

reaction was usually impressive when both the aortic nerves had previously been severed, as in B, but was also evident (A) when the aortic nerves had been left intact and allowed to compensate for interruption of the homologous afferents in the vagus. Also the effects of vagal blockade upon basal blood pressure, which were barely apparent when other baroreceptive pathways were working, became constantly clear, and often dramatic (up to 40 mm Hg increase of the pre-existing hypertension), when in addition to aortic nerves severing both carotid sinuses were previously denervated.

As vagal section constantly induces respiratory changes, it was thought essential to test whether the pressor response to vagal interruption might not be secondary either to a rise in alveolar carbon dioxide concentration or to hemodynamic alterations resulting from the changed respiratory mechanics. The first possibility was ruled out in a large series of animals in which alveolar CO<sub>2</sub> concentration was continuously measured by means of an infrared analyzer. In most preparations, vagal section or blockade, although inducing definite increases of the carotid occlusion response or of basal arterial pressure, did not modify alveolar CO<sub>2</sub> concentration. Also the second possibility could be excluded, by showing persistence of the circulatory effects of vagal interruption in decerebrate animals immobilized with gallamine, triethiodide and artificially ventilated.

Finally, a third series of experiments was conceived to appraise the role played by aortic accessory afferents (i.e. fibers originating from the aortic region, although running in the vagus rather than in the aortic nerve) in the circulatory effects of vagal sectioning. As baro- and chemoceptive fibers from the aortic region are likely to run intermingled along the peripheral afferent paths, completeness

of aortic receptive area deafferentation after interruption of the aortic nerves was tested by intraventricular injection of small amounts (50–100 µg) of potassium cyanide, a classical stimulant of aortic and carotid bodies chemoreceptors. The respiratory and circulatory responses to the drug, always clear-cut whenever at least one aortic nerve was intact (both carotid sinuses being preliminarily ablated in this group of experiments) completely disappeared upon section of the aortic nerve in spite of the fact that the left vagus remained untouched. This demonstrates that no important contribution of chemoceptive (and presumably baroreceptive) afferents from the aortic region is carried through the left vagus nerve.

To sum up, our experiments have succeeded in demonstrating the quantitative importance of the tonic reflex influence exerted by afferent vagal fibers on arterial pressure, its independence of concomitant respiratory changes, and that its origin is different from that of the classical afferent fibers running in the aortic nerve. A more precise localization of the origin of the vagal afferents will be the subject of a subsequent report.

*Riassunto.* La sezione del vago cervicale, dopo precedente sezione dei nervi aortici, produce un cospicuo aumento della risposta pressoria all'occlusione carotidea, o, nell'animale con denervazione seno-carotidea, della pressione arteriosa basale. Questo fenomeno non dipende dalle concomitanti variazioni respiratorie, né dalla sezione di eventuali fibre di origine aortica decorrenti nel vago.

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### Tumor-Producing Capacity of Transplanted Lung and Spleen Taken from Yoshida Sarcoma Bearing Rats

Clinical and experimental evidence accumulated in recent years indicates that tumor cells, in general, circulate earlier and with greater frequency than it was formerly supposed<sup>1-4</sup>. This fact has a special bearing on the problem of metastases, and many questions must be answered before the precise relationship between circulating tumor cells and localized secondary growth can be established. We report here some preliminary observations on the problem.

Yoshida sarcoma cells circulate very precociously. Our unpublished observations have shown that heart-blood taken from tumor-bearing animals, as early as four days after tumor implant, is able to reproduce the growth when injected subcutaneously to recipient animals. Nodular metastases are not seen in Yoshida tumor-bearing rats,

even in terminal stages, although a considerable number of tumor cells can always be seen within vessels of practically every organ. The reason why these cells do not give rise to nodular metastases is not entirely understood as yet.

