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Protein-Energy-Requirement Studies in Developing Countries: Results of International Research

Edited by William M. Rand, Ricardo Uauy, and Nevin S. Scrimshaw

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25. THE EFFECT OF DIFFERENT INTAKES OF CALORIES ON THE NITROGEN INTAKE FOR NITROGEN EQUILIBRIUM WITH HABITUAL DIETS BASED ON CORN AND BEANS

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Objectives

1. To determine the effect of energy intake on nitrogen intake for nitrogen equilibrium.
2. To determine the nitrogen needs for nitrogen equilibrium in three rural diets of different protein quality.

Experimental Details

Subjects

A total of 24 healthy young adult male subjects were used in two studies. In study 1, 18 subjects were involved, while 10 subjects participated in study 2. All subjects performed their usual chores. Their characteristics are described in table 1.

Study Environment

The subjects lived in their homes in Guatemala City and worked at INCAP. All of their meals were eaten in the Metabolic Unit of the Division of Food and Agricultural Chemistry. The daily ambient temperature ranged from 21° to 25° C. Relative humidity ranged from 72 to 85 per cent. Guatemala City is 1,510 m above sea-level.

Experimental Diets

Based on actual dietary intakes of rural adult Guatemalan populations, a series of basal diets were formulated for each level of protein intake, as shown in table 2. These diets provided from 2,217 kcal/day when protein intake was practically 0 kg/day to 720 kcal/day when protein intake was equivalent to 0.7 g P/kg/day. Calculations of actual dietary intakes in rural areas showed caloric intake to be equivalent to 36 kcal/kg/day. Therefore, the protein intake and the differences in energy intake were made up from diets based on lime-treated common corn, lime-treated opaque-2 corn,

TABLE 1. Physical Characteristics of the Subjects (36 kcal/kg/day)

Study	Group	Subject	Age (yrs)	Height (cm)	Weight (kg)
1	Common corn	J.M.	27	165	66.0
		J.P.	27	167	62.7
		H.R.	24	158	62.0
		A.G.	32	168	59.0
		G.C.	23	168	58.0
		J.A.S.	22	156	48.2
	Opaque-2 corn	M.N.	26	170	70.0
		R.S.	24	158	64.2
		O.B.A.	28	159	61.8
		J.A.A.	30	168	60.0
		R.C.	27	164	51.8
		O.H.	25	168	48.2
	Common corn plus chicken	H.G.	27	170	65.0
		J.R.	35	170	65.0
		C.E.	30	168	62.0
		O.B.O.	25	162	61.3
		A.O.S.	19	160	51.5
		J.D.V.	38	153	50.9
2	Common corn	F.S.	23	168	52.0
		V.D.	20	152	52.0
		L.S.	27	158	55.6
		A.L.	33	162	57.3
		W.J.S.	25	168	60.5
		E.M.	22	160	61.1

beans, and poultry meat, as shown in table 3. It was necessary to feed a few individuals who had a higher weight additional calories. In study 2, the level of calories was 45 kcal/kg/day and only the common-corn diet was assayed.

Experimental Design

Protein Quality Assay. Short-term Nitrogen Balance Method

After a four-day adaptation period with a high-quality diet fed at 0.6 g protein/kg/day and 36 kcal/kg/day, the subjects were fed a low-nitrogen diet for three days. The low-nitrogen diet is that shown in table 2, but all protein sources were replaced with cassava. During the last two days, quantitative faecal and urine collections were made. The low-N diet was followed by feeding the experimental diets in study 1 to provide

TABLE 2. Food Intake. Basal Diets (g/day)

Food	0.0	0.2	0.4	0.6	0.7
Instant coffee	3	3	3	3	3
Apple marmalade	40	40	30	30	—
Cassava patty	615	—	—	—	—
Sugar	30	30	30	30	25
Soup[a]	400	400	400	—	—
Guisquil	300	—	—	—	—
Orange	100	100	100	100	100
Banana	50	50	50	50	50
Vegetable oil	80	25	36	36	36
Artificial flavoured drink (glasses)	3	4	3	2	—
Margarine	—	20	20	20	20
Sweet bread	—	40	40	40	40
Cooked rice	—	100	100	100	100
Raw tomatoes	—	100	100	100	50
Raw cabbage	—	100	100	100	50
Boiled potatoes	—	50	50	50	50
Analysed nitrogen (g/day)	1.575	1.866	1.639	1.730	1.603
Calculated calories[b]	2,217	1,550	1,264	1,264	720

a. Made with corn starch, herbs, margarine.
b. Adjusted to 36 kcal/subject with the experimental diets (table 3) and with N-free food items (candies, soft drinks).

