

Absorption Tests on Protein-Deficient Rats.

Absorption of S^{35} -Methionine and I^{131} -Triolein

In the process of digestion and absorption, the small intestine takes an active part. In recent years, therefore, the digestive and absorptive activity of the small intestine has been thoroughly studied by gastro-enterologists.

In recent years, by using isotope-marked nutrients, a more exact tracing of the absorption mechanism was made possible (HORZELA¹, STANLEY², RUFFIN³, BERKOWITZ⁴, CSERNAY⁵).

The available literature of nutritional science presents only very few data about the influence of nutritional deficiency or of overfeeding on absorption. According to our investigations (BEDÖ and KEMÉNY⁶), it became evident that a protein deficient diet, fed for a long period, affected only to a small extent the absorption of the different sugars in rats. The aim of the present investigations was to study the effect of a protein deficient diet on the absorption of S^{35} -methionine and I^{131} -triolein.

Experimental. Investigations were performed on male white rats, weighing 200–300 g, of our Institute's inbred strain, using the method of KERTAI et al.⁷. After opening the abdominal wall, the pilorus and the large intestine, at the level of the ileocecal valve, were ligated and glass cannules inserted. The intestine was washed with physiological saline at 37°C, and after removing the intestinal content, the experimental material was injected into the intestine.

The degree of absorption was calculated from the difference between the amount of isotope injected and that washed out at the end of the experimental period.

The composition of the diet was as follows:

Protein deficient diet		Control diet	
Starch	82%	Starch	64%
Fat	10%	Fat	10%
Salt mixture after Sós	4%	Casein	18%
Dried yeast	3%	Salt mixture after Sós	4%
Cod-liver oil	1%	Dried yeast	3%
		Cod-liver oil	1%

To satisfy the nibbling urge of the animals and to ensure the necessary ballast material, 2% sawdust was mixed to the diet. The test was carried out in nembutal narcosis (4 mg/100 g body weight i.p.). On the 8th, 16th, 24th and 32nd day of the administration of the protein deficient diet, 5–8 animals were used in a group simultaneously with control rats.

Amino acid absorption was tested with carrier-free S^{35} -methionine. In 2 ml physiological sodium chloride, 20 μ g 3 μ Ci S^{35} -methionine was injected into the small intestine of each animal. Absorption time was 20 min.

The absorption of fat was tested by using I^{131} -triolein. Per animal 4 μ Ci, i.e. 0.026 ml, I^{131} -triolein was injected in 2 ml physiological sodium chloride. Chemical composition of the isotope: one part I^{131} -triolein + two parts Tween-80 emulgator. The triolein absorption test lasted 30 min.

The samples washed out from the intestine of each animal were diluted to 25 ml. From the diluted material 0.5 ml aliquots were taken. Radioactivity was measured in the usual way by using a Geiger tube with a 1.7 mg/cm² thick end window of mica.

Results and discussion. The results are shown in Figures 1 and 2. In Figure 1 the percent absorption of S^{35} -methionine is shown in progression of protein deficiency

as compared with the controls. From the 24th day of protein deficiency, a mild but not significant decrease was observed in the amino acid absorption.

Figure 2 shows the absorption of I^{131} -triolein. From the 24th day of the protein deficiency, an essential decrease could be observed in fat absorption ($p < 2\%$).

Amino acids are absorbed by the active phosphorylation of the epithelium, similar to the absorption of

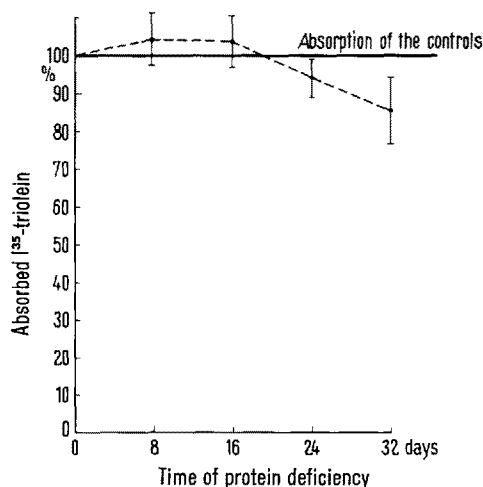


Fig. 1. S^{35} -methionine absorption in protein-deficient rats.

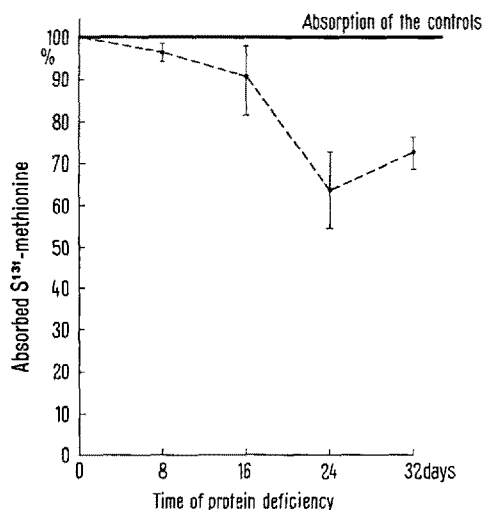


Fig. 2. I^{131} -triolein absorption in protein-deficient rats.

¹ T. HORZELA, Z. SZYBINSKI, and S. KONTUSEK, *Acta physiol. polon.* 13, 149 (1962).

² M. M. STANLEY and S. J. THANNHAUSER, *J. lab. clin. Med.* 34, 1634 (1949).

³ J. M. RUFFIN, W. W. SHINGLETON, G. I. BAYLIN, and J. C. HYMAN, *New Engl. J. Med.* 255, 594 (1955).

⁴ D. BERKOWITZ, M. N. CROLL, and B. SHAPIRO, *Gastroenterology* 42, 572 (1962).

