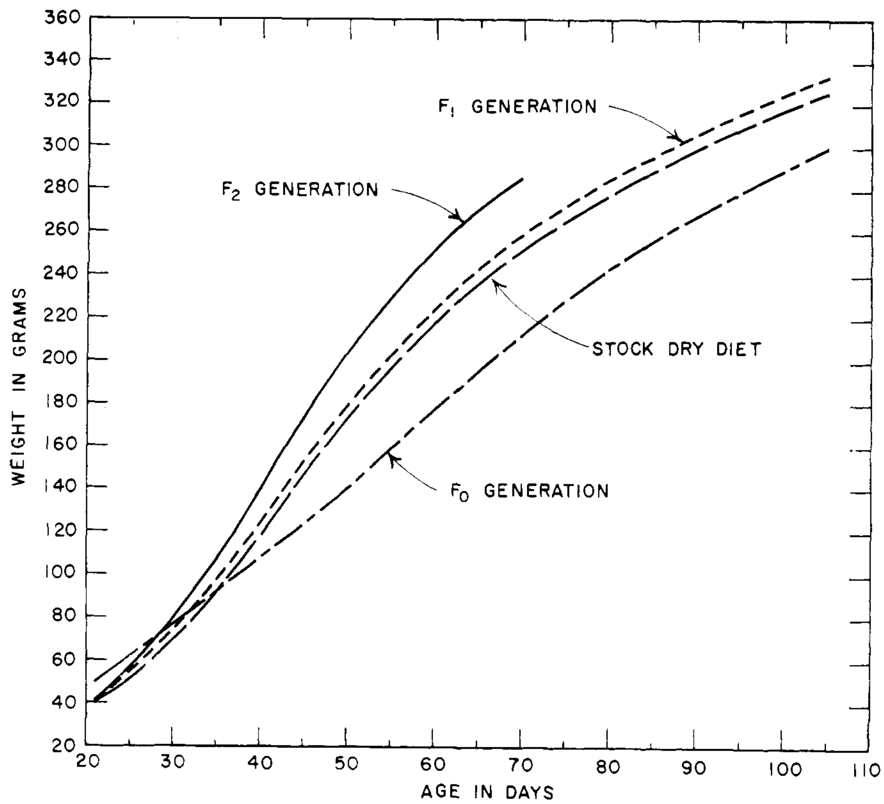

Figure 1. Growth curves of three generations of male rats fed only vegetable fat milk product, compared with colony norm for males on stock dry ration

ess (*4*), even when this preheating corresponded to standard pasteurization time and temperature conditions.

Schantz, Elvehjem, and Hart (*11*) found that 4% butterfat, corn oil, coconut oil, cottonseed oil, or soybean oil homogenized with mineralized skim milk gave good growth results in rats—although the rats on the butterfat milk made somewhat better and more efficient gains than the others during the first 2 or 3 weeks of the experiment. Freeman and Ivy (*7*) noted that rats fed evaporated milk grew more than those fed a "filled" milk containing coconut oil, during a 97-day experiment; the difference being greater during the second half of the experiment than in the first 49 days. In studies recently reported by Dryden and associates (*5*), very little difference was found between the effect of butterfat and vegetable fat on reproduction and lactation in rats, confirming the observations of most previous workers, as cited by these authors.

The investigation reported here was undertaken to assess the nutritive value for rats of a concentrated filled milk containing dry milk solids and vegetable fats, and to compare it with a conventionally processed evaporated milk and a high-temperature, short-time sterilized evaporated milk.

Experimental

Milk Diets. The milk products used in this investigation were manufactured by commercial processes as described below.

The concentrated vegetable fat milk product (VFMP) was prepared by combining spray-dried, nonfat milk solids, vegetable fats, and water. These ingredients were homogenized, sterilized by high-temperature, short-time treatment, and aseptically canned. The vegetable fat used was 90% coconut fat and 10% cottonseed fat, added in sufficient amounts to give approximately 3.95% total fat when diluted with an equal volume of water. The concentrated milk was fortified before canning with approximately 1 mg. of thiamine, 1.3 mg. of vitamin B_6, 1.5 mg. of vitamin A, and 845 U. S. Pharmacopoeia units of vitamin D per liter.

