Results and discussion. Distal segments of olfactory vesicles display elliptic transsections, the diameters measuring 1400 Å and 900 Å, respectively. The double membrane is coated on its outside by a thin (60 Å), osmiophilic layer, and, more conspicuous, on its inner aspect by a relatively electron dense, homogenous 100 Å thick film. Exactly in the small axis of the ellipse from this interior coat originate dark staining branches (120 × 100 Å), which split in a fairly constant distance of 500 Å from their origin in 2 V-shaped arms, which in their turn insert into the central tubuli. This pattern was the only one to be observed in a few hundred transsections.

The tubuli are composed by 13 protofilaments, visible only in transsections. Tannic acid outlines the subunits and forms a cog wheel pattern (Figure). The V-shaped arms contact the tubuli in such a manner that 4 protofilaments lie between them in the center of the cilium, and the 9 left filaments remain outside. The interior border of the protofilaments is supported by a thin (30 Å) ring of highly osmiophlic substance. Lateral processes of tubuli that could correspond to dynein arms as seen in various cilia of the 9+2 pattern, were virtually absent in all preparations.

Active movement of olfactory cilia has been a matter of dispute in the past[12]. Since olfactory cilia are likely to harbour receptor sites in their membrane, because of their particulate location in the upper storey of the olfactory surface, active motility was thought to facilitate both contact with stimulating molecules and cleaning of the cell surface after registration of odour substances on the membrane[13].

Direct evidence for ciliary beating has been achieved by direct microscopical observation of the frog olfactory mucosa[6]. The proximal thick segments appeared to beat uncoordinated and very vigorously, especially after elimination of the distal segments. Distal segments seemed to be moved passively and to retard the movements of proximal parts.

The present study demonstrates that tubuli of distal segments lack dynein bridges, which have been convincingly demonstrated to be essential for active movement in human sperm[14]. Since the two tubules found in distal segments seem to be extensions of the central pair in the 9+2 pattern of proximal shafts, the lack of dynein bridges is actually not surprizing, because they are devoid of these arms from their origin. According to the sliding filament hypothesis the cental pair is the anchoring site of radial spokes of peripheral doubletts. It mediates the conversion of the linear sliding of peripheral tubules into a bending of the whole cilium[7]. Thus, the possibility is excluded that the central pair of microtubuli acquires dynein bridges somewhere distal from the 9+2 portion.

As in many other instances, the tubuli are composed by 13 protofilaments, which are very prominent in transsections after tannic acid impregnation[9]. The osmiophilic submembranous coat in cilia may play an active role in the stabilization of intramembranous particles in olfactory cilia, as recently demonstrated in abundance by freeze etching[8]. The Y-shaped connections of this layer with the tubuli may provide an inner skeleton for distal ciliary segments, resembling the horizontal interconnecting filaments in microvilli of the intestinal brush border[15].

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Failure of ^{3}H-Serine to Induce Radioactivity in Presumed Glycinergic Retinal Neurons[1]

B. BAUER and B. EHINGER

Department of Histology, Biskopsgatan 5, S–22362 Lund (Sweden), 24 May 1976.

Summary. ^{3}H-serine does not label retinal neurons selectively when injected intraocularly in rabbits, as would have been expected if it had been converted to neutrotransmitter glycine. The reason is unknown, but one possibility is that the conversion was blocked during the conditions of the experiment.

Glycine has recently been advanced as a putative retinal transmitter in rabbits. It is present in the retina in freely extractible form in comparatively high concentration[2,3]; it is actively and selectively accumulated by certain amacrine neurons with a high-affinity mechnnism[4,5], and it is also stored in a comparatively protected pool[6]. Moreover, it has recently been demonstrated to be releasable by light stimulation both in vivo and in vitro[7,8]. Glycine thus fulfills several of the main criteria of a transmitter substance.

Glycine is usually regarded as formed from serine by removing a hydroxyl group. It was thus of interest to see whether the neurons being labelled by ^{3}H-glycine would also be labelled by ^{3}H-serine. In a previous report, this was not the case[6], but there the tritium label was in the hydroxyl group of serine, and any glycine formed would not necessarily have been radioactive.

Rabbits were therefore injected intravitreally with 25 µCi L-serine-^{3}H (generally labelled), 4 or 24 h before processing the eyes for diffusion-free autