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Time Delayed Protein Complementation

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ABSTRACT

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by

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The time delayed complementation of plant proteins was tested in weanling Sprague-Dawley male rats by measuring growth rates as well as plasma and tissue free amino acids. Diets of pinto beans with rice, corn or wheat were pair-fed in combination or alternating 4 times a day at 6 and 11 am, and 4 and 9 pm for 4 weeks. Plasma, liver and muscle samples were obtained 2 hours postprandial, a time determined most suitable for sacrifice, and analyzed for free amino acids via ion-exchange chromatography. There were no significant differences in growth of rats fed combination or alternating diets during week 3 and 4 with the exception of the limiting corn-bean combination and alternating diets in week four. Step-wise multiple regression of the data show plasma free amino acids most correlated to growth included asparagine, 15.1% contribution; and arginine, tyrosine, histidine, 1-methylhistidine and cystathionine, each contributing <5%. The following amino acids were negatively correlated with growth: threonine, 29.2%; and leucine, phosphoethanolamine, taurine and serine <6.5%. This study confirms previous work in this laboratory which suggested

that vegetable proteins fed 5 hours apart in combined or alternating fashion had equal growth promoting capacity. Contrary to expectations, the limiting amino acids in these diets -- methionine, lysine and tryptophan -- were not the postprandial plasma amino acids most related to growth rate. More study is necessary to correlate the limiting amino acids in proteins and tissue free amino acids with the growth rate of the rat.

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TIME DELAYED PROTEIN COMPLEMENTATION

by

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Each person whose signature appears below certifies that this manuscript in his opinion is adequate, in scope and quality, in lieu of a thesis for the degree Master of Science.

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INTRODUCTION

Early studies on delayed amino acid supplementation suggest that supplements of the limiting amino acids to deficient diets are most efficiently utilized for growth in animals fed the supplement together with the meal (1, 2). It is recognized that all amino acids must be simultaneously present in rat tissues in appropriate amounts for optimal protein synthesis (3). However, it is believed that amino acid supplements may be given within 4 to 5 hours of the deficient meal, since there is evidence that amino acids may be stored for short periods of time (3, 4). Successful delayed amino acid supplementation of 6 hours for threonine (5) and 16 hours for lysine (6) have been reported in rats. Recently, Baker and Izquierdo (7) showed that chicks fed lysine deficient diets of corn and sesame seed meal also had comparable weight gains whether given a lysine supplement mixed with the diet or a diet containing a double dose of lysine for 12 hours followed by no lysine supplementation for 12 hours. The available data (5, 6, 8 - 10) support a delayed time supplementation premise. On the other hand, methionine has shown a supplementary effect of only 1 hour after a meal (9, 11). Therefore, the time delay for amino acid supplementation must depend upon the specific most limiting amino acids.

A few studies in the past have tested the possible delayed complementation of proteins (12 - 15). Recent reports show the delayed complementary effects of plant proteins (16, 17) but very few selected protein combinations have been studied. Mills and Canolty (16) found an inverse relationship between complementary capacity and time when using time intervals of 0, 1, 2 and 3 days. Sanchez et al. (17) used time intervals of 5 h and found comparable growth promoting capacity in rice and mung beans fed in combination or alternating.

Growth rate of animals has almost exclusively involved the measurement of delayed protein complementation and delayed amino acid supplementation. It is recognized that all amino acids must be simultaneously present in the tissues in appropriate amounts for optimal protein synthesis. Therefore, we assume that amino acid levels in tissues vary with the rate of growth and may elucidate the mechanism for the phenomenon of delayed protein complementation. The purpose of this study was to test time delayed complementation in animals fed different commonly used cereal-legume protein combinations and to identify the levels of amino acids most conducive to growth by measuring amino acid concentrations in blood and tissues samples.

METHODOLOGY

Animals

One hundred male, weanling, albino Sprague-Dawley rats were obtained from Harlan Sprague Dawley and housed in individual raised bottom stainless steel cages at the Loma Linda Medical Center Animal Care Facility. The room was maintained at 70 - 72° F, 40 - 50 % humidity with complete darkness for 16 hours per day. Prior to the feeding of the test diets, the rats were placed on a standard rodent diet (Purina Rodent diet #5001) for 5 - 7 days. The rats were randomly divided into diet groups of 10 animals each and weighed. The rats were pair-fed 4 times daily and water was allowed ad libitum.

Diets

Pinto beans were obtained in the precooked powdered form from Inland Empire Foods. Polished white rice was obtained from Capitol Milling in a powdered form. Powdered wheat and corn were purchased from Centennial Mills and Giusto's Specialty, respectively. To increase the protein content of the rice and corn diets, the sodium hydroxide (0.2%) extraction procedure of Fujita et al. (18) was used. The protein contents of each diet was determined by the standard Kjeldahl method described by the Association of Official Analytical Chemists (AOAC) (19).

Table 1 shows the composition of the diets. American Institue of Nutrition (AIN) Vitamin Mixture 76 and AIN Mineral Mixture 76 were obtained from U.S. Biochemicals. Alphacel was obtained from ICN Nutritional Biochemicals. Each diet was mixed in the Hobart vertical cutter mixer (Hobart, model VCM25), analyzed for protein content and stored at 4^{o} C.

Experiments

A preliminary study was conducted to determine the postprandial sacrifice time for the 100 pair-fed rats in the delayed protein complementation study. For this, plasma free amino acids were measured at fasting and at 1, 2 and 4 h postprandial (Figure 1).

Ten groups of 10 rats each, were fed the test diets 4 times daily for one hour with 4 hour intervals between meals. Groups 1, 2, 3 and 4 were given the pinto-bean, rice, corn and wheat diets, respectively for each meal. Groups 5, 6 and 7 received combination diets of rice-bean, corn-bean and wheat-bean, respectively. Groups 8, 9 and 10 were given diets of pinto-bean alternating with rice, corn or wheat. The feeding schedule of the various diets is given in Table 2. Food was allowed ad-libitum for the first meal to identify the rat with the least food intake. Thereafter animals were pair-fed the amount of food consumed by the rat that ate the least. The food intakes of every meal were recorded for each rat and rats were weighed weekly. Protein efficiency ratios (PER) were calculated. At the end of the four week period, the last meal consisted of 2 grams of feed given after an overnight fast. Rats were bled by heart puncture at 2 h postprandial while under CO_2 anaesthesia. The liver and gastrocnemius muscles were removed rapidly and placed on dry ice for quick-freezing and stored at -30^{o} C. Due to technical problems about half of the muscles were analyzed. Amino acids were analyzed by ion-exchange chromatography (20) via a Beckman 7300 Amino Acid Analyzer (21).

Data Analysis

Statistical comparisons were made for combined and alternating diet groups in relation to the bean diet, rather than reporting comparisons between all diets.

Protein efficiency ratios (PER) were calculated by group. Significance of the data was determined statistically for plasma, liver and muscle amino acids by one-way analysis of variance with diet as the independent variable. Duncan's multiple range test was used to determine differences between means. Multiple correlation between weight gain and plasma amino acids was measured by Multiple step-wise regression analysis (22).

RESULTS

Growth

Table 3 shows the food intake and weight gain of rats and PER values of the various diets. Rats on the bean or corn diets restricted the food intake for the other groups, causing loss of weight for the first two weeks. At this point, the next limiting diet was chosen to control food intake for all the other diets. By this modification, food intake and growth rate for the last 2 weeks were comparable to previous results (17) indicating that beans or corn were more limiting than proteins used previously. For this reason the rest of this report will focus on week 3 and 4 although data is provided for the entire study period.

