the data of COMROE and his co-workers[1] has been constructed from Table 3 taken from that paper.

From Figure 2 we observe the following: There is a relatively close resemblance between the data of the four separate experimental groups. The curve for the regeneration is exponential and similar to those obtained through isotope studies. It may be concluded, therefore, that this enzyme exists in the body in the same dynamic state as, for example, serum albumin. We may from these curves also obtain the half life time of the enzyme, approximately 6–7 days. This may be compared with the reported[2] half life time value for serum albumin of 20 days obtained for normal man. Actually, the half life time obtained with the D.F.P. procedures is not strictly comparable to that gotten from the isotope method. We are at present attempting a mathematical formulation which will make it possible to translate one into the other. This paper has been presented, since it offers a new method for determining the dynamic state of an enzyme. The data for serum cholinesterase has been presented above, but the procedure may be used generally for such determinations with other enzymes. With D.F.P., important information may be obtained not only for serum cholinesterase, but for true cholinesterase as it occurs in many tissues.

M. G. LEVINE

Kabat-Kaiser Institute, Vallejo, California, November 29, 1952.

Zusammenfassung

Eine Methode wird dargestellt, mittels welcher das dynamische Verhalten und die Halblebenszeit eines Enzyms berechnet werden kann. Es wird ein Inhibitor hinzugefügt und nachher die Regeneration, *in vivo*, bestimmt. Das Enzym ist «Pseudocholinesterase» und der Inhibitor ist Di-isopropyl-Floro-phosphate.

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Host Resistance to Tumor Implantation, its Alteration and Relationship to the Individual Metabolic Pattern[1]

We have been working in this laboratory for several years with a transplantable fibrosarcoma, the implantation incidence of which was relatively low in a closely-bred strain of SHERMAN rats. An attempt to explain this low incidence led to the consideration of a number of possible causes.

If, for any reason, transplanted fragments do not possess a comparable potential of active cells, due to variation within the tumor stock itself, or if certain variables in the technique of grafting are not adequately controlled, one may expect a lowered incidence. On the other hand, it is plausible to assume, the tumor stock and variations in technique being controlled, that some individuals are more susceptible than others to a tumor implant. Under such conditions, one should be able, by challenging with grafts, to segregate tumor-resistant from tumor-susceptible animals. With such groups available, a new approach to the problem of the nature of resistance to cancer becomes possible through a comparative study of the metabolism of these respective groups of animals.

Our first efforts were, therefore, concerned with establishing a reliable tumor transplantation technique and eliminating possible variation within the tumor stock. An early study[1] showed that the age of the host bearing a stock tumor had a distinct effect on viability of the tumor. It was demonstrated that when rats were implanted with tumors at the age of 10–15 days, the tumors which developed within 2–3 weeks were better for transplantation purposes than those grown in older animals. Such tumor stocks from young animals were subsequently used. It was also shown that all portions of such tumors are equally viable[2]. By propagating the tumor in young animals at regular intervals, and utilizing stocks of the proper age, variations within the tumor stock have been definitely controlled.

The methods for handling the tumor during the process of grafting have likewise been carefully standardized. Grafting is carried out in a constant-temperature room at 24°C, which was shown to be more favorable to viability than 37°C. Graft size, within a certain range, is a critical factor, but it can be controlled so that implantation incidence following replicate grafting with fragments from the same or similar stock tumors in the same animals does not differ significantly[3]. As a matter of fact, segregation of resistant from susceptible animals has been achieved by challenging with quadruple grafts.

Other studies, in which standard techniques and tumor stocks were used, showed that the resistance to implantation of a significant number of individuals within an experimental group may be affected by specific factors and that it can be altered by nutritional means. Age of individuals receiving grafts is a determining factor[4]. Dietary supplementation with pyridoxine favors implantation[5], as does supplementary administration of thymic extracts[6]. Phenylalanine supplements, however, lowers incidence of successful implants[7], while riboflavin has no effect on incidence, but does affect growth of the tumor after implantation[8].

Having shown that resistant animals can be segregated from susceptible ones and that dietary supplementation can alter resistance, it is of interest to know what may be the physiological basis for the phenomenon of resistance.

It would seem that implantation and growth of tumors, being subject to nutritional control, constitutes another case of what is characterized as "genetotrophic" diseases by R. J. WILLIAMS *et al.*[9]. Individual and strain variation in regard to tumor susceptibility may simply be an expression of metabolic pattern variation. Quantitative analytical determination of the constituents of representative body fluids, such as urine, have revealed that

[1] Presented at the Second International Congress for Biochemistry (Section on Cancer), Paris, July 1952. Aided by a grant from the Damon Runyon Memorial Fund.

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[2] J. B. LOEFER, R. B. MEFFERD, Jr., and R. M. NETTLETON, Jr., Texas Rep. Biol. Med. *10*, 598 (1952).

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[4] J. B. LOEFER and N. G. GILLES, Cancer *4*, 1259 (1951). – J. B. LOEFER, Cancer *5*, 163 (1952).

[5] J. B. LOEFER, Can. Res. *11*, 481 (1951).

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[7] J. B. LOEFER and R. B. MEFFERD, Jr., Texas Rep. Biol. Med. *10*, 614 (1952).

[8] R. B. MEFFERD Jr. and J. B. LOEFER, Texas Rep. Biol. Med. *10*, 619 (1952).

