

Wochen beobachtet wurden – lebten in Aquarien von ca. 80 l Inhalt, an deren Stirnseiten Aluminiumblechelektroden angebracht waren. Die Temperatur des Wassers wurde durch einen Reglerheizer konstant auf 26°C gehalten. Während der Beobachtungen wurde die Heizung ausgeschaltet. Ein Absinken der Temperatur um 2–3°C (grössere Temperaturschwankungen sind infolge der relativ hohen Raumtemperatur nicht aufgetreten) zeigte keinen deutlichen Einfluss auf die Entladungen.

Die Entladungen wurden mit Hilfe eines Elektronenstrahloszillographen (Philips GM 3156/01) beobachtet und mit einem Elektrokardiographen (Siemens Einkoffer-Elektrokardiograph) aufgezeichnet. Die Registrierungen erfolgten stets erst 1 h nach dem Einschalten der Geräte. Ferner wurden nur die Registrierungen ausgewertet, die von Tieren stammten, die bereits mindestens 2 Tage unter Untersuchungsbedingungen lebten. Eine Ausnahme hiervon stellen allerdings die Registrierungen des Kampfverhaltens dar, bei denen ein Tier zu einem im Versuchsbekken befindlichen hinzugesetzt wurde und dessen Entladungen sofort registriert wurden. Beim Einschalten der Apparatur wurde des öfteren die Elektrode angegriffen. Das Verhalten hierbei unterscheidet sich nicht von den ersten Phasen des Kampfverhaltens.

Resultate. Bei Ruhe erfolgen die Entladungen in den allermeisten Fällen mit einer Frequenz von 5–15 Hz (Figur, a), die Stärke der Einzelimpulse liegt bei einem Abstand von 25 cm zu Elektrode um 45 mV.

Beim langsamen Schwimmen (Figur, b) wird die Frequenz etwas höher (15–25 Hz), bei schnellem Schwimmen

(Figur, c) erfolgen die Entladungen mit einer Frequenz von 20–40 Hz. Höhere Frequenzen konnten auch bei Appetenzverhalten registriert werden (bis zu 60 Hz, meist aber zwischen 30 und 50 Hz) (Figur, d).

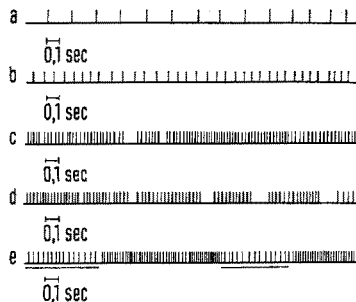
Bei Komfortverhalten wurden keine charakteristischen Entladungen registriert, ihre Frequenzen lagen fast im ganzen Bereich, mit Ausnahme der Extremwerte. Die Tiere zeigen ein eigenartiges Scheuern und Reiben an festen Unterlagen, über die sie sich vorwärts- und rückwärtsschwimmend schieben und keineswegs nur über einen festen Gegenstand schnellen und dabei anstreifen.

Bei Kampfverhalten – bisher wurde nur ein zweites Tier in ein bereits besetztes Becken gesetzt – erfolgt, wie von MÖHRES^{5,6} bereits beschrieben, sofort eine wesentliche Erhöhung der Frequenz der Entladungen. Bei Attacken einzeln gehaltener Tiere auf Metallgegenstände wurden während des Breitseitimonierens Entladungen mit Frequenzen um 40 Hz, bei Rammstössen zwischen 50 und 60 Hz aufgezeichnet. Dass Kampfverhalten durch die Entladungen eines gleichartigen Tieres ausgelöst wird, steht nach den Untersuchungen MÖHRES' ausser Zweifel. Es soll nun in nächster Zeit versucht werden, den Tieren Entladungen von artcharakteristischen Frequenzen und Impulsstärken zu bieten, während sich die Fische allein im Becken befinden.

Summary. Observing the discharge-frequencies of the electrical organ of *Gnathonemus petersii*, it was found that there are characteristic frequencies in relation to different behaviour as: slow and fast swimming, at rest, at appetive and aggressive behaviour. No such characteristic frequencies could be detected in relation to comfort movements.

