- (3) HOTCHKISS reaction and metachromasia have given clear positive slide images in the central nervous systems of fishes, amphibia and lower mammalia such as rodents and chiroptera<sup>1</sup>. Doubtful and weak colorations were obtained with the central nervous system of birds and higher mammalia<sup>2</sup>.
- (4) Histochemical colouring methods may not give true images on structure and position of metachromatic and Hotchkiss positive substances.

It is well known that these histochemical reactions involve precipitated material in a form which is probably far from what it is in real life.

Nevertheless, slide images obtained by infiltration of coloured materials (such as India ink), in the nervous tissue centres and successive glia coloration clearly help to affirm that hyaluronidase sensitive substances are lodged in the so-called interstitial spaces of the central nervous system.

- (5) The data hereby reported agree with the old ideas expounded by such anatomist as Deiters, Virchow and Gierke<sup>3</sup> whereby the existence of an homogeneous substance between the neuroglia, nerve cells, fibres and axon terminations.
- (6) Systematic comparison between histochemical reactions and glia make-up, show, up to now, that the intercellular matrix of white matter has a certain antagonism for histochemical reactions and fibrous glia development. Nevertheless, when the fibrous glia are highly developed the histochemical reactions are very weak.

As our researches were approaching completion, many other authors reported works with data agreeing in many respects with our experimental results which will soon be subject of a separate report.

FREEDMANN<sup>4</sup> has published a brief report whereby the application of his experimental method demonstrates the existence of Hyaluronidase/Hyaluronic acid systems in the intercellular matrix of the gray matter of the central nervous system.

Hess<sup>5</sup> has shown, with his histochemical methods, the existence of mucopolysaccharides in the intercellular matrix of the gray matter of the central nervous system of the different mammalia. In all probability the mucopolysaccharides form the ground substance of the central nervous system. The experimental data presented by Hess agree fully with the results obtained from our researches.

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## Résumé

Réactions histochimiques et enzymatiques, l'action positive du facteur diffuseur de Duran-Reynalds montrent que dans les espaces intercellulaires du système nerveux central des vertébrés existent des substances mucopolysaccharides. La glie (Virchow) est constituée par une substance amorphe mucopolysaccharide et par la texture glio-vasculaire.

- <sup>1</sup> A. BAIRATI jr. and M. PATERNO, Atti Soc. it. Anat. Napoli 1952, pubbl. in Monit. Zool. it. Suppl. 61, 107 (1952) (fishes). E. BARTOLI and G. BERTACCINI, ibidem (birds) pag. 109. H. GIERKE, Arch. mikr. Anat. 25, 95 (1885): 26, 110 (1886).
- mikr. Anat. 25, 95 (1885); 26, 110 (1886).

  <sup>2</sup> F. Massari and G. Marsico, Atti Soc. it. Anat. Napoli 1952, pubbl. in Monit. Zool. it. Suppl. 61, 110 (1952) (mammalia). H. Gierke, Arch. mikr. Anat. 25, 95 (1885); 26, 110 (1886).
  - <sup>3</sup> H. GIERKE, Arch. mikr. Anat. 25, 95 (1885); 26, 110 (1886).
  - <sup>4</sup> B. Freedmann, Anat. Rec. 115, 265 (1953).
  - <sup>5</sup> A. Hess, J. Comp. Neurol. 98, 69 (1953).

## The Significance of Temperature and the Daily Light – Dark Period in the Formation of Resting Buds

As has already been shown, increased storage temperatures are able to produce in resting winter buds a further deepening of dormancy<sup>1</sup>. Furthermore, those buds which had recently emerged from dormancy could be rendered dormant again by raising the temperature sufficiently<sup>2</sup>, this secondary resting period exhibiting all the typical characteristics of the first "natural" one. These observations, coupled with the fact that in the case of most plants the resting period begins in the summer, have convinced the author that the "natural" resting period is mainly due to the influence of high summer temperatures<sup>3</sup>.

In the present communication, an attempt has been made to investigate the influence of temperature and photoperiodism upon the formation of resting buds. It has become clear that here, too, temperature is the main factor determining the occurrence or non-occurrence of such dormancy. Within certain limits, however, the effect of this factor is suppressed by the duration of the daily light period.

The experimental material comprised plants of *Hydrocharis morsus ranae L*. which had been allowed to develop from winter buds, the dormancy of the latter having been broken by storing them at 5°C. The plants were maintained in tap water at temperatures of 10, 15, 20, and 25°C, respectively, both in the dark and under artificial daylight. The daily periods of illumination lasted, respectively, 3, 6, 9, 12, 15, 18, and 21 h, as well as continuous illumination.

The results clearly indicate that the environmental temperature is of decisive importance in inducing the formation of dormant buds. At 10°C, and independent of the length of the daily light-period, no resting buds occur, the plants continuing their growth by the production and development of non-resting, stolon-forming buds. On the other hand, resting buds tend to occur at 15, 20, and 25°C, though only when the daily light-period is not too long.

From a photoperiodic standpoint, the formation of resting buds at the above temperatures must be considered a short-day character, i.e., it occurs when the duration of the daily light-period is neither too long nor too short. Resting buds are not produced under conditions of continuous illumination. Here a decisive factor in the initiation of resting-bud formation is the duration of the daily dark-period, the length of which must not lie below a definite minimum value.

Resting buds are also formed by plants maintained entirely in the dark, on a saccharose solution, at 18-22°C room temperature. From this it may be concluded that the significance of the daily light-period lies only in its effect on the synthesis of carbohydrates which are necessary not only for the formation of resting buds but also for storage in the latter or in the adjacent tissues.

If the duration of the daily light-period exceeds the length characteristic for each temperature, light inhibits the formation of resting buds. Where the daily light-period is too long, i.e., where the corresponding dark-period is too short, plants maintained at temperatures of 15°C and higher continue growth by the formation of non-resting stolon-forming buds.

- <sup>1</sup> A. Vegis, Symbolae Bot. Upsalienses 10, 2 (1948).
- <sup>2</sup> A. Vegis, Physiol. Plant. 2, 117 (1949).
- <sup>3</sup> A. Vegis, Svensk Bot. Tidskr. 43, 671 (1949).

In comparing the shortest light-periods which are essential, at the various temperatures, for stolon formation, i.e., for continued growth, it is obvious that the lower the temperature the shorter the light-period necessary to bring such growth about. However, as the temperature rises, the daily light-period necessary for continued growth becomes correspondingly longer.

On the other hand, the maximum daily light-period which permits the formation of resting buds becomes longer as the temperature rises, while the dark-period, essential to resting-bud formation, becomes correspondingly shorter. Thus at 15°C, with a daily dark-period of only 3 h, no dormant buds are formed. On the other hand, 50% of the plants formed resting buds when the dark period was increased to 6 h, and 100% when the dark-period was 9–18 h long. A 3-hour dark-period at 20°C caused resting-bud formation in 4 out of 10 plants, while a dark-period of 6–18 h at the same temperature caused resting buds to form on all plants. Again, all plants produced resting buds at 25°C and a dark-period 3–18 h long.

Though higher temperatures were not used in these experiments, the rapid decrease observed in the minimum daily dark-period required to initiate resting-bud formation leads us to expect that with a further increase in temperature even continuous illumination could not hinder the formation of resting buds. This means that after a certain temperature has been reached, formation of resting buds is apparently independent of the length of the daily light-period. That is to say, the short-day reaction is substituted by a day-neutral one. Such a change in the photoperiodic behaviour of plants, as regards the initiation of flower formation, has been reported in a number of cases¹.

In this manner, light and temperature are antagonistic factors. While light promotes growth and inhibits the initiation of resting-bud formation, a high temperature suppresses growth and provides the conditions necessary for the formation of resting buds. The light factor, acting a minimum number of hours per day, is able to neutralize the inhibiting effect of temperature, provided that the latter does not exceed a certain limit. As the temperature rises, however, the length of the light period must be increased. Finally, a point is reached where even continuous illumination can no longer compensate for the inhibiting effect of temperature.

The inhibitory effect of light on the formation of resting buds and on the initiation of the dormant condition is likely due to the presence of auxin², and possibly to other active substances formed in the leaves. The fact that resting-bud formation is promoted by a high temperature is manifestly connected with the appearance of inhibiting substances³. It should be noted in this connection that Snow⁴, with regard to the occurrence of correlated inhibitions, has suggested an antagonistic effect between auxin and inhibiting substances.