TABLE 3. Average Food Intake (g/day). Protein Sources

Group		(g protein/kg/day)			
		0.2	0.4	0.6	0.7
Common corn	Tortilla	143.3	286.4	429.5	501.1
	Bean powder[a]	15.4	31.1	46.5	54.4
Common corn plus chicken	Tortilla	103.8	206.9	311.4	363.3
	Bean powder	11.2	23.4	33.6	39.0
	Chicken[b]	12.9	25.9	38.8	45.3
Opaque-2 corn	Tortilla	140.4	281.7	422.0	492.0
	Bean powder	15.5	31.1	46.5	54.3

a Served fried with vegetable oil and salt, added before frying.
b. Boiled chicken deboned and chopped.

TABLE 4. Summary of Nitrogen Retention at Various Levels of Nitrogen Intake of Different Dietary Treatments

Basal Diet Variable Component	Energy Intake (kcal/kg/day)	Subjects	Average N Intake (mg/kg/day)				
Common corn	36	6	26.5 ± 2.4[a]	64.1 ± 3.8	93.0 ± 3.2	127.1 ± 3.4	141.3 ± 3.2
			− 50.3 ± 9.4[a]	− 2.8 ± 6.1	3.6 ± 11.1	19.0 ± 18.7	28.8 ± 8.0
Opaque-2 corn	36	6	26.5 ± 2.1	64.4 ± 03.7	93.4 ± 3.5	127.5 ± 3.5	141.3 ± 3.0
			− 58.7 ± 12.4	− 13.7 ± 12.5	0.1 ± 10.9	17.7 ± 20.6	29.4 ± 27.2
Common corn plus chicken	36	6	27.7 ± 1.6	64.8 ± 4.5	93.7 ± 4.1	128.5 ± 4.7	143.2 ± 5.2
			− 47.2 ± 14.0	− 5.6 ± 15.2	6.1 ± 16.3	21.4 ± 20.0	23.0 ± 22.5
Common corn	45	6	22.8 ± 1.8	47.0 ± 0.9	88.6 ± 1.4	127.6 ± 1.4	—
			− 74.3 ± 14.1	− 44.6 ± 11.3	9.8 ± 7.9	14.8 ± 20.1	—

a. Standard deviation.

TABLE 5. Regressions between Nitrogen Intake and Nitrogen Retained of Rural Diets made with Common Corn, Opaque-2 Corn, and Common Corn with Chicken

Rural Diet	Calorie Intake (kcal/kg/day)	Number of Subjects	Number of Observations	Regression Equations	r^2 (%)
Common corn	36	6	30	NR = − 58.07 + 0.64 Ni P (B) ≤ 0.001	70.9
Opaque-2 corn	36	6	30	NR = − 70.54 + 0.72 Ni P (B) ≤ 0.001	70.1
Common corn and opaque-2 corn	36	12	60	NR = − 64.29 + 0.68 Ni P (B) ≤ 0.001	73.0
Common corn plus chicken	36	6	30	NR = − 79.84 + 1.38 Ni − 0.0047 Ni^2 P (B) ≤ 0.003 P (B) ≤ 0.05	70.4
Common corn	45	6	24	NR = − 123.30 + 2.23 Ni − 0.0089 Ni^2 P (B) ≤ 0.000091 P (B) ≤ 0.003	87.1

TABLE 6. Regressions between Nitrogen Retained and Weight Losses of Subjects Fed Diets Made with Common Corn, Opaque-2 Corn and Common Corn with Chicken at 36 kcal/kg/day