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Charakteristische Frequenzen von Entladungen bei (a) Ruhe, (b) langsamem Schwimmen, (c) schnellem Schwimmen, (d) Appetenzverhalten, (e) Kampfverhalten (unterstrichen: imponieren, nicht unterstrichen: Rammstoss). Nach Registrierungen umgezeichnet.

Time Required for Heterogenous Induction in Chick Embryo Ectoderm¹

GALLERA² has recently published results that indicate that the time required for neural induction in chick embryo ectoderm is relatively long. Using the living Hensen's node as inductor he showed that at least 6 h of contact is required to produce a neuroid reaction in the ectoderm. Eight and a half h of contact is required to elicit a typically neural response in the ectoderm.

It is well-known that various adult tissues act as heterogenous inductors to produce a neural reaction in amphibian ectoderm. It has also been demonstrated that chick liver and kidney act as heterogenous inductors for chick embryo ectoderm (PASTERNAK and McCALLION³). In the present experiments we have attempted to establish the time required for chick liver to induce a neural response in chick embryo ectoderm.

The chick embryo materials used in our experiments were obtained from White Leghorn eggs supplied by a commercial hatchery. The eggs were incubated at 38°C. Small fragments of liver from 14- to 20-day old embryos were fixed overnight in 70% alcohol. Prior to their use in operations they were washed thoroughly in sterile physiological saline solution. Ectoderm of broad streak stage embryos was used as the reacting material. Two procedures were followed. In the first of these the embryo was explanted, ventral side up, onto a suitable agar medium (BUTROS⁴) and a fragment of liver was inserted between the epiblast and hypoblast. After 4 or 6 h the

¹ Work supported by a grant from the National Research Council of Canada.

² J. GALLERA, *Experientia* 21, 218 (1965).

³ L. PASTERNAK and D. J. McCALLION, *Can. J. Zool.* 40, 585 (1962).

⁴ J. BUTROS, *J. exp. Zool.* 152, 57 (1963).

inductor was removed. The piece of ectoderm that had been in contact with the inductor was excised and raised for a further 20–24 h on fresh culture medium. In control experiments the ectoderm and inductor, together with any hypoblast attached to the inductor, were transferred to fresh culture medium for a further 20–24 h. In the second procedure the hypoblast was removed from a large area of epiblast and the inductor was placed on the ventral surface of the ectoderm. The ectoderm with the inductor attached was excised and cultured in fresh medium. After 4 or 6 h the inductor was removed from the explant which was then cultured for a further 20–24 h. In those cases which served as controls both ectoderm and inductor were left together for 24–28 h.

According to our observations intimate contact between the inductor and the ectoderm is essential for induction. Therefore, all of those explants in which there was any doubt at the time of removal of the inductor that

intimate contact had not been maintained were discarded. This applied especially to those cases in which the inductor was inserted between the epiblast and hypoblast because the latter frequently grew between the inductor and the epiblast.

The explants were fixed in Bouin's fluid overnight, embedded in paraffin, sectioned and stained in Hematoxylin and eosin.

Histological observation of the sections revealed that, of 16 control explants with a sufficient contact between inductor and ectoderm, 10 had begun to differentiate in a neural direction with a clearly recognizable neural plate or neural tube (Figure 1), 4 were composed of more or less neuroid cells, and 2 remained completely undifferentiated. Of the 11 explants from which the inductor had been removed after 6 h of contact, 8 had begun to differentiate in a neural direction (Figure 2), 2 were neuroid, and 1 remained undifferentiated. Of the 9 explants from which the inductor had been removed after 4 h of contact, 1 was neuralized, 3 were neuroid (Figure 3) and 5 remained undifferentiated.