The total weight gain was significantly greater (<0.05) in rats fed the rice or corn combination diets, than the single protein sources. Wheat was not significantly improved when fed with beans except during week 3 and 4. There were no significant differences in weight gain between the combination and the alternation diets during week 4.

Amino Acids

Table 4 shows the multiple correlation between growth rate and plasma amino acid levels. In a multiple step-wise regression analysis, the level of plasma asparagine is the most highly correlated of amino acids related to growth, followed by arginine, histidine, tyrosine, 1-methylhistidine, and cystathionine. Serine, taurine, phosphoethanolamine, leucine and threonine were negatively correlated to growth. If only essential amino acids are considered, the results show threonine, 29.2% (inverse); and arginine, 14.4% ; tryptophan, 7.4% and histidine, 2.4%., all directly related to growth.

Table 5 shows the 2 h postprandial plasma amino acid concentrations of those amino acids correlated positively or negatively with growth. Differences for plasma phosphoethanolamine were similar to those for threonine and serine with the exception that there were no significant differences between bean and bean/wheat alternation diet groups. Plasma threonine and serine concentrations were significantly higher in rats fed beans than any combination or alternating diet group. However, there were no significant differences in plasma threonine and serine between combination or alternation diet groups except threonine in the bean/corn alternating diet group. While there were significant differences between the plasma leucine concentrations of rats fed bean/wheat combination or alternation diets, leucine levels of bean/corn combination or alternation fed rats were not significantly different. Rats fed beans with rice in combination or alternation diets did not have significantly different leucine levels than rats fed the bean diet alone

Table 6 shows the 2 hour postprandial free amino acid concentrations in liver. As in plasma, threonine and serine were significantly higher in the bean group than other combination or alternating diet groups. Of all combination and alternating diet groups, only bean/rice and bean/wheat had significantly lower concentrations of leucine than the bean diet group. Only the bean/rice fed rats had plasma histidine concentrations significantly lower than bean fed rats. Table 7 shows the values for muscle. Muscle free threonine concentration was again higher in the bean group compared to combination or alternating diet groups.

Table 8 compares the most limiting amino acids in the diets with egg as the standard protein. Table 9 compares the actual intake of the most limiting amino acids to the recommended amino acid allowances for the rat.

DISCUSSION

The available data suggest that proteins complement one another from meal to meal (23). However, conclusions from recent data question this premise and propose that proteins must be combined in the same meal for optimal growth (16). The present study was conducted to compare the growth promoting capacity of various complementary proteins fed alone, together at the same meal or alternately at separate meals and to correlate tissue free amino acid levels with growth . Growth data from this study (Table 3) confirm previous works (5, 7, 8, 17) which support the adequacy of vegetable proteins fed in a time lapse manner. Sanchez et al. (17) have shown that rice and mung beans (limiting in lysine and methionine, respectively) fed separately at alternate meals, five hours apart, had the same growth promoting capacity and protein quality as when fed together at the same meal. Others have shown that the interval for delayed supplementation of deficient diets depends upon the limiting amino acid. Lysine was effective up to 16 h after feeding (6), threonine up to 6 h (5) and methionine only 1 h (9, 10). Mills and Canolty (16) tested 0, 1, 2 and 3 day complementary effects of wheat germ/mungbean as well as sesame seed/black bean combinations and found an inverse relationship between complementary capacity and time.

These time intervals were much too long (days rather than hours) and should not be used to conclude that complementary proteins must be present at each meal for optimal growth of animals. Our previous data (17) show that complementary proteins spaced at time intervals similar to customary meals among humans promotes equal growth in rats as when fed the proteins together in the same meal. To explain this phenomenon, Nasset and Ju (24) hypothesized the existence of amino acid pools which assist growth in time delayed administration of plant proteins. Thus, the available data on delayed protein complementation suggests that the degree of delayed complementation depends upon the amino acid which is limiting in the protein and the time interval between feedings.