The conventional evaporated milk (CEM) was processed with the usual evaporation stage, fortified with 845 U. S. Pharmacopoeia units of vitamin D per liter, and sterilized in the can at 247° F. for 15 minutes.

The third product (HTST) was produced by the same evaporation stage followed by high-temperature, short-time sterilization and aseptic canning. Vitamin D was added as in the conventional evaporated milk.

The latter two products were derived from the same milk shed, so that they differed principally in the processing conditions. Because of differences in behavior of the nonfat milk solids, milk from a different dairy pool was used to obtain a spray-dried powder with suitable characteristics for manufacturing the vegetable fat milk product.

The three products are compared in Table I with respect to their vitamin content. These values are averages of determinations made on three different lots of each milk used during the present study, and are generally in agreement with values that have been obtained repeatedly on these products in control testing. Values for fresh whole milk are included for comparison.

To avoid excessive storage periods, new lots of milk were prepared at intervals of 3 to 4 months during the experiment. Storage periods were thus kept well within the 12- to 14-month storage time on which there seems to be agreement that no loss of nutritive value occurs (*4*, *13*).

The milks were diluted with water (about 1 to 1) to approximate whole milk concentration before feeding. The dilutions were calculated on the basis of nitrogen determinations to give equal protein concentrations in the three products as fed. Each diet was supplemented at the time of mixing with 30 mg. of iron, 1.5 mg. of copper, and 1.5 mg. of manganese per liter of milk, as ferric ammonium citrate, copper sulfate, and manganous sulfate. The diets were prepared daily and fed *ad libitum* once a day in drinking bottles of a type usually used for providing water. With this system

Table I. Comparison of Vitamin Content of Processed Milk Products and Fresh Cow Milk

Vitamin and Unit/100 Ml. Whole Milk Basis[a]	*CEM*	*HTST*	*VFMP*	*Mature Cow Milk*[b]
Vitamin A, total, USP units	142	131	232	(190)[c]
Thiamine, γ	26.5	27	57	43
Riboflavin, γ	207	190	210	158
Niacin, γ	128	100	99	85
Vitamin B_6, γ	32.5	29	80	48
Pantothenic acid, γ	350	355	323	350
Choline, mg.	16.8	17	14.9	13
Vitamin B_{12}, γ	0.067	0.103	0.207	...

[a] As diluted for feeding in this experiment.
[b] (*10*) Table V.
[c] Calculated from values for preformed A and carotenoids.

no problems of spoilage or creaming were encountered.

Growth Study. The experimental animals were male and female weanling Long-Evans rats obtained from Simonsen Laboratories, Gilroy, Calif., where a homogeneous breeding colony has been maintained for over 5 years without any admixture from other sources. The 21-day-old weanlings were fed a stock diet for 3 days, during which the milk diets were provided in lieu of drinking water. The weight range was narrowed at the end of this time so that each test group was made up of 10 males in the range from 60 to 68 grams, averaging 63.3 grams, and 15 females in the range from 50 to 64 grams, averaging 56.7 grams. From the fourth day on, only the respective milk diets were fed. A control group of six males and six females was reared on a dry stock diet (Purina Lab Chow) to provide normal tissues for histopathological comparison.

The rats were caged individually in suspended wire-bottomed cages in air-conditioned quarters. Individual body weights of all animals were recorded twice weekly until the rats were 15 weeks old. Daily records of milk intake were kept for three 10-day periods during the experiment. The first 10-day record was begun as soon as the rats were placed on all-milk diets. The second and third 10-day records were made during the second and third months, respectively.

Reproduction Study. After the 12-week growth study was completed, the female rats in each diet group were mated with males of the same group. At each mating one male was caged with three or four females for 1 week and then removed. When the weight gain of a female indicated pregnancy, she was transferred to a large individual cage with clean wood shavings for nesting material. The young were counted on the first day after birth. On the second day all of the live young were weighed and litters of more than eight were reduced to eight. The young were weighed again at weaning, 21 days after birth. Throughout the reproduction study, both the adults and young were fed only their respective milk diets.

