In the transport of amino acids across the normal intestinal mucosa the magnitude of the absorptive process exceeds the efflux with an ultimate gain by the host of the nutrient amino acids from the luminal contents of the intestine in a dynamic fashion (Fig. 6).

Summary. Net active absorption of amino acids, as mixtures at blood level, has been demonstrated. Both absorption and efflux occur simultaneously across the intact intestine. The transport of amino acids is an active process and is indeed dynamic.

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Effects of Dietary Iodinated Casein on Components of the Electron Transport System of Chicken Liver.* (29967)

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Several recent studies of the effects of thyroxine on rat liver(1,2,3,4,5) and skeletal muscle(4) have presented evidence that the effects of thyroxine on electron transport are secondary to an effect on protein synthesis. Studies in our laboratory on rat liver(1) have shown a general thyroxine-induced increase of all components of the succinoxidase system. Since this should be a basic metabolic phenomenon, it was decided to investigate a second vertebrate class.

Methods and materials. Hubbard White Mountain chickens were placed on experiment at 4 weeks of age, treated for 4 weeks and sacrificed by severing the neck arteries. For treatment, the birds were divided into 8 groups of 5 males and 5 females, and while Groups 1-4 were kept at 70°F, Groups 5-8 were kept at 85°F. Groups 1 and 5 received the basal diet and in all other groups, iodinated casein was substituted for glucose monohydrate in the basal diet at levels of 0.05% (Groups 2 and 6), 0.10% (Groups 3 and 7) and 0.20% (Groups 4 and 8). Livers were rapidly removed following sacrifice and chilled in 20 ml of the homogenizing medium (0.25 M sucrose).

Previously described assays were used for determination of cytochrome oxidase activity

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TABLE I. Effects of Iodinated Casein on Succinic Oxidase Activity of Liver Homogenates from Chickens Kept at 70° and 85°F.

Ambient temp	Iodinated casein (% of diet)	Succinic oxidase activity* Males — cyt c	Males + cyt c	Males + cyt c/ — cyt c	Females — cyt c	Females + cyt c	Females + cyt c/ — cyt c
70°F	.00	1.7 (.7)	4.8 (1.3)	2.8	1.4 (.5)	4.6 (.9)	3.3
"	.05	3.8 (.7)	6.5 (.4)	1.7	2.4 (.6)	6.0 (.7)	2.5
"	.10	3.1 (.7)	6.4 (.6)	2.0	2.6 (.7)	7.0 (.8)	2.7
"	.20	4.4 (1.1)	7.9 (1.1)	1.7	3.8 (1.0)	9.0 (.9)	2.4
85°F	.00	1.4 (.6)	4.6 (.5)	3.3	1.5 (.3)	5.4 (.6)	3.6
"	.05	2.3 (.5)	6.1 (2.0)	2.7	1.9 (.8)	4.7 (1.1)	2.5
"	.10	3.4 (.4)	6.6 (.6)	1.9	3.1 (1.0)	6.9 (1.0)	2.2
"	.20	2.1 (.1)	6.9 (1.5)	3.3	1.7 (.6)	5.6 (1.4)	3.3

( ) Standard error of mean.

* μatoms of oxygen/min/g liver (wet wt).

TABLE II. Effects of Iodinated Casein on NADH Oxidase Activities of Liver Homogenates from Chickens Kept at 70° and 85°F.

Ambient temp	Iodinated casein (% of diet)	NADH oxidase activity* Males — cyt c	Males + cyt c	Males + cyt c/ — cyt c	Females — cyt c	Females + cyt c	Females + cyt c/ — cyt c
70°F	.00	2.3 (1.4)	6.7 (1.2)	2.9	2.2 (.6)	8.6 (1.7)	3.9
"	.05	8.0 (2.0)	12.4 (3.4)	1.6	4.2 (1.7)	10.1 (2.9)	2.4
"	.10	6.0 (2.4)	11.2 (1.4)	1.9	4.3 (1.4)	13.4 (2.4)	3.1
"	.20	7.9 (1.2)	13.7 (3.9)	1.7	8.2 (2.2)	14.7 (3.7)	1.8
85°F	.00	2.4 (.9)	7.3 (1.7)	3.0	1.6 (.2)	6.5 (2.3)	4.1
"	.05	5.5 (1.9)	12.6 (2.5)	2.3	3.4 (1.8)	8.5 (3.9)	2.5
"	.10	7.6 (2.3)	11.5 (2.6)	1.5	7.5 (3.6)	11.5 (3.4)	1.5
"	.20	4.8 (1.9)	14.6 (1.6)	3.0	3.3 (1.1)	9.8 (6.3)	3.0

( ) Standard error of mean.