All amino acid concentrations are lowest at 2 h postprandial in food-restricted, young growing rats (Fig. 1). These findings are in contrast to increases in most plasma, liver and muscle free amino acids in 200 g rats force-fed adequate amounts of food (25).

Plasma threonine appears to be the essential amino acid most statistically correlated with growth, followed by some (arginine, leucine, histidine and tryptophan) but not all essential amino acids for the growing rat (Table 4). Furthermore, several non-essential amino acids (asparagine, phosphoethanolamine, tyrosine, 1-methlyhistidine, taurine, cystathionine and serine) in plasma were also correlated with growth. The metabolism of most of these latter nonessential amino acids is closely related to the metabolism of essential amino acids. Threonine and serine were the only plasma amino acids significantly different in concentration between the bean group and all other combination or alternating diet groups (Table 5). Since both amino acids undergo direct deamination via dehydratases, it would be interesting to determine levels of serine (threonine) dehydratase to estimate the rate of degradation of these amino acids in the liver. There were no significant differences in concentrations of threonine or serine in plasma of alternation and combination diet groups. It is well recognized that the amount of amino acids in dietary protein is reflected in tissue amino acid concentrations (26). This does not account for the high levels of threonine and serine in plasma and tissue of rats fed the bean diet. However, deficiency or excess in one plasma amino acid can cause changes in concentrations of other fasting (25) and postprandial (27) plasma amino acids. Thus, the plasma amino acid concentrations may not directly reflect the amino acid composition of dietary proteins.

Protein quality is estimated by comparing the essential amino acids contained in foods to a standard protein such as whole egg (28). Protein quality is also directly proportional to the essential amino acid content of foods relative to the organism's needs (23). While the limiting amino acid is not the same for each diet using these two different standards, the three most limiting amino acids by both approaches are methionine, lysine and tryptophan. Our expectation was that the most limiting amino acids, in relation to body needs, would be the least concentrated in plasma after the meal. However, growth was not correlated to the plasma concentrations of these three limiting amino acids. Threonine is more concentrated in plasma, liver and muscle among animals fed the poorest protein quality (tables 5, 6, and 7). The high sensitivity of plasma threonine to diet may not be a true estimate of the quality of the protein (29). Also, there are limitations involved when restricting calories and protein. Leverton's research (30) implied that the lower the intake of protein and calories, the greater the need for including high quality protein in each meal if nitrogen is to be well utilized. A deficiency of a single amino acid can inhibit transport systems or enzymic activity involving a number of other amino acids (31). However, in spite of multiple amino acid deficiencies and restricted calories, a time delayed complementation is evident in this study.

Data is needed to correlate tissue free amino acids with the rate of syntheses of circulating and cellular proteins in an effort to identify the amino acids that limit the growth process at different times postprandial and under restricted or unrestricted food intakes.

Table 1

Diet Composition

Components	Composition
	%
Protein[1]	10
AIN Mineral Mixture	3.5
AIN Vitamin Mixture	1
Corn Oil	2
Alphacel	2
Choline Chloride	0.1
Starch[2]	81.4

[1]Percent protein content of pinto bean, rice, corn, wheat, rice concentrate and corn concentrate were 23.51, 8.72, 10.16, 14.97, 29.25 and 44.5, respectively.

[2]Cornstarch was added to make 100%.

Table 2

Experimental Design of Delayed Time Complementation[1]

Diet Groups	Feeding Sequence			
	6-7 AM	11-12 AM	4-5 PM	9-10 PM
Single protein source				
Pinto beans (B)	B	B	B	B
Rice (R)	R	R	R	R
Corn (C)	C	C	C	C
Wheat (W)	W	W	W	W
Combined				
Pinto bean - Rice	BR	BR	BR	BR
Pinto bean - Corn	BC	BC	BC	BC
Pinto bean - Wheat	BW	BW	BW	BW
Alternating				
Pinto bean or Rice	R	B	R	B
Pinto bean or Corn	C	B	C	B
Pinto bean or Wheat	W	B	W	B