The first litters were discarded after weaning, and the females mated again after a rest of 10 days or more. Of the second litters, 15 females and 10 males in each diet group were kept after weaning and reared on the milk to 15 weeks of age. The cycle was repeated until the third generation second litter young were reared to 10 weeks of age. A departure from the usual mating routine was made in producing the final litters, when all the females were mated to males of one group rather than to those of their respective milk diet groups.

After the first reproduction cycle was completed, two males and two females randomly selected from the stock diet group and from each milk diet group were sacrificed and examined for gross pathological changes. Histological sections of the liver, kidney, and large and small intestine were prepared and stained for micropathological examination.

Table II. Summary of Growth Responses of Three Generations of Rats Maintained on Milk Diets

Generation and Diet Group	Number[a] and Sex of Rats	Age, Weeks: 3	5	7	9	11	13	15	Standard Deviation of Final Weights
		Average Body Weights, G.							
F_0									
CEM	9 M[b]	50	88	121	163	200	233	263	±69.3
	13 F[c]	48	79	116	144	161	171	190	21.9
HTST	9 M[b]	50	89	127	170	201	233	259	46.5
	15 F	48	84	120	148	169	186	202	31.4
VFMP	10 M	50	91	136	191	231	272	302	26.2
	15 F	48	88	136	158	179	200	220	24.8
F_1									
CEM	10 M	29	83	149	212	256	294	319	31.3
	13 F	29	73	123	158	182	201	213	14.5
HTST	10 M	42	92	176	250	291	325	355	37.5
	15 F	35	82	129	161	182	200	215	18.9
VFMP	10 M	40	97	173	237	277	309	334	36.2
	15 F	37	84	130	160	185	203	215	18.9
F_2									
CEM	9 M	33	84	152	216	238	(10 weeks)		24.0
	9 F	31	76	122	153	166			12.3
HTST	10 M	42	109	178	244	263			27.9
	10 F	41	97	140	165	173			11.9
VFMP	10 M	42	109	193	265	286			25.8
	10 F	41	97	147	180	190			18.1

[a] Number of animals that completed growth period.
[b] Does not include one animal that died during period.
[c] Does not include two animals that died during period.

Table III. Average Daily Milk Intake of Rats for Three 10-Day Periods

Diet Group	Number and Sex of Rats	Age of Rats during Test Period, Days: 28–38	48–58	77–87
		Ml./Rat/Day		
CEM	9 M	49.9	65.4	90.4
	13 F	50.9	65.0	73.3
HTST	9 M	52.0	66.0	85.5
	15 F	53.6	67.1	77.3
VFMP	10 M	52.4	82.5	104.1
	15 F	60.0	75.2	82.0

Results

Growth and Milk Intake. The average body weight gains for all three generations of milk-fed rats are summarized in Table II. In the parent generation, the weight gains of the rats fed the vegetable fat milk product exceeded those of the rats fed either the conventional or the high-temperature, short-time process evaporated milks, for both males and females. This was true also of the third generation animals. In the second generation, the average weight gain of the males on the vegetable fat milk product diet was midway between those of the other two milk groups, while the average weight gains of females were nearly identical on the three milk diets. Weight gains and final body weights were higher in the second and third generations on all three milk diets than they were in the first generation, in spite of starting with lower body weights at weaning. A graphic comparison of the growth curves for three generations of male rats fed vegetable fat milk product is given in Figure 1. A normal growth curve based on colony experience with large numbers of male Long-Evans rats from the same source reared on dry stock ration is included for comparison.

The average daily milk intake of males and females of the first generation in each diet group are given in Table III for each of the three test periods. Differences between the groups generally parallel the differences noted in weight gains.

Reproduction Performance. Table IV shows the performance of female rats in the study of two reproduction cycles of two litters each. The most significant differences were with respect to conception rate and weight of young at

Table IV. Summary of Reproduction and Lactation Data for Two Generations of Female Rats Fed Mineralized Milk Diets

