Differentiation Tendencies of Eye Potency Field in Early Chick 
(Gallus domesticus) Embryo*

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Paper includes a preliminary investigation on prosencephalic differentiation tendencies with special reference to differentiation of eye potency field at primitive streak and head process stages of chick embryo. The technique involves intracoelomic culturing of small ectodermal grafts measuring about 0.2 x 0.2 mm prepared from the presumptive prosencephalic area for 12 days and analysing them histologically. The study of 18 grafts reveals that loci for eye field appear anterolaterally on peripheral zone of presumptive prosencephalic ectoderm at primitive streak stage and shift medially during head process stage. Other prosencephalic differentiation tendencies are elaborated at head process stages.

The organs forming potency maps of early chick embryo constructed by Rawles and Rudnick for medium head process and primitive streak stages were based upon differentiation of grafts having all germ layers. Neural differentiation tendencies studied at these stages by Rao were based on differentiation of ectodermal grafts in the absence of other germ layers; but it was not possible for him to specify exact loci of different structural elements of brain on these blastoderms because his grafts were large in size and rectangular in shape. The present investigation was undertaken to study the finer mode of changes of prosencephalic differentiation tendencies with special reference to the differentiation of eye potency field at Hamburger and Hamilton stages 4 and 5 (primitive streak and head process stages). The technique involved intracoelomic culturing of grafts prepared from the presumptive prosencephalic ectoderm during these stages.

Material and Methods

The hatchable red and white crossbreed leghorn eggs were obtained from Naya Bungalow Govt. Poultry Farm, Shillong. They were incubated at 37.5°C. Pans full of distilled water were kept inside incubators to maintain 60-70% humidity. For experiments Hara's guidelines and Spratt's fate maps were followed. The presumptive prosencephalic ectoderm was stripped free of underlying endoderm and mesoderm at H & H stages 4 and 5 and cut into 5 grafts—1 median (M) and 4 peripheral (A, B, C, and D) each measuring about 0.2 x 0.2 mm according to the operation plan shown in Fig. 1. The grafts were transplanted into coelom of another 2½ days old chick embryo and cultured for 12 days. The recovered grafts were fixed in Bouin's—picro—for-
structures were observed. The histological analysis has been compiled in Table 1.

In the 1st set of experiments performed at H and H stage 4, 4 out of 7 grafts showed sensory layer of retina of which 2 were accompanied by pigmented layer. In the 2nd set of experiments at H and H stage 5, when length of the head process ranged from 0 to 0.30 mm, 3 out of 7 grafts showed sensory layer of retina of which 2 were accompanied by pigmented layer. One graft showed only pigmented layer. In the 3rd set of experiments when length of the head process ranged from 0.31 to 0.60 mm, 2 out of 4 grafts showed sensory layer, of which 1 was accompanied by pigmented layer. It can be noted that eye structures were encountered in 3 peripheral (AB, B and C) and 1 median (M) grafts in the 1st set of experiments and in peripheral grafts (A, B and C) in the 2nd set. In the 3rd set they were observed only in the median grafts. These results indicate the possibility that at H and H stage 4 the loci of eye vesicle rudiments first appear in the peripheral region, perhaps antero-laterally in the presumptive prosencephalic ectoderm, and then shift to its median region at H and H stage 5.

Figs 2-6—Cross-sections of grafts showing: (3) telencephalic cortex (TC), (2) Sensory and pigmented layer of retina (SL, PL), (4) prosencephalic neural mass (PNM), (5) feather germs (FG) and (6) cartilage (CAR), x 400

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Discussion

Hara\(^4\) and Rao\(^4\) are the first workers to study the neural differentiation tendencies in early chick embryo by intracoelomic grafting technique. As reported by Rao\(^4\), the neural differentiation tendencies are elaborated during head process stages. Similar observations have been made in the present investigation also. While referring to the differentiation tendencies of the eye, he writes, "Among the prosencephalic structural elements, those belonging to the eye (retina together with tapetum) are the most frequently encountered ones in the grafts from the median areas. In the lateral areas they are on the whole much less frequent." His median grafts were large covering more than half of the presumptive prosencephalic area. In the present investigation, 5 grafts were prepared from this area exploring the possibility of studying much finer mode of differentiation.

The organ forming potency maps of Rawles\(^1\) and Rudnick\(^2\) for medium head process and primitive streak stages show different loci for the eye rudiments. Rawles\(^1\) cultured different pieces of blastoderm on the chorioallantoic membrane and showed that the eye potency field develops from the middle as well as lateral regions of the presumptive prosencephalic area. Her grafts, however, included all the three germ layers and so a clear picture of the eye potency field may not be possible because there was constant interaction between all the germ layers. The grafts in the present series of experiments were stripped free of endoderm and mesoderm and thus, they were purely ectodermal. The lens structures were never observed either by Rao\(^4\) or in the present investigation. Rawles\(^1\) found lens element in 1 out of 18 grafts but she did not show the loci of lens and eye vesicle rudiments separately. The eye potency field as shown in Rudnick's\(^2\) fate map are located just lateral to the Henson's node in the region of presumptive mesencephalic area of Spratt\(^1\). The experiments in the present investigation reveal that the loci of the eye potency field appears laterally on the peripheral region of the Spratt's presumptive prosencephalic area during H and H Stage 4 and shift medially in front of the head process during H and H stage 5. More experimental data would, however, be required for determining the exact loci of the eye potency field. In a review on earlier work Rudnick\(^2\) visualises a continuous medio-lateral separation of eye field in the anterior medullary plate at an early stage. It would be interesting to investigate the pattern of such morphogenetic behaviour.

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