[1]All diets were pair-fed to the group with least food intake

Table 3

Mean (±SD) food intake and growth of rats and PER of diets containing single, combined or alternating vegetable proteins over a 4 week period.[1]

Diet	Total food intake	Total wt gain	Wt gain wk3	Wt gain wk4	PER wk4
	g	g	g	g	g
Single protein source					
Beans	185.4 ± 11.5^{a}	2.1 ± 5.8^{a}	3.3 ± 0.8^{a}	4.7 ± 1.1^{a}	0.97 ± 0.19^{a}
Rice	211.9 ± 12.2^{c}	26.4 ± 3.5^{b}	11.7 ± 0.2^{d}	$10.6\pm 1.8^{c,d,e,f}$	$1.70\pm0.14^{b,c}$
Corn	172.5 ± 16.5^{b}	8.1 ± 3.9^{c}	3.9 ± 2.4^{a}	$6.7\pm 2.8^{a,b}$	1.40 ± 0.59^{b}
Wheat	214.4 ± 11.3^{c}	25.0 ± 3.0^{b}	9.4 ± 1.3^{c}	$9.7\pm 2.2^{c,d}$	1.55 ± 0.28^{b}
Combined protein source					
Beans-Rice	214.6 ± 11.2^{c}	33.8 ± 6.3^{d}	12.1 ± 1.4^{d}	14.0 ± 2.9^{g}	2.22 ± 0.37^{d}
Beans-Corn	211.9 ± 13.9^{c}	22.3 ± 7.7^{b}	$8.7\pm 1.3^{b,c}$	$10.3\pm 2.4^{c,d,e}$	$1.65\pm0.24^{b,c}$
Beans-Wheat	214.9 ± 12.8^{c}	27.7 ± 4.6^{b}	11.9 ± 1.8^{d}	$12.6\pm 3.0^{e,f,g}$	$1.99\pm0.33^{c,d}$
Alternating protein source					
Beans/Rice	210.9 ± 15.0^{c}	23.5 ± 4.3^{b}	$10.3\pm 1.8^{c,d}$	$12.8\pm 3.3^{f,g}$	2.07 ± 0.48^{d}
Beans/Corn	202.7 ± 14.2^{c}	12.3 ± 4.3^{c}	6.8 ± 1.5^{b}	$8.7\pm 2.4^{b,c}$	1.52 ± 0.30^{b}
Beans/Wheat	211.0 ± 9.3^{c}	21.6 ± 5.1^{b}	$10.0\pm 1.3^{c,d}$	$12.0\pm 1.9^{d,e,f,g}$	1.98 ± 0.35^{d}

[1] Ten rats per group; mean initial weight for the 10 groups was 67.4±10.7 g, none significantly different from another.

[a] Different superscripts denote statistical differences ($p < 0.05$) between means. Similar letters denote no significant differences between groups.

Table 4

Multiple Regression Analysis of Plasma Amino Acid Concentrations versus Weight Gain

Amino Acid	Contribution
Overall[1]	%
Asparagine	15.1
Arginine	4.9
Histidine	4.7
Tyrosine	3.4
Cystathionine	2.4
1-Methylhistidine	1.8
Serine	1.5 (Inverse)
Taurine	2.3 (Inverse)
Phophoethanolamine	3.3 (Inverse)
Leucine	6.2 (Inverse)
Threonine	29.2 (Inverse)
Essential Amino Acids[2]	
Arginine	14.4
Tryptophan	7.4
Histidine	2.4
Threonine	29.2 (Inverse)

[1]Multiple correlation analysis for all amino acids.

[2]Multiple correlation analysis for essential amino acids.