Generation and Diet Group	No. of Parent Females	Fraction Conceiving at Mating, 1st	2nd	3rd	Average Litter Size	Young Weaned, %	Average Weight of Young, G., 2nd day		Weaning	
F_1, first litter							All		Male	Female
CEM	13	6/13	3/7	4/4	9.1	65.0	6.6		32.3	29.0
HTST	14	7/14	6/7	1/1	9.4	61.1	6.7		33.0	30.3
VFMP	15	11/15	3/4	1/1	9.0	79.2	6.7		37.3	34.9
F_1, second litter										
CEM	12	8/12	2/2	...	9.1	50.0	6.5		28.4	29.3
HTST	14	9/14	1/2	...	10.0	85.5	6.6		35.7	32.9
VFMP	14	10/14	2/2	...	9.3	95.0	6.9		38.1	36.1
F_2, first litter							Male	Female		
CEM	13	7/13	6/6	...	10.0	58.0	7.2	6.4	38.6	35.5
HTST	15	13/15	2/2	...	9.3	80.2	7.0	6.5	39.9	39.1
VFMP	15	13/15	2/2	...	9.5	98.3	7.1	6.6	41.5	41.2
F_2, second litter										
CEM	12	10/12	2/2	...	10.8	86.4	6.6	6.5	31.0	30.0
HTST	15	12/15	2/3	...	9.7	91.0	6.2	6.1	40.2	41.0
VFMP	15	15/15	...	...	10.0	88.6	7.1	6.4	42.3	41.1

weaning. In general, the females fed conventional evaporated milk required the most rematings to secure conceptions, and those fed the vegetable fat milk product the least. Weaning weights in the former group were lower throughout the reproduction study than those in the other two groups. In the latter groups, the second litters produced in each generation had weaning weights equal to or more than those of the corresponding first litters, while in the group fed the conventional evaporated milk the reverse was true. This was especially pronounced in the F_2 litters, when the range of body weights of weanlings in the conventional evaporated milk group did not even overlap with that of either of the other two groups.

Condition, Mortality, and Pathology. The general condition of all rats on the milk diets was good throughout this study, with the exception of four animals in the parent generation that developed severe diarrhea early in the experiment and one female in the second generation that suffered the same affliction. No differences in condition or coat appearance attributable to the diets were noted.

Mortality in all three milk-fed groups during the initial growth study totaled four animals. Three of these were in the group fed the conventional evaporated milk and one in the high-temperature, short-time process group. All four deaths were attributed to the severe diarrhea noted above. One second-generation female in the conventional evaporated milk group died during her first lactation period from undetermined causes. During the growth study with the parent generation moderate diarrhea was present in the majority of the rats in all milk diet groups early in the experiment but did not persist beyond the second week except in the four animals that failed to recover. Diarrhea, even of moderate degree, was almost entirely absent among the second and third generation rats.

No gross pathological changes were observed in any of the animals sacrificed for tissue specimens. The rats on milk diets showed more deposition of fat in the viscera than was seen in the animals fed the stock diet. This was to be expected in view of the higher fat content of the milk diets. Histological sections of the livers, kidneys, and small and large intestines, prepared in a routine manner and stained with hematoxylin and eosin, revealed no significant pathological alteration in any of these organs. Evidence of fat globules was seen in most of the livers and within the lumen of the proximal and distal convoluted tubules of the kidneys. These changes were minimal and there was no significant alteration in the structure of the liver or of the epithelial structure of the tubules or the glomeruli of the kidneys. No pathological alterations were seen in the sections of large or small intestine.

Discussion

According to these results, a concentrated milk product prepared from nonfat milk powder, vegetable fats, and water, and fortified with vitamins, is nutritionally equal or superior to the conventional in-can-sterilized evaporated milk or the high-temperature, short-time sterilized, aseptically canned evaporated milk, when judged by the criteria of growth and reproduction performance of rats fed *ad libitum* on the mineralized milk diets. As the milk solids in the vegetable fat milk product underwent somewhat more severe heat treatment than did the high-temperature, short-time sterilized evaporated milk, the spray-drying process, followed by preheating and sterilizing of the vegetable fat milk product, did not impair the nutritive value.

The better over-all performance of rats on this milk product may be attributable to the significantly higher content of vitamin A, thiamine, vitamin B_6, and vitamin B_{12} in this milk, although the lowest values in the other milks in each case are well above minimum requirements. Comparison with "adequate allowances" cited by Albritton (*1*) shows that the vitamin A intake of 0.34 mg. per kg. of body weight in the poorest case (high-temperature, short-time sterilized milk) was about 35 times more than required, or well above the level for storage. Lowest thiamine intake on the conventional evaporated milk and high-temperature, short-time sterilized milks was 0.34 mg. per kg. of body weight in the lactating females, some seven times the requirement for lactation. Similarly, the minimum intake of vitamin B_6 was 0.39 mg. per kg. of body weight in lactating females as compared to an "adequate allowance" of 0.05 mg. per kg. Encountering any deficiency of these vitamins seems unlikely.

