Relative Phase Difference as an Index of the Quantity of the Secretory Material in the Neurosecretory Cell of *Iphita limbata*

In an attempt to devise a quantitative method to estimate the neurosecretory material in the cell, the refractive index of the cytoplasm of the ordinary neurone and of the neurosecretory cell was studied using BARER's[1] double-immersion method. The brain of the plant bug *Iphita limbata* Stal. was processed as outlined in an earlier work[2]. Phase measurements on the cytoplasm of the neurosecretory cells of pars intercerebralis and that of the adjacent ordinary neurones were taken when the sections were immersed in liquid paraffin which was successively displaced thoroughly by xylol and methyl salicylate by the irrigation technique[3]. Phase measurements of the same cells along the same region were taken when the cells were immersed in methyl salicylate. An AO Baker interference microscope with $\times 40$ shearing system and a tungsten lamp with green filter were used for the present study. Measurements were taken only where there was no overlap of the images. Refractive indices of the immersion media were found by means of an Abbe refractometer. Refractive index of the neurosecretory cell and of the ordinary neurone was calculated[4]. It was found that the refractive index of the fixed and dehydrated neurosecretory cell was 1.568 ± 0.017 and that of the ordinary neurone, 1.567 ± 0.013, which were not significantly different from one another. These findings are in good agreement with those of BARER and JOSEPH[5] for the refractive index of pure protein (1.58).

Though these dingings revealed that in fixed and dehydrated sections neurosecretory cells and ordinary neurones had identical refractive indices, the neurosecretory cells showed higher phase retardation than the ordinary neurones in similar preparations[2]. As the optical path is the product of thickness and refractive index, the concentration of dry matter in the neurosecretory cell and the ordinary neurone is presumably expressed in thickness of these cells when fixed and dehydrated. So the relative phase difference of the neurosecretory cell in tissue sections, which may be considered as the ratio of phase retardation of the neurosecretory cell divided by phase retardation of ordinary neurone in the same section and in the same medium, should represent correctly the ratio of their concentration of dry matter during live condition. The fact that the relative phase retardation of the neurosecretory cell is identical in liquid paraffin (2.028 ± 0.405) and in methyl salicylate (2.027 ± 0.497) within the limits of experimental error, supports this view.

This hypothesis was further evaluated in the following study. Brains of female *Iphita* in the nearly gravid condition when the neurosecretory cells were full, as well as of animals which just started oviposition when the cells were depleted[6], were dissected out, processed and phase measurements in liquid paraffin taken as described above. In all cases, phase retardation value of the individual neurosecretory cell in each brain section was divided by the phase retardation value of an adjacent ordinary neurone in the same brain section, and its relative phase retardation calculated. Measurements were taken on all neurosecretory cells whose thickness as judged visually was not less than the thickness of the section. The findings are summarized in the Table. It may be seen that the relative phase retardation of the neurosecretory cell of the animals after oviposition was only 1.753 ± 0.385, which is considerably lower than the value for neurosecretory cells from females before oviposition (2.028 ± 0.478).

Slides were subsequently stained by GOMORI's[7] method and the observations[6] confirmed. It is concluded that the relative phase retardation of the neurosecretory cell defined as phase retardation of the neurosecretory cell relative to that of the ordinary neurone may be taken as an index of the quantity of the secretory material in the cell[8].

Zusammenfassung. Die sogenannte Phasenverzögerung in neurosekretorischen Zellen beim weiblichen Käfer *Iphita limbata* vor und nach der Eiabsonderung ist verschieden und wird als Index für die Quantität der neurosekretorischen Substanz betrachtet.

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Relative phase retardation of the neurosecretory cells when mounted in liquid paraffin, in nearly gravid *Iphita* and in those which have started oviposition

	Animal almost gravid	Started oviposition
Animals	4	4
Brain sections	113	108
Neurosecretory cell sections	260	264
Neurones	132	108
Relative phase retardation in liquid paraffin (mean $\pm$ S.D.)	2.028 ± 0.478	1.753 ± 0.385

[1] R. BARER, Nature *172*, 1098 (1953).
[2] V. K. K. PRABHU, Proc. Sem. Int. Cell Biology, Bombay (1966), p. 428.
[3] R. BARER and S. JOSEPH, Q. J. microsc. Sci. *96*, 423 (1955).
[4] R. BARER and D. A. T. DICK, Expl. Cell Res., Suppl. *4*, 103 (1957).
[5] R. BARER and S. JOSEPH, Q. J. microsc. Sci. *95*, 399 (1954).
[6] K. K. NAYAR, Proc. Ind. Acad. Sci., B. *47*, 233 (1958).
[7] G. GOMORI, Am. J. Path. *17*, 395 (1941).
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Hemmung von SH-Enzymen durch Ethacrynsäure

DUGGAN und NOLL[1] machen die Blockierung essentieller SH-Gruppen für die Wirkung von Ethacrynsäure (2, 3 - Dichlor - 4 - (2 - methylenbutyryl) phenoxyessigsäure) verantwortlich. Die Hemmung gewisser Energieübertragungsprozesse in Mitochondrien[2-4] kann ebenfalls auf diese Weise gedeutet werden; höchstwahrscheinlich ist ja eine SH-Gruppe an einem Zentrum, das zwischen Atmungskette und dem Oligomycinangriffspunkt liegt für die Energiekonservierung von ausschlaggebender Bedeutung[5-7].