Table 5

Plasma free amino acids (means ± SD) for rats fed 10 different diets[1]

Amino	Single				Combined			Alternating		
Acid	Bean(B)	Rice(R)	Corn(C)	Wheat(W)	B-R	B-C	B-W	B/R	B/C	B/W
	nmol/ml	nmol/ml	nmol/ml	nmol/ml	nmol/ml	nmol/ml	nmol/ml	nmol/ml	nmol/ml	nmol/ml
Petn[2]	78±21	55±15	64±12	80±12	57±17[a]	63±17[a]	50±16[a]	61±22[a]	55±11[a]	72±12
Thr	899±394	454±157	361±109	324±28	199±63[a]	322±67[a,b]	165±63[a]	203±103[a]	412±55[a]	181±40[a]
Ser	960±252	576±214	642±106	615±57	688±70[a]	677±91[a]	666±45[a]	606±69[a]	700±63[a]	586±42[a]
Asn	68±6	71±17	55±10	56±1	76±9	61±8	62±7	61±9	59±6	48±4[a]
Leu	121±19	140±14	113±26	92±19	116±12	143±20[c]	92±13[a]	107±15	141±20[c]	59±22[b]
Tyr	42±8	80±24	54±18	53±7	67±8	63±8	47±6	79±17	51±11	48±7
Trp	26±8	42±27	36±24	54±10	28±6	14±3	24±6	28±10	11±6[a]	32±9
His	129±14	123±35	139±39	139±22	141±19	130±21	138±17	109±12	124±15	123±12
Arg	145±15	142±38	118±16	104±15	177±27[c]	170±26[c]	118±15[a]	154±23	129±9	89±32[b]
Tau	119±68	188±64	211±72	180±25	106±49	96±24	101±52	161±121	88±31	143±49

[1] Only amino acids which were significantly associated with growth (table 4) are presented.

[2] Phosphoethanolamine

[a] Letters denote statistical differences from the bean diet ($p<0.05$). Similar letters denote no significant differences between groups.

Table 6

Liver free amino acid (means ± SD) for rats fed 10 different diets[1]

Amino Acid	Single Bean(B)	Rice(R)	Corn(C)	Wheat(W)	Combined B-R	B-C	B-W	Alternating B/R	B/C	B/W
	nmol/g	nmol/g	nmol/g	nmol/g	nmol/g	nmol/g	nmol/g	nmol/g	nmol/g	nmol/g
Petn[2]	344±115	248±462	152±46	145±138	453±214	286±143	254±145	261±87	319±152	202±110
Thr	3608±1733	1650±905	1320±603	1197±631	455±163[a]	1060±316[a,b]	434±167[a]	431±117[a]	1512±466[b]	625±499[a]
Ser	6703±1623	5509±2342	3699±1679	4519±1621	2662±769[a]	4547±694[b]	2517±732[a]	2503±349[a]	4235±1108[b]	2683±1418[a]
Asn	945±2959	617±1871	--	3±9	--	--	--	--	--	4±12
Leu	318±71	278±93	344±135	232±56	280±50	280±32	252±55	229±38[a]	298±41	213±56[a]
Tyr	141±141	151±981	136±63	90±22	107±24	94±14	82±15	116±25	87±12	95±24
Trp	1094±3357	737±2093	44±61	22±46	22±22	14±17	24±23	47±48	29±56	41±74
His	765±165	575±265	753±91	706±144	731±91	668±76	717±68	585±65[a]	676±57	658±132
Arg	13±27	5±9	17±14	10±15	7±9	9±8	6±9	10±11	8±8	15±19
Tau	741±238	1599±1230	974±168	1005±367	710±163	681±112	662±159	767±170	752±186	732±174

[1] Only amino acids which were significantly associated with growth (table 4) are presented.

[2] Phosphoethanolamine

[3] Letters denote statistical differences from the bean diet ($p<0.05$). Similar letters denote no significant differences between groups.