By changing the particular time at which the plants are exposed to the temperature and photoperiodic conditions favouring resting-bud formation, the latter can be promoted at any season. On the other hand, if the

<sup>4</sup> R. Snow, New Phytologist 36, 283(1937).

plants are cultivated during that season when dormant buds are normally formed in nature, and under the conditions of temperature and light favourable to growth, no resting buds are formed. All these observations negate the existence of an endogenous annual rhythm.

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## Résumé

Les bourgeons dormants ne se forment que dans le cas où la température dépasse un certain niveau critique (entre 10 et 15°C) correspondant à une période quotidienne d'obscurité qui ne doit pas s'abaisser au delà d'une certaine durée minimale. Plus la température monte, plus cette durée peut être raccourcie. Dès l'instant où ces conditions ne sont pas réalisées, l'accroissement continue.

## Teneur en acide ribonucléique de différents génotypes chez *Drosophila melanogaster*

On tend actuellement à attribuer à l'hétérochromatine un rôle dans la synthèse des acides nucléiques. Caspersson et Schultz¹ ont montré que la teneur en A.N. (déterminée par absorption en lumière ultra-violette) du cytoplasme d'oocytes et celle des nucléoles et des chromosomes de glandes salivaires subit des variations liées à la teneur du noyau en hétérochromatine. Callan² d'autre part, se basant sur des déterminations chimiques, n'a pas observé d'influence due à la présence d'un chromosome Y supplémentaire sur la teneur en A.R.N. du cytoplasme de l'œuf.

Nous avons tenté de vérifier chez Drosophila melanogaster si des variations importantes du génotype ou du phénotype sont accompagnées de variations dans la teneur en A.R.N. de l'organisme.

Un premier résultat (Tabl. I) se rapporte à la teneur en A.R.N. de mâles normaux (XY) et de mâles sans Y (XO). Deux cultures parallèles, issues de parents isogéniques, fournissent les deux types de mâles. La teneur en A.R.N., déterminée par la méthode de Ogur et Rosen³ et rapportée au poids sec, diffère de moins de 5% pour les femelles et les mâles de ces deux cultures. De plus, comme les femelles de deux générations sont génétiquement identiques, le rapport entre les valeurs obtenues pour les femelles et les mâles nous permettrait de déceler un écart dû au génotype des mâles. Or ces deux rapports sont identiques. Nous devons donc admettre que l'absence du chromosome Y, malgré sa richesse en hétérochromatine, est sans effet sur la teneur globale de l'organisme en A.R.N.

Un deuxième résultat se rapporte à un type Minute, M² caractérisé par une taille réduite, des soies diminuées et une durée de développement prolongée de deux jours. Ce même phénotype peut être déterminé par de nombreuses mutations distribuées tout au long des quatre chromosomes. Les types Minutes constituent donc une classe assez particulière de mutants. On les a parfois interprétés comme étant dûs à des déficiences de régions

<sup>&</sup>lt;sup>1</sup> R. H. ROBERTS and B. E. STRUCKMEYER, J. Agric. Res. 56, 633 (1938); 59, 699 (1939).

<sup>&</sup>lt;sup>2</sup> A. Vegis, Acta Soc. Biol. Latviae 7, 87 (1937). – H. U. Amlong and G. Naundorf, Gartenbauwiss. 12, 116 (1938). – J. P. Bennett and F. Skoog, Plant Physiol. 13, 219 (1938). – G. Borgström, The transverse reactions of plants, Thesis, Lund (1939).

<sup>&</sup>lt;sup>3</sup> T. Hemberg, Acta Horti Bergiani 14, 133 (1947); Physiol. Plant. 2, 24 (1949); 2, 37 (1949); 4, 437 (1951); 5, 115 (1952).

<sup>&</sup>lt;sup>1</sup> T. Caspersson et J. Schultz, Nature 142, 294 (1938). – J. Schultz et T. Caspersson, Arch. exp. Zellforsch. 22, 650 (1939).

<sup>&</sup>lt;sup>2</sup> H. G. Callan, Nature 161, 440 (1948).

<sup>&</sup>lt;sup>3</sup> M. Ogur et G. Rosen, Arch. Biochem. 25, 262 (1950).