Perhaps it will be possible by further pursuing these researches to learn something about the contribution of the endocrine glands to the diabetic disturbance of the metabolism.

L. LASZT.
H. VOGEL.

Department of Physiology,
Nutrition Division,
University, Fribourg.
Jan. 21.

[1] Laszt, L., *Experientia*, **1**, 7 (1945).

Thermostable Endotoxin of Rickettsiae

Kligler and Oleinik[1] reported the presence of a labile toxin in yolk-sac cultures of Rickettsiæ. The following experiments were designed in order to test whether the toxic effects of Rickettsiæ are wholly brought about by this labile toxin, or are due also to the action of a thermostable toxin.

Rickettsiæ suspensions were prepared as described by Kligler and Oleinik[1]. The micro-organisms were thrown down by centrifugation and suspended in saline at a concentration of at least 6 × 10⁸ micro-organisms per c.c. of suspension. The suspension was then heated for two hours at 100° C.

The following effects were produced by heated suspensions of *R. prowazeki* or *R. mooseri* in rats weighing 25–30 gm. One hour after intra-abdominal injection of 0·5–1·0 c.c., there was a drop of body temperature to less than 35·0° C. as observed by Zahl and Hutner[2] after injection of Gram-negative organisms into mice: a further effect induced by the same injection was decrease of liver glycogen. In one experiment the value for the controls ranged about 2·5–3·0 gm. per cent, while the corresponding value for rats treated with Rickettsiæ was 0·7–1·0 gm. per cent. Cameron, Delafield and Wilson[3] found a similar change in rats which were injected with a toxic fraction of *Salmonella typhimurium*. In guinea pigs, leucopenic reactions occurred 1 hour after an intraperitoneal injection of 1 c.c. of the Rickettsia suspension. The observed decreases in the numbers of the white cells were of the order of more than 50 per cent of the values found at the beginning of the experiment. In this effect, Rickettsiæ resemble Gram-negative micro-organisms[4]. The Schwartzman[5] technique applied to the Rickettsiæ revealed neither the preparing nor the injury-producing factor. The results of a typical experiment are tabulated below:

Agent used for skin preparation	Size of skin lesion at site of intracutaneous injection after intravenous injection of	
	R. prowazeki	*Proteus X* 19
Proteus X 19	No reaction	60 mm. × 35 mm.
S. dysenteriæ	,, ,,	30 ,, × 25 ,,
Myc. butyricum	,, ,,	10 ,, × 10 ,,
Cor. xerose	,, ,,	No reaction
R. prowazeki	,, ,,	,, ,,

3 × 10⁸ micro-organisms heated at 100° C. for two hours were injected intracutaneously to effect skin preparation. At least 2 × 10⁹ micro-organisms were used in the intravenous injection. The experiments with Rickettsiæ were also performed with twice this number of micro-organisms, but again yielded negative results.

Summarizing, it can be said that although Rickettsiæ contain neither the preparing nor the injury-producing factors of Schwartzman, they are capable of eliciting most of the endotoxic effects that are usually produced by Gram-negative Eubacteriales.

L. OLITZKI.
E. OLEINIK.
J. W. CZACZKES.
A. KUTZENOK.

University-Hadassah Typhus Laboratory,
Department of Hygiene,
Hebrew University,
Jerusalem.
Jan. 8.

[1] Kligler, I. J., and Oleinik, E., *Nature*, **154**, 462 (1940).
[2] Zahl, P. A., and Hutner, S. H., *Proc. Soc. Exp. Biol. and Med.*, **56**, 156 (1944).
[3] Cameron, G. R., Delafield, M. E., and Wilson, J., *J. Path. and Bact.*, **51**, 223 (1940).
[4] Olitzki, L., Avinery, Sh., and Bendersky, J., *J. Immunol.*, **41**, 361 (1941).
[5] Schwartzman, G., "Phenomena of Local Tissue Reactivity" (Paul B. Hoeber, Inc., New York, 1937).

Dose Control in Radiotherapy

A LIMITED attack has been made on the problem of X-ray skin tolerance dose-levels as related to the size of the area treated and the extent of the non-irradiated neighbourhood.

It was found in animal experiments[1] that the severity of skin reactions to X-rays depends on the size of the area irradiated, and by means of various devices (grid, sieve, anulus, etc.) it was shown that, the dose per unit area remaining unchanged, the reaction becomes a minimum when the normal tissues can exert their maximum protective role. A relationship of a function of the perimeter/area ratio and the dose required to produce similar degrees of reaction was surmised. Following lines of approach similar to those pursued by Lecomte du Nouy[2] in the studies of cicatrization of wounds, a function of the cube root of the perimeter/area ratio was found significant. In an endeavour to avoid permutation of dosages, a formula has also been studied which, taking into account the area and its shape, would fit the relevant clinical data.