Table 7

Muscle free amino acid (means ± SD) for rats fed 10 different diets[1]

Amino Acid	Single				Combined			Alternating		
	Bean(B)	Rice(R)	Corn(C)	Wheat(W)	B-R	B-C	B-W	B/R	B/C	B/W
	nmol/g	nmol/g	nmol/g	nmol/g	nmol/g	nmol/g	nmol/g	nmol/g	nmol/g	nmol/g
Petn[2]	200±234	146±228	162±324	251±194	275±136	299±208	168±146	270±146	178±167	210±132
Thr	2580±496	1581±352	1156±337	1615±1168	626±176[a]	1004±236[ab]	352±111[a]	591±174[a]	1302±149[b]	571±165[a]
Ser	4458±911	3210±563	3106±448	4326±2389	3588±534	3247±479	3609±441	3347±345	4117±561	3283±617
Asn	164±30	200±36	173±55	269±189	205±58	160±20	128±114	145±86	204±18	151±38
Leu	139±36	148±22	206±122	188±141	140±59	315±409	135±11	108±23	225±171	101±43
Tyr	87±17	135±38	120±37	141±80	121±34	168±138	100±8	134±27	122±63	106±25
Trp	--	14±11	5±110	18±24	3±5	--	--	2±5	4±8	--
His	240±77	329±37	361±45	447±260	424±105	399±39	382±46	321±133	377±45	312±51
Arg	51±38	114±18	60±25	130±69	70±26	70±19	34±30	75±64	57±20	35±31
Tau	5763±2376	9479±1605	9508±1131	7239±2391	5363±718	6595±941	6548±1402	5572±47	6672±1053	6843±187

[1] Only amino acids which were significantly associated with growth (table 4) are presented.

[2] Phosphoethanolamine

[a] Letters denote statistical differences from the bean diet ($p<0.05$). Similar letters denote no significant differences between groups.

Table 8

Percent of amino acid relative to egg as 100%[1]

Amino Acid	Diets						
	Single				Combined or Alternating		
	B	B	C	W	B:R	B:C	B:W
	% Relative to Egg						
ile	67	67	59	52	67	63	60
leu	87	94	143	76	90	115	82
lys	107	54	40	42	80	74	74
met	33	66	59	47	50	46	40
cys	36	64	65	107	50	50	72
phe	92	86	86	80	89	89	86
tyr	61	77	92	72	69	76	66
thr	81	67	73	60	74	77	70
trp	58	64	44	72	61	51	65
val	64	80	67	61	72	66	62
arg	89	118	66	72	104	78	80
his	117	97	113	95	107	115	106

[1]Amino Acid Content of Foods & Biological Data on Proteins; FAO, 1970.

Table 9

Percent of amino acid relative to rat RDA as 100%[1]

Amino Acids	Single				Combined			Alternating		
	B	R	C	W	B-R	B-C	B-W	B/R	B/C	B/W
	% Relative to RDA									
ile	29	37	23	30	34	33	34	36	30	31
leu	32	45	50	39	40	53	41	43	49	39
lys	27	18	10	15	25	23	26	26	21	24
met/cys	11	27	18	31	19	19	23	20	17	22
phe/tyr	29	40	31	39	36	38	39	38	35	37
thr	27	29	23	27	30	32	32	32	29	29
trp	21	30	15	36	27	23	32	28	21	30
val	22	36	22	29	30	28	30	32	26	28
arg	97	167	67	107	135	104	118	144	95	112
his	32	34	29	35	35	39	39	37	39	37