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there are marked and constant differences in the quantities of materials excreted–differences which may be as great as 1500%. These workers have correlated certain individual metabolic patterns, particularly those involving vitamin deficiencies, with certain diseases and have shown that these may be successfully treated by dietary supplementation, i.e., they are genetotrophic. They report the extremely encouraging conclusion that even though a physiological condition rests upon hereditary roots, a nutritional attack may be successful in modifying or alleviating it.

Utilizing this concept, we have determined the quantities of more than twenty compounds present in the urine of tumor-susceptible and -resistant rats. Daily and long range excretion variation has been compensated for by pooling five 24-hour urine specimens per individual rat, and repeating the procedure a second time two months later. Quantitative determinations in replicate were made upon each of these two pooled individual samples. Values obtained were corrected for the weight of individual rats so as to make comparisons possible. The number of grams of feed required to be consumed per rat to yeld an excretion of one milligram of a given substance was determined. The Table presents typical data for a single substance (leucine), and serves to illustrate the method employed.

Utilizing the median value for each compound excreted, animals may be ranged according to whether they excrete more or less than this "normal" value. Reported in this manner, it is possible to differentiate most of the susceptible animals by utilizing singly any one of several compounds. These separations are statistically significant, but in each case one or two animals demonstrate aberrant behavior, such that it is impossible to use the results of a single determination. The difference becomes much more striking, however, when several compounds are considered simultaneously.

J. B. LOEFER and R. B. MEFFERD Jr.[1]

From the Southwest Foundation for Research and Education, and Trinity University, San Antonio, Texas, U.S.A., November 18, 1952.

Evaluation of leucine excretion as a means of differentiating between tumor-resistant and tumor-susceptible rats.

Weight in grams*	Milligrams leucine excreted per rat per day	Grams feed consumed per day	Corrected leucine excreted per day**
Susceptible rats			
322	0·26	25·7	88·6
401	0·29	28·3	108·8
351	0·24	26·3	105·2
497	0·33	35·4	147·5
368	0·23	25·1	109·1
450	0·26	25·7	121·4
329	0·24	26·6	98·5
			$\bar{x}_1 = 111{\cdot}3$
Resistant rats			
305	0·27	23·6	73·8
369	0·26	29·2	112·3
405	0·28	27·6	110·4
378	0·29	25·2	90·0
335	0·29	24·1	75·3
270	0·26	17·6	50·3
340	0·25	27·4	101·5
406	0·32	28·1	96·9
280	0·27	23·6	67·4
357	0·33	25·8	75·9
364	0·29	25·8	89·0
			$\bar{x}_2 = 85{\cdot}7$

* Median weight: 364 (weighted to equalize classes).
** Grams feed consumed per day per individual

(milligrams leucine excreted per day per rat) $\left(\frac{\text{median weight}}{\text{individual weight}}\right)$

$x_1 - x_2 = 25{\cdot}6$; $n_1 = 7$, $n_2 = 11$; d. f. = 16; $Sx^2 = 6387{\cdot}7$; $t = 2{\cdot}66$, $P < 0{\cdot}02$; significant at $P < 0{\cdot}03$ level by median test[1].

[1] A. M. MOOD, *Introduction to Theory of Statistics* (McGraw-Hill, New York, 1950), pp. 394.

Zusammenfassung

Es wurde ein Fibrosarkom der Ratte transplantiert, welches sich nicht in allen Tieren der Sherman-Rasse einpflanzen lässt. Über zwanzig Komponenten des Urins von sowohl resistenten wie auch von empfänglichen Tieren wurden analysiert. Die statistische Behandlung der Resultate zeigte, dass bei Tieren aus gleicher Zucht deutliche quantitative Unterschiede in den Urinanalysen existieren. Die Leuzinausscheidung in einem Versuch mit 18 männlichen Ratten übertraf bei den empfänglichen Tieren um 30% diejenige der resistenten von demselben Alter ($P < 0{,}02$). Auch die Ausscheidung anderer Komponenten ist bei den Empfänglichen verschieden von der resistenter Tiere.

[1] Post-Doctoral Fellow in Cancer Research of the Damon Runyon Memorial Fund.

Mammalian Specificity of Fibrinogen

There is no uniformity in the results of investigations concerning the antigen properties of fibrinogens. In some papers[1] considerable differences were reported between fibrinogens of different animals, in others[2] a limited relationship was demonstrated between the fibrinogens of some mammals, while in a third group of papers[3] the relationship is stated to be rather remote. It seems important to clarify this problem both from a theoretical and from a practical point of view. By the fractionation of human plasma different derivatives of fibrinogen are prepared which are successfully used in therapy in different lines of medical practice. The extensive use of these products, however, could not be realized, owing to the fact that they are produced from *human* plasma, a raw material available only in limited amounts, and–in spite of voluntary donors–very expensive. In addition the fibrinogen level in human plasma is very low, and a continous production of the therapeutic derivatives cannot easily be organized. Hence, different substitutes are used such, as gelatina-foam, oxidized cellulose, polyethylen, etc., which are unable to compensate natural proteins in the organism. Since all fibrin products are derivates of fibrinogen, first of all the antigen properties of fibrinogen were studied.

Materials and methods

Preparation of fibrinogen from mammalian and fowl plasma. Fibrinogen was prepared by LAKI's method[4].

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