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Amino Acids in Blood Plasma of Young and Aged Adults.* (30776)

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Concentration of amino acids in blood, urine and a variety of other biological materials has been examined by a number of investigators. However, little attention has been given to aged individuals in this regard (1,2,3). An interest in the protein metabolism of elderly human subjects has permitted the collection of data to fill the void.

Early in the investigation it was necessary to decide whether or not clinically detectable senile brain damage would have a major effect on plasma amino acid levels. Since there was no basis for the decision, the subjects employed were divided into 2 groups, those who showed brain damage and a second group which appeared normal in this respect. The data collected have permitted a statistical comparison between the 2 groups of aged individuals and between each of these and the control group of healthy young adults.

Materials and methods. The subjects were patients at the Jewish Home for the Aged, Miami, Fla., selected for this study by Dr. Charles Beber, its Medical Director. Eleven normal and 16 brain damaged geriatric patients (ages 79-92) were used. Thirteen healthy adults (ages 22-40) comprised the control group and were either laboratory and hospital staff or selected control subjects.

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The medical basis for distinguishing between the normal and brain damaged geriatric groups was the face-hand test, neurological and mental status testing. A negative or questionable result in the face-hand test was considered normal, while a positive test was interpreted as that of a senile individual. A normal neurological and mental status test indicated a normal individual, and a brain damaged individual with no evidence of a stroke or gross neurological disorders, was indicated by abnormal results on these tests.

A fasting sample of venous blood was drawn before breakfast from each subject. Each 15-20 ml sample was delivered into a 30 ml centrifuge tube containing one drop Sequester-Sol, Cambridge Chemical Products, Inc., Dearborn, Mich. (dipotassium ethylenediaminetetraacetate) per 5 ml blood. After mixing, the blood was centrifuged and the plasma removed, measured, and stored at −5°C.

Five ml plasma were diluted with 20 ml triple distilled water and ultrafiltered at 375 psi nitrogen pressure for 5 hours through an Ara-Flo 65 unit (Applied Research Associates), with a one ml wash of triple distilled water at the end of the run. The volumetric recovery was 90-95%; the filtrate was then flash evaporated to dryness and brought back to the original (5 ml) plasma volume with pH 2.2 sodium citrate buffer. Duplicate fil-

TABLE I. Representative Values of Normal and Brain Damaged Geriatrics in Micromoles Per Liter Plasma.

Amino acids	Normal geriatric #	Mean	S.E.	Brain damaged geriatric #	Mean	S.E.
Histidine	11	51.8	3.0	14	52.3	5.8
Threonine	11	109.5	19.5	15	112.8	13.0
Valine	11	171.5	13.9	14	178.1	13.9
Methionine	10	20.8	1.3	15	19.5	2.3
Leucine	10	89.8	7.3	15	88.9	7.1
Phenylalanine	10	43.6	3.0	13	50.6	5.5

trations and analysis of several 10 ml plasma samples were then run to verify the advantages of this technique over figures obtained on plasmas which were not diluted for ultrafiltration.

Analyses for free amino acids in the plasma samples were conducted on a Beckman-Spinco Model 120 Amino Acid Analyzer by the method of Spackman, Stein and Moore(4). The buffer change was made at 14 hours simultaneously with the temperature change from 30°C to 50°C on the 150 cm column. Arginine was not determined; therefore, no temperature change was required for the 50 cm column.

Results. Ranking tests and t-tests were employed in a comparison of the plasma amino acid levels of normal and brain-damaged geriatric individuals.

The tests which were applied gave no indication of significant differences between the two groups.† Therefore, we have pooled the data from both groups into one category, "geriatric," and used these figures for comparison with the normal young adult data.

When the pooled data for the concentration of amino acids in the plasma of these geriatric individuals were compared with the corresponding values from a group of healthy young adults, the sole significant difference ($P<.01$) was found in the case of ornithine (Table II). It should be noted that while only ornithine showed a significant increase in the geriatric over the healthy adult, 12 of the 18 amino acids exhibited higher means in the geriatric than the normal adult. These were: methionine, isoleucine, leucine, tyrosine, phenylalanine, lysine, proline, glutamic acid, glycine, alanine, valine and ornithine. Of these amino acids, mean values of glutamic acid and valine were both elevated in the geriatric groups in excess of 20 micromoles per liter plasma over the young adult group.