Rural Diet	Weight Losses	Number of Subjects	Number of Observations	Regression Equations	r^2 (%)
Common corn	High	3	15	NR = − 61.550339 + 0.67957 Ni P (B) ≤ 0.00044	65.6
	Low	3	15	NR = − 84.354209 + 1.544372 Ni − 0.005529 Ni^2 P (B) ≤ 0.001945 P (B_2) ≤ 0.027418	86.6
Opaque-2 corn	High	3	15	NR = − 89.596578 + 1.458469 Ni − 0.003984 Ni^2 P (B) ≤ 0.000921 P (B_2) ≤ 0.051018	93.1
	Low	2	10	NR = − 59.838841 + 0.504027 Ni P (B) ≤ 0.00316	69.2
Common corn plus chicken	High	3	14	NR = − 108.646796 + 2.379523 Ni − 0.011026 Ni^2 P (B) ≤ 0.001811 P (B_2) ≤ 0.007086	75.6
	Low	2	10	NR = − 60.158831 + 0.664204 Ni P (B) ≤ 0.00027	86.0

TABLE 7. Regressions between Nitrogen Retained and Weight Losses of Subjects Fed Diets Made with Common Corn at Two Different Calorie Intakes

Rural Diet	Weight Losses	Number of Subjects	Number of Observations	Regression Equations	r^2 (%)
45 kcal/kg/day Common corn	Gain	2	8	$NR = -126.80957 + 2.581358\ Ni - 0.011596\ Ni^2$ $P\ (B) \leq 0.001391\ P\ (B_2) \leq 0.004855$	97.2
	Loss	2	8	$NR = -162.147021 + 3.197631\ Ni - 0.014768\ Ni^2$ $P\ (B) \leq 0.013716\ P\ (B_2) \leq 0.041228$	89.6
36 kcal/kg/day Common corn	High	3	15	$NR = -61.660339 + 0.67957\ Ni$ $P\ (B) \leq 0.0004$	65.6
	Low	3	15	$NR = -84.354209 + 1.544372\ Ni - 0.005529\ Ni^2$ $P\ (B) \leq 0.001945\ P\ (B_2) \leq 0.027418$	86.6

0.2, 0.4, 0.6, and 0.7 g P/kg/day. In study 2 the highest protein level fed was 0.6 g P/kg/day. These levels of protein were given for two days each, with quantitative faecal and urine collections every 48 and 24 hours, respectively.

At the end of each experiment, the samples of food, faeces, and urine were analysed for nitrogen. The results were analysed statistically by linear regression to estimate protein needs for nitrogen equilibrium and estimation of protein requirement.

Summary of Main Results

Table 4 summarizes the average nitrogen balance in diets fed at 36 kcal/kg/day and the common corn/bean diet at 45 kcal/kg/day.

The statistical analysis carried out for the three diets made with common corn, opaque-2 corn, and common corn plus chicken at an intake of 36 kcal/kg/day indicated no difference between the opaque-2-corn and common-corn diets, but the common corn plus chicken was different from the other two because it showed a significant quadratic component, as indicated in table 5. As calorie intake was low, all subjects lost weight. The diet with chicken at 0.7 g P/kg/day was no longer giving a linear response, while the other two at this level did show a linear response, suggesting that protein was being used as a calorie source.

Statistical analyses were made between the weight changes of individuals, and the relationship between nitrogen intake and nitrogen retention for each dietary treatment. These are shown in tables 6 and 7.

Within each dietary treatment, the individuals were divided into two groups: those who lost or gained more weight and those who gained or lost less weight. At a calorie intake of 36 kcal/kg/day all individuals with all diets lost weight; however, those eating common corn with a higher weight loss showed a linear regression, while those with a lower weight loss showed a significant quadratic component. The opposite was true according to the statistical analysis for subjects with greater weight loss as compared with those with lower weight loss on diets of higher protein quality, that is, that of opaque-2 corn and that with common corn plus chicken. With respect to the common-corn diet fed at 45 kcal/kg/day, some subjects gained weight while some lost weight. In this case, with "adequate calories" both groups showed a significant quadratic component.

Conclusions

The interpretation of the regression equations suggests different methods of adaptation to the experimental conditions imposed with respect to calorie intake and its interaction with protein quantity and quality in each diet.