One first approach to the problem is to determine, by means of a biological test, if these cells observed in several organs are viable or not. In order to assess this point, fragments of lung and the whole spleen were removed from tumor-bearing Wistar rats, at 3, 5, 7, 9, and 11 days after tumor implantation, and transplanted subcutaneously to recipient normal animals. The results are registered in the Table.

No significant differences were seen between lung and spleen with regard to their capacity to give rise to tumors when transplanted. This finding is of particular interest when we consider that it is a well established fact in oncology that the spleen is an exceedingly uncommon site for metastases localization<sup>5,6</sup>. These observations indicate that Yoshida sarcoma cells present in these organs are viable and that their inability to produce a secondary growth, when the organs are *in situ*, must be due to other

Days after tumor implant	Organs transplanted			
	Lung		Spleen	
	Tot. No.	No. Pos.	Tot. No.	No. Pos.
3	4	0	5	0
5	4	3	5	2
7	4	4	5	4
9	4	4	5	5
11	4	4	5	5

<sup>1</sup> H. C. ENGELL, *Acta chir. scandinav. Suppl.* 201, 1 (1955).

<sup>2</sup> R. PIMENTA DE MELLO, *Hospital (Rio de J.)* 57, 119 (1960).

<sup>3</sup> A. A. SANDBERG and G. E. MOORE, *J. Nat. Cancer Inst.* 19, 1 (1957).

<sup>4</sup> T. YOSHIDA, *Acta Union int. contra Cancrum* 16, 496 (1960).

<sup>5</sup> R. A. WILLIS, *The Spread of Tumors in the Human Body* (Butterworth Co., London 1952).

<sup>6</sup> I. ZEIDMAN, *Cancer Res.* 17, 157 (1957).

conditions not related to their intrinsic aptitude to multiply.

**Zusammenfassung.** Werden Lunge und Milz von Yoshida-Sarkomratten bereits fünf Tage nach der Tumortransplantation entnommen und in normale Ratten implantiert, so zeigt der Umstand, dass sich aus diesen Organen Tumoren entwickeln, *in situ* belassen aber keine Metastasenknötchen bilden, dass dies auf andere Bedin-

gungen als solche, die mit der Lebensfähigkeit der Zellen zusammenhängen, zurückzuführen ist.

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### Changes in the Distribution of Glucose-<sup>14</sup>C in Alternative Catabolic Pathways Induced by Kinetin-Analogue in the Callus Tissue of Carrot (*Daucus carota* L.)

It is known that kinetin affects growth and differentiation of tissue callus cultures<sup>1</sup>. It appeared to be of interest whether the action of this type of substance bears some relationship to the regulation of the activity of alternative pathways of glucose catabolism, as had been demonstrated that  $\beta$ -indoleacetic acid<sup>2,3</sup> and animal hormones<sup>4,5</sup> are able to change the distribution of glucose in these pathways.

This report contains the results of measurements of the production of <sup>14</sup>CO<sub>2</sub> from glucose-(1-<sup>14</sup>C) and glucose-(6-<sup>14</sup>C) by the callus tissue of carrot (*Daucus carota* L.) after a short treatment with 6-benzylaminopurine. The experimental material was obtained from the Institute for Plant Physiology of Poznań (Poland). The tissue was passaged four times in the course of four months in our laboratory and maintained on a modified GAUTHERET's medium<sup>6</sup> at 22–24°C in daylight. Callus tissue was sectioned to pieces about 1–3 mm<sup>3</sup>, and washed in water. 0.5 g material thus prepared was incubated in 50 ml Erlenmeyer flasks with 6 ml 0.033 M phosphate buffer of pH 5.3, containing different concentrations of 6-benzylaminopurine which was synthesised at the Department of Radiobiology of this Institute. Darkened flasks were shaken in a water bath at 25°C. After 2 h, 1 ml glucose-(1-<sup>14</sup>C) or glucose-(6-<sup>14</sup>C) of a total activity of 330  $\mu$ C and

mass of 4 mg was added. Carbon dioxide was trapped in 28% KOH placed in a cup suspended from the stopper of the flask. After terminating the incubation, the K<sub>2</sub><sup>14</sup>CO<sub>3</sub> was converted to Ba<sup>14</sup>CO<sub>3</sub>,<sup>7</sup> which was collected on a filter paper. This filter paper was then used for measuring the radioactivity with an end-window GM counting tube (Frieseke-Hoepfner apparatus). The values of radioactivity were referred to zero mass of Ba<sup>14</sup>CO<sub>3</sub>. The efficiency of measurement was 1  $\mu$ C = 10<sup>6</sup> c.p.m.