Detailed data will be published shortly.

In radium therapy the dosage system of Paterson and Parker[3] has withstood the test of clinical application. However, only the maximum and minimum range of doses was suggested. The penumbral zone of normal tissues outside the irradiated volume which exert an influence on the reaction, both by removing the tissue demolition products and by supplying nutritional fluids and contributing to repair, should be taken into account in the assessment of the dose to be delivered.

Introducing in the computation the shell area/volume ratio index, relative optimal effective dosage figures for volumes of different sizes and shapes can be obtained[4].

In the treatment of malignant tumours, the maintenance of an efficient connective tissue is probably of no less importance than the delivery of a certain dose of radiation. This is in harmony with experimental findings in genetics[5] showing that the effect of ionizing radiations on cells is not the whole story of the events leading to the resolution of tumours, but that there are other concomitant physiological factors. Clinical data corroborate also the above statement.

The suggested approach allows the assessment of workable dosage-levels for different volumes of tissue without resorting to empiricism and leads one to consider the implanted volume not in an abstract way but as an integral part of the living body.

B. JOLLES.

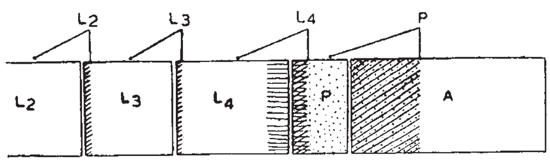
Radiotherapy Department,
General Hospital,
Northampton.
Dec. 15.

[1] Jolles, B., *Brit. J. Radiol.*, **14**, No. 159 (March 1941).
[2] Lecomte du Nouy, *J. Exper. Med.*, **29** (1919).
[3] Paterson, R., and Parker, H. M., *Brit. J. Radiol.*, **7** (Oct. 1934); **11** (1938).
[4] Jolles, B., *Brit. J. Radiol.*, in the press.
[5] Muller, H. J., *J. Gen.*, **40** (1940).

Concealed Phases in the Metamorphosis of Insects

WORK on the biology of insects is becoming more exact than formerly. Attempts are more frequently made to control with precision some of the physical factors, particularly temperature and humidity; and it is not uncommon to find that the mean and standard deviation are given of the temperature and duration of the various instars. In some instances, even the wave-length of the light to which the insects were exposed during the experiment is stated. This increasing accuracy has not, curiously enough, resulted in a closer examination of some of the assumptions that are universally made.

In biological work in which the different instars are distinguished, it is generally regarded as axiomatic that the shedding of the cuticle marks the end of one instar or stage and the beginning of another. Snodgrass[1] and a few others have directed attention to the fact that the beginning of a new instar does not date from the shedding of the old cuticle but begins when the old cuticle is loosened from the epidermis and a new cuticle is secreted beneath the old. The new instar thus invariably appears several hours to two or three days or more before the cuticle of the previous instar is shed. This fact should be kept in mind when interpreting published figures for the duration of the larval instars, but is usually of no importance so long as it is recognized that the moults serve as convenient landmarks in the life of the larva.



pharate phases.
pupal instar, as usually reckoned.
'prepupal instar'.

The precise discrimination of the instars is, however, of considerable importance when dealing with the adult or imaginal stage. The imaginal stage of the Holometabola consists of two instars[2,3], the first of which is known as the pupa and is usually spoken of as the pupal stage. The duration of the pupal or first imaginal instar is usually reckoned from the time the last larval skin is shed to the time the adult emerges from the pupal cuticle, and any activities between these moults, such as wriggling, crawling, or swimming, are attributed to the pupa. I think that the three examples given below clearly show that our conception of certain biological phenomena is fundamentally altered when the instars are more accurately defined.

1. One beetle and a few flies are the only insects known to reproduce pædogenetically. A gnat, *Tanypus boiemicus* Kieff., is unique in that it has[4–7] a pædogenetic pupa. The imago may lay eggs a few minutes after shedding the pupal cuticle, or it may fail to shed the effete pupal cuticle and lay its eggs in the pupal case. The failure of the imago to shed its pupal cuticle cannot alter the fact that it is an imago at the time the eggs are laid, as is very clear from Zavrel's[7] figure. This, therefore, is an example of a parthenogenetic species and not a pædogenetic one.

2. According to Weber[8], Imms[9], and nearly all other writers, the pupæ of the Megaloptera and Neuroptera are able to walk and the pupæ of the Trichoptera are able to swim. In every case it is the adult, enclosed in the now effete pupal cuticle, that is crawling or swimming.