The vegetable fat milk product was significantly higher in vitamin B_{12} as a result of supplementation in manufacturing, but Bixby and associates (*3*) found no effect on rat growth rate from supplementing mineralized cow's milk with vitamin B_{12}. On the other hand, the heat treatment applied in the conventional in-can-sterilization process may have had a deleterious effect on nutritive value. The growth rate of the rats fed the conventional evaporated milk was not significantly affected, but the over-all performance in reproduction and lactation was poorer in this group as judged by fertility, weaning rates, and weights of young at weaning, even in comparison with the rats fed the high-temperature, short-time sterilized milk, which was derived from the same milk pool and was essentially equal in other respects than the processing conditions.

Replacement of the butterfat with vegetable fats did not apparently affect nutritive value in the product investigated here. Bixby and coworkers (*3*) stated that removing the butterfat and replacing it with corn oil depressed the growth rate of rats, but no data were given or references cited on this point.

The results of the current study differ from those of Freeman and Ivy (*7*), both with respect to the weight gains of rats fed the vegetable fat milk product compared to those fed conventional evaporated milk and with respect to the occurrence of diarrhea. The three products used in the present study had equal fat contents, 7.90% before dilution, whereas the filled milk used by Freeman and Ivy (*7*) contained only 6% fat as compared to the 7.74% fat in their evaporated milk, introducing a substantial caloric difference that might have influenced the deposition of body fat and consequently the weight gains. Nevertheless, these authors associated the relatively poorer growth in their filled milk group in the second half of the experimental period with the high incidence of loose stools, and suggested that failure of the intestinal tract to adapt to the vegetable fat might account for the observations.

With three generations of rats in this study, the only difficulty in adapting to the diets was among the first generation animals that had been weaned to a dry ration before starting on the milk diets, and the incidence of loose stools was higher among the rats getting butterfat in the milk than among those getting the vegetable fat. The condition improved markedly in all groups after the first few weeks of the experiment. The litters of the second and third generations had the respective milk diets available *ad libitum* during the nursing period, and made the transition uneventfully from mother's milk to the respective diet milk.

These results are not necessarily inconsistent with those of other workers. The data showing a loss of protein efficiency in evaporated milks indicate only about 10% loss, and this is manifested only under critical conditions in the rat, when the requirement for cystine, cysteine, and methionine becomes the limiting factor (*9*). Under the *ad libitum* feeding conditions in the present experiment, differences in protein nutritive value were not reflected significantly in the growth responses of the rats.

The greater physiological stress imposed on the females by repeated pregnancy and lactation provides a more critical measure of the nutritional adequacy of the diets. The conventionally processed milk appeared to be nutritionally less adequate than the other two milks, while the vegetable fat milk product appeared to be the most adequate, although statistical study of the differences with respect to the individual criteria of conception rates, weaning rates, weaning weights, and weight gains does not show consistently significant differences.

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Received for review July 15, 1957. Accepted October 1, 1957. Investigation supported by Foremost Research Foundation, San Francisco, Calif.

TOXICITY IN MEAT SCRAP

Effect of Trichloroethylene-Extracted Meat Scrap on Young Cattle

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Calves fed meat scrap produced by azeotropic extraction-dehydration with trichloroethylene developed a moderate to severe thrombocytopenia and relative lymphocytosis and, in one instance, leucopenia. These effects are similar to those induced in young cattle by feeding certain specimens of trichloroethylene-extracted soybean oil meal. Development of toxic properties in cattle feeds processed with trichloroethylene is not restricted to soybeans.

The extraction of oil-bearing seeds, particularly of soybeans, with trichloroethylene has been initiated at various times in several countries, but the attractive features (*16*) of this process have been overshadowed by the almost invariable occurrence of cases of fatal aplastic anemia when the extracted residues were fed to cattle (*12*). For this reason, commercial production of trichloroethylene-extracted soybean oil meal has apparently been abandoned,

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