Discussion. It is clearly realized that the level of amino acids in the blood plasma of individuals is the result of many factors, of which one of the most important is the amount of dietary protein(1,2). No attempt has been made to control this variable beyond the use of fasting blood samples and the fact that the meals for all geriatric subjects were prepared in a common kitchen. If, as has been suggested(5), plasma amino acid levels in general mirror protein intake, our subjects show no evidence of protein lack; on the contrary, the levels tend to be higher in the geriatric individuals than in the young adults.

Other investigators have reported finding significantly lower plasma amino acid levels in geriatric individuals than in young adults (3). As seen in Table II, we did not find this to be true with our geriatric group, and although not significantly elevated, the mean values of 12 of the 18 amino acids were higher in the geriatric than in the young adult.

Glutamic acid and valine showed the greatest difference, both being at least 20 micromoles per liter higher. These differences were not significant.

It is of interest that the levels of glutamic acid as seen in Table III were about 2½ times the values reported by investigators using methods of protein removal other than ultrafiltration. Our glutamic acid values do not include citrulline as that amino acid was eluted on the trailing edge of the glutamic acid peak and was readily identified in all runs by its lack of inversion of the suppressed 570 and 440 mμ wavelengths, as occurs with the glutamic acid peak(6).

Summary. Plasma free amino acid levels have been determined for 27 geriatric individuals. Sixteen of the patients showed senile brain damage. Thirteen young adults comprised the control group. Comparison has been made between the 3 groups, normal geri-

† George W. Snedecor, Statistical Methods, 5th ed., Iowa State College Press, 1956.

TABLE II. Plasma Amino Acid Levels (μmole/liter) in Normal Adults and Geriatric Individuals.

Amino acid	Normal adult Mean	Normal adult Range	Pooled geriatric Mean	Pooled geriatric Range	Difference between means (ger. minus norm adult)
Ornithine*	51	17-116	79.9	16-155	*28.9*
Ammonia	261.5	29-562	168.9	25-610	-91.7
Lysine	130.5	75-175	163.2	43-393	*32.7*
Histidine	57.6	28-90	52.1	0-83	- 5.5
Taurine	56.1	32-94	54.6	24-117	- 1.5
Aspartic acid	15.2	2-51	14.6	0-42	- .6
Threonine	137.1	68-168	111.4	36-266	-25.7
Serine	108.6	84-133	101.6	31-250	- 7.0
Proline	149.6	97-262	154.1	0-332	*4.5*
Glutamic acid	138.8	38-345	165.1	19-371	*26.3*
Glycine	217.0	117-447	229.5	109-368	*12.5*
Alanine	300.2	194-433	311.9	14-566	*11.7*
Valine	153.5	92-290	175.2	95-284	*21.7*
Methionine	19.1	14-25	20.0	0-38	*.9*
Isoleucine	46.3	29-85	49.2	20-69	*2.9*
Leucine	80.6	50-122	89.3	39-131	*8.7*
Tyrosine	48.7	27-76	51.5	12-114	*2.8*
Phenylalanine	45.8	26-64	47.6	21-90	*1.8*

* P <.01.

TABLE III. Influence of Deproteinization on Glutamic Acid Concentration (μmole/liter).

Ultrafiltration				Picric acid							Rapid dialysis
Young adult	Normal geriatric	Brain damaged geriatric	(7)	(8)	(3)	(9)	(10)	(11)	(12)	(13)	(14)
N.A.	N.G.	B.D.	Walker	Nyhan	Ackerman	Stein	Stein	Frame	Knauff	Christensen	McMenany
138	165	155	130	60	72	68	47	30	60	36	<20

atric, brain damaged geriatric, and normal young adults. In the 2 geriatric groups, there was no significant differences between brain damaged and non-brain-damaged; therefore, the data were pooled and compared with the young adult group. It was found that the ornithine concentration was significantly higher in the geriatric group.

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