As may be seen from the Table 6-benzylaminopurine affects the release of <sup>14</sup>CO<sub>2</sub> from glucose-(1-<sup>14</sup>C) and from glucose-(6-<sup>14</sup>C) in the course of a 4 h incubation of tissue with glucose so that the ratio of C<sub>6</sub>/C<sub>1</sub> is somewhat raised. This effect, indicating the change of the ratio between the alternative respiratory pathways, is marked at concentrations when the total release of <sup>14</sup>CO<sub>2</sub> is reduced (0.80 and 4.00 mg/l) as well as at the lowest concentration used when 6-benzylaminopurine is observed to stimulate the overall formation of <sup>14</sup>CO<sub>2</sub> (0.20 mg/l).

It follows from the above results that 6-benzylaminopurine exhibits a pronounced effect on the catabolism of glucose-<sup>14</sup>C in the callus tissue of carrot in the sense that it decreases the activity of the pentose cycle and increases that of glycolysis.

**Zusammenfassung.** Im Callusgewebe von *Daucus carota* L. wurde mittels speziell markierter Glucose nach 6stündiger Behandlung mit 6-Benzylaminopurin (0,2–4,0 mg/l) der vergrößerte Anteil der Atmung vom glycolytischen Typ festgestellt.

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The effect of 6 h treatment with 6-benzylaminopurine on the catabolism of specifically labelled glucose in the callus tissue of carrot (*Daucus carota* L.). The tissue was pretreated with 6-benzylaminopurine for 2 h, whereafter labelled glucose was added and incubation prolonged for further 4 h

Concentration of 6-benzylaminopurine in mg/l in solution with labelled glucose	Radioactivity of Ba <sup>14</sup> CO <sub>3</sub> in c.p.m./0.1 g dry weight. Mean values and standard deviations		C <sub>6</sub> /C <sub>1</sub> and coefficient of variation %
	Glucose-(1- <sup>14</sup> C)		
	-(6- <sup>14</sup> C)		
0	1885 ± 11	662 ± 6	0.35 ± 3.4
0.20	1910 ± 12	723 ± 7	0.38 ± 3.4
0.80	1666 ± 11	740 ± 7	0.44 ± 3.5
4.00	1481 ± 10	773 ± 7	0.52 ± 3.5

### Über die Beziehung zwischen Muttertier und Jungen beim Mufflon (*Ovis aries musimon*, Pall.)

SCHLOETH<sup>1</sup> gibt an, dass sich bei Camargue-Rindern Mutter und Kind gegenseitig optisch und akustisch kennen. Über die Mutter-Kind-Beziehung entwickelt sich nach COLLIAS<sup>2</sup> und SCOTT<sup>3</sup> bei Schafen und Ziegen die

Herdensozietät. HEDIGER<sup>4</sup> lässt offen, ob sich diese Beziehung durch Prägung im Lorenzschen Sinn oder durch

<sup>1</sup> R. SCHLOETH, Säugetierkundl. Mitt. 5, 145 (1958).

<sup>2</sup> N. E. COLLIAS, Ecology 37, 228 (1956).

<sup>3</sup> J. P. SCOTT, Animal Behaviour (Univ. Press, Chicago 1958).

<sup>4</sup> H. HEDIGER, Tierpsychologie im Zoo und im Zirkus (Verlag Reinhardt, Basel 1961).