* μatoms of oxygen/min/g liver (wet wt).

(6), and reduced nicotinamide adenine dinucleotide (NADH)- and succinic-cytochrome c reductase activities(1). Succinic oxidase and NADH oxidase activities were determined polarographically. The former system contained 100 μmoles of phosphate, pH = 7.2; 40 μmoles of succinate, pH = 7.0; and 5-10 mg protein in a final volume of 2.0 ml. The latter system contained 80 μmoles of phosphate, pH = 7.4; 0.5 μmole of NADH; and 5-10 mg of protein in a final volume of 2.0 ml. The degree of "openness" to cytochrome c was determined by adding 1 μmole of cytochrome c to each system. All assays were performed on whole homogenates at 38°C.

Coenzyme Q (CoQ) was determined by the method of Crane and Dilley(7) and protein by the biuret reaction(8).

Reduced β-NAD and cytochrome c were supplied by Sigma Chemical Co., iodinated casein containing 7% iodine and 1% thyroxine activity was supplied by Nutritional Biochemicals Co., and inorganic reagents were supplied by Fisher Chemical Co.

Results. As shown in the Tables there are 2 distinct effects, one of the environmental temperature and another of the dietary iodinated casein. There also seems to be an interrelationship between these 2 effects since the effects of iodinated casein express themselves differently at the 2 temperatures.

If the controls are compared at the 2 different temperatures, it can be seen that there is no significant difference in either the succinic oxidase (Table I) or NADH oxidase (Table II) activities either in the presence or absence of exogenous cytochrome c, or in the cytochrome c oxidase activity (Table III). However, both the succinic- and NADH-

TABLE III. The Effects of Iodinated Casein on NADH- and Succinic Cytochrome c Reductase Activities and Cytochrome c Oxidase Activity of Liver Homogenates from Chickens Kept at 70° and 85°F.

Ambient temp	Iodinated casein (% of diet)	NADH-cyt c reductase* Male	NADH-cyt c reductase* Female	Succinic-cyt c reductase* Male	Succinic-cyt c reductase* Female	cyt c oxidase† Male	cyt c oxidase† Female
70°F	.00	15.7 (7.4)	17.2 (5.3)	4.7 (1.5)	5.2 (.9)	25.9 (5.7)	37.0 (8.8)
"	.05	22.0 (2.8)	24.8 (7.5)	6.0 (1.3)	6.2 (.9)	25.4 (6.9)	38.1 (9.6)
"	.10	22.3 (6.9)	26.6 (7.4)	6.3 (.6)	7.9 (1.3)	47.1 (9.4)	50.5 (7.8)
"	.20	31.0 (9.3)	32.4 (9.8)	11.5 (2.3)	11.4 (1.5)	34.0 (5.6)	32.6 (9.0)
85°F	.00	27.1 (8.2)	28.2 (8.5)	7.6 (1.2)	7.1 (.8)	23.1 (3.2)	27.4 (4.4)
"	.05	21.6 (6.6)	19.6 (6.0)	6.8 (1.5)	5.8 (1.7)	32.4 (10.6)	32.4 (10.3)
"	.10	21.5 (6.6)	31.8 (9.7)	7.0 (1.1)	8.4 (1.9)	26.1 (4.8)	32.8 (2.9)
"	.20	19.6 (5.9)	17.8 (5.4)	7.0 (1.1)	5.6 (1.0)	55.4 (7.7)	32.4 (7.5)

( ) Standard error of mean.

* μmoles of cytochrome c reduced/min/g liver (wet wt).

† μatoms of oxygen/min/g liver (wet wt).

cytochrome c reductase activities appear to be elevated by the 15°F increase in ambient temperature (Table III).

In comparing the effects of iodinated casein at the 2 temperatures, it can be seen that at 70°F, with the exception of the cytochrome c oxidase activity, all activities are progressively increased by each of the 3 dose levels (Tables I, II and III). In the birds maintained at 85°F, the succinic and NADH oxidase activities in the absence of exogenous cytochrome c are increased by the 0.05 and 0.1% levels while the 0.2% level depresses the activities to levels which approach the controls or the 0.05% level (Tables I and II). In the presence of exogenous cytochrome c, there is a difference in the pattern of change and the depression by the 0.2% level is either absent or less pronounced. This difference in effect on the two systems is best expressed by the ratio of the activity in the presence to the activity in the absence of cytochrome c. Comparing the control values at the 2 temperatures, it can be seen that there is no significant difference. At 70°F all 3 levels of iodinated casein depress the ratio to approximately the same degree while at 85°F only the 0.05 and 0.1% levels depress the ratio and at the 0.2% level the ratio returns to the control value (Tables I and II).

From Table III it can be seen that the succinic-cytochrome c reductase activity in the controls appears to be higher at 85°F than at 70°F, and although the differences for the individual sexes are not statistically significant, the consistency of the results in both sexes strongly suggests that the NADH-cytochrome c reductase activity is also increased. Similarly, feeding of iodinated casein at 70°F causes a progressive increase in both succinic- and NADH-reductase activities up to 2-fold that of the controls, and although many of the differences for NADH-cytochrome c reductase are not statistically significant, the results are consistent in both sexes. At 85°F there is certainly no increase and possibly a slight decrease of activity with increasing levels of iodinated casein.

Cytochrome c oxidase activity at 70°F does not show the consistent pattern of change found with the reductases. If the increase at the 0.1% level can be taken as significant the effect is limited to a narrow range since the 0.2% level is back to the control value (Table III). At 85°F no consistent pattern of change is observed.

Table IV shows that at 70°F the CoQ content of the liver is increased by feeding iodinated casein at all 3 levels. At 85°F there is a trend analogous to that seen in the NADH and succinic-oxidase activities at 85°F, but the changes are small and are not statistically significant.

Discussion. Under thyroxine treatment at a normal temperature(1) or under cold stress (9) the rat responds by increasing its electron transport potential. The results obtained at 70°F are in good agreement with those obtained previously in studies on liver

TABLE IV. Effects of Iodinated Casein on Coenzyme Q Content of Liver Homogenates from Chickens Kept at 70° and 85°F.

Ambient temp	Iodinated casein (% of diet)	Coenzyme Q* Male	Female
70°	.00	77 (35)	76 (30)
"	.05	102 (10)	84 (13)
"	.10	120 (20)	103 (18)
"	.20	114 (18)	137 (28)
85°	.00	86 (12)	89 (13)
"	.05	97 (20)	102 (11)
"	.10	97 (37)	99 (24)
"	.20	88 (8)	60 (15)

( ) Standard error of the mean.
* μg of Coenzyme Q/g liver (wet wt).

homogenates from thyroxine treated rats(1). The differences in response obtained from chickens kept at 85°F are of interest, and at present cannot be explained. Further work on the status of the electron transport and protein synthetic mechanisms of both the rat and chicken under various thyroid states and at several environmental temperatures should be examined in the hope of gaining a better understanding of the role of thyroxine in thermoregulation and of changes in responsiveness of the tissue to a given thyroxine titer.

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Effect of Dodecyl Succinic Acid on *Pseudomonas aeruginosa* and Its Reversal by Formaldehyde and Iron. (29968)

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Anionic detergents react with positively charged groups on proteins(1,2). The positive charge is primarily contributed by amino and amide groups and anionic compounds are more effective in acid solutions(3). Their effect on bacteria is probably the result of a similar interaction with proteins of the cell membrane since work with protoplasts(4,5) shows membrane disruption in the presence of the compounds. If reaction with amino groups is important for the action of these compounds on bacteria, then pretreatment with small amounts of formaldehyde should counteract some of their effects. Cations such as calcium and iron which may be present on cell membranes could also react with anionic detergents and thus addition of small amounts of these ions to the cells may also counteract the effects. To test these possibilities a strain of *Pseudomonas aeruginosa* was used. This organism as well as many other Gram negative ones rapidly decreases in size when placed in salt solutions(6). Upon incubation a slow increase in size occurs as a result of the influx of ions with their water of hydration(7). The change in size can be measured by the change in optical density and the validity of the method has been checked with electron micrographs(7). The effect of detergents on the rate of cell size increase was studied in the presence and absence of formaldehyde and ferrous salts. Some experiments on the effect of mercury and EDTA are also included.

Materials and methods. A strain of *Pseudomonas aeruginosa* maintained in this laboratory for 15 years was grown at 34° for 24 hours on Difco nutrient broth. The cells were spun down and then washed twice with water by centrifugation. They were then sus-