⁵ L. CSERNAY, A. BIRÓ, and V. VARRO, *Orv. Hétl.* 105 évf. 14, 637 (1964).

⁶ M. BEDÖ and T. KEMÉNY, in press.

⁷ P. KERTAI, J. GYÖKÖSSY, and G. LUDÁNY, *Kísér. Orvostud.* 4, 373 (1955).

methionine (WISEMAN⁸). The absorption mechanism may be influenced by several factors, as for instance by the preceding food intake (DONHOFFER⁹). In the case of protein deficiency, production of the protein-splitting enzymes in the pancreas may suffer an injury. For the absorption of S³⁵-methionine, no protein-splitting enzymes are necessary and so it is obvious that its absorption through the intestinal wall is not considerably affected by the protein-deficient condition of the organism.

Remarkable was the difference in the absorption of I¹³¹-triolein after protein deficient diet had been given for a longer period. I¹³¹-triolein is the ester of three oleic acid groups and one glycerine molecule. Each oleic acid group contains a double bond. According to its chemical nature the administered isotope is a fat, thus when investigating its absorption, one must consider the extensive digestive activity preceding fat resorption (VERZÁR and KUTHI¹⁰, FRAZER¹¹). For the changes in the absorption of I¹³¹-triolein, the secretive activity of the intestinal epithelial cells, as well as of the bile and pancreas, may be responsible. To elucidate to what an extent these three factors take part in this action needs further investigation. Besides, it is possible that the energy stores of the protein deficient organism are too much decreased for the

mechanism of fat resorption. This hypothesis is supported by our investigations, performed on similar animals, when, after double sugar loading in progression of protein deficiency, the protein and ATP content as well as the activity of the ATPase and the alkaline phosphatase gradually decreased.

Zusammenfassung. Die Wirkung der Eiweissmangeldiät auf die Resorption von S³⁵-Methionin und I¹³¹-Triolein wurde untersucht. Die Resorption von Methionin wurde nicht wesentlich beeinflusst, während die Fettresorption nach 24tägigem Eiweissmangel signifikant vermindert ist.

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September 21, 1964.*

⁸ G. WISEMAN, *J. Physiol.* 120, 63 (1953).

⁹ Sz. DONHOFFER, *Pflügers Arch. ges. Physiol.* 246, 92 (1942).

¹⁰ F. VERZÁR and S. KUTHY, *Biochem. Z.* 210, 265 (1929); 225, 267 (1930); 230, 451 (1931).

¹¹ A. C. FRAZER, *Physiol. Rev.* 20, 561 (1940); 26, 103 (1946).

Transcallosal, Extracallosal, and Geniculo-Cortical Responses during Physiological Sleep and Wakefulness

The excitability of neurons in the lateral geniculate body and visual cortex undergoes clear-cut variations during paradoxical sleep¹⁻⁴. The aim of this investigation was the analysis of transcallosal (TCR), extracallosal (ECR) and visual cortex (VCR) responses and their interaction during the natural phases of sleep and wakefulness.

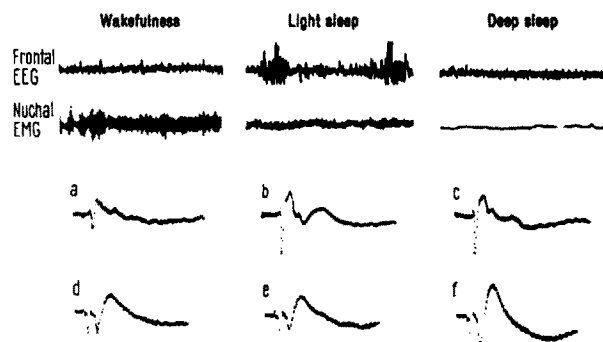
Experiments were made on cats, unrestrained, with chronic bipolar stimulating electrodes implanted in the lateral geniculate body of one side and in the white matter underneath the lateral gyrus of the opposite hemisphere. The responses were monopolarly recorded from the same cortical point in the lateral gyrus of the side where the lateral geniculate body was stimulated. A variable number of responses (50-300) was integrated in a CAT 400 Mnemotron and then photographed on an oscilloscope. The EEG (from frontal areas) and the EMG (from nuchal muscles), recorded with an inkwriter electroencephalograph, gave information on the degree of wakefulness and stage of sleep of the animal during the experiment.

Stimulation of white matter underneath the lateral gyrus, with single shocks (1/sec, 0.05-0.1 msec, 2-4 V) evoked in the homologous point of the opposite hemisphere a TCR with 2 msec latency followed by the ECR with the positive peak at 50 msec and the negative one at 90 msec from the stimulus (Figure b).

During *attentive wakefulness* the TCR fluctuated in amplitude and showed, when integrated, the lowest values observed. Usually its negative component was strongly reduced. The ECR was rarely present during wakefulness and when integrated was always very small in amplitude (Figure a).

In *light sleep* (with synchronized EEG), the TCR had the maximal amplitude. The ECR showed all its compo-

nents. An integration of both responses revealed the highest values observed (Figure b).



Transcallosal, extracallosal and geniculo-cortical responses during wakefulness, light sleep and deep sleep. Unrestrained cat with chronic implanted electrodes. Each photogram represents an integration of one hundred responses. a, b, c: Responses of the lateral gyrus to stimulation of the homologous point of the white matter of the opposite hemisphere during attentive wakefulness (a), light sleep (b) and deep sleep (c). d, e, f: Responses of the same cortical point of the lateral gyrus to stimulation of the lateral geniculate body during the same episodes of wakefulness (d), light (e) and deep sleep (f). The sweep of photograms a, b and c lasts 125 msec and of d, e and f 31.25 msec.

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