These results would indicate that 4 h of contact between inductor and ectoderm is usually not sufficient for an induction leading to clearly definable neural differentiation. It would seem, however, that 6 h of contact is adequate for such induction. Although the small number of cases does not allow statistical consideration, it is interesting to note that the percentages of neural, neuroid and undifferentiated cases are similar in both the control and the 6-h series (63, 25, 13 and 73, 18, 9 respectively). We know from other observations that chick ectoderm induced by liver after prolonged cultivation will develop into well-differentiated neural tissue. Whether a contact of 6 h would be equal to continuous contact in this respect is yet to be determined.

The results of our experiments with heterogenous inductors suggest an induction time at least 2 h shorter than that suggested by the results obtained by GALLERA with the living Hensen's node as inductor. There is some evidence that the natural inductor is mesodermal elements derived from the node rather than the node itself (HARA⁵ and PASTERNAK and MCCALLION⁶). It may be that the difference in time noted above is the time required for the production of this material in GALLERA's experiments. On the other hand, it may be that the inductor substances are more concentrated in or more rapidly diffusible from the fragments of liver than Hensen's node. It may be deduced from HARA's experiments that not more than 2 h would elapse from the definitive streak stage to the time when the ectoderm in front of the node can be shown to have been induced. Thus, the time required for induction under natural conditions may be even shorter than that observed under the conditions of our experiments.

Résumé. Le foie de l'embryon de poulet de 14–20 jours d'incubation sert d'inducteur pour découvrir la durée nécessaire pour déclencher des inductions neurales dans l'ectoblaste du poulet. Nos résultats ont montré que 4 h de contact entre l'inducteur et l'ectoblaste n'est pas suffisant à l'induction d'une différenciation neurale. Après 6 h de contact l'ectoblaste réagissant se différencie au sens neural.

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Department of Zoology, University of Toronto, Toronto 5 (Canada), 16th May 1967.

⁵ K. HARA, Ph.D. Thesis Utrecht (1961).

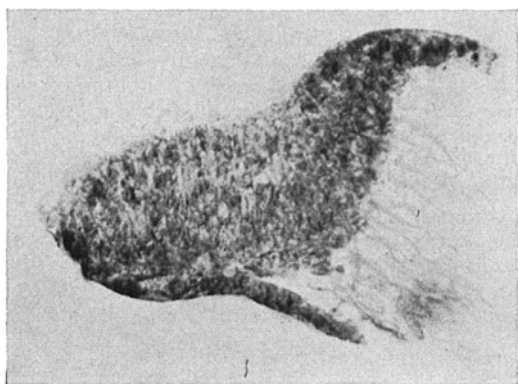


Fig. 1. Neural plate induced by chick embryo liver in continuous contact with the ectoblast. The inductor is seen at the right.

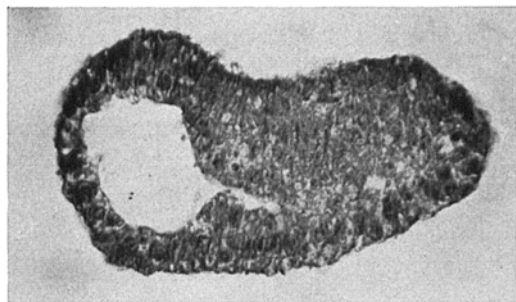


Fig. 2. Neural plate induced by chick embryo liver which had been left in contact with the ectoblast during 6 h.

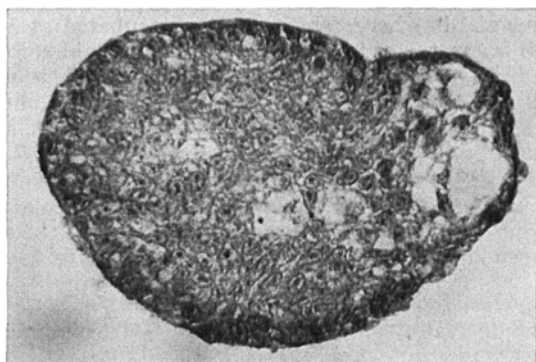


Fig. 3. A neuroid mass induced by chick embryo liver that had been left in contact with the ectoblast during 4 h.