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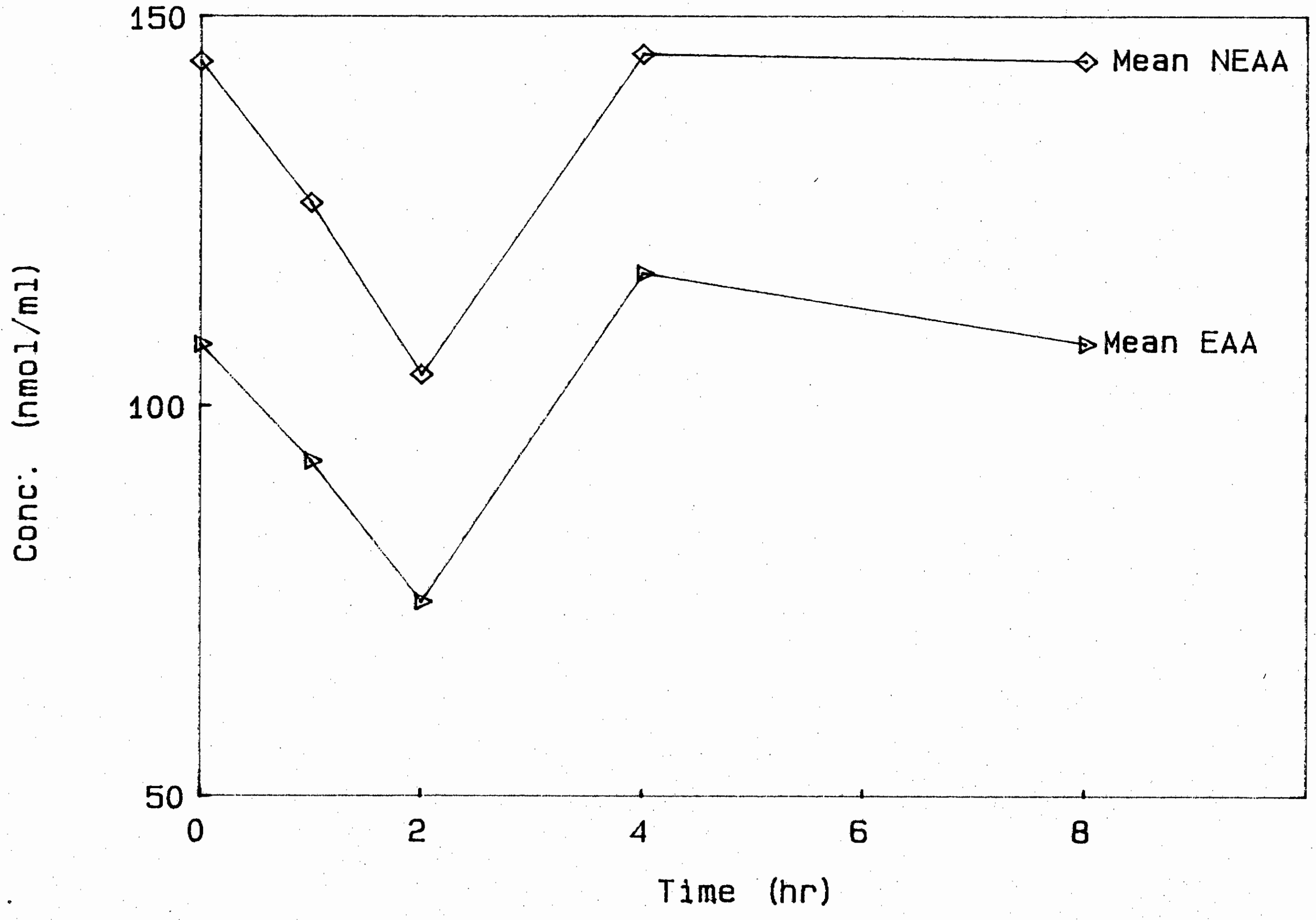
Figure 1. Mean Postprandial Essential and Nonessential Rat Plasma Amino Acid Levels. Essential amino acid levels were lysine, arginine, leucine, threonine, histidine, methionine and tryptophan. Nonessential amino acids levels were serine, taurine, tyrosine, asparagine, phosphoethanolamine and 1-methylhistidine. There were 4 rats at 1 hour, 4 rats at 2 hours, 2 rats at 4 hours and 4 rats at fasting. The fasting value was also used as the 8 hour steady state value.

Figure 1

Mean Rat Plasma AA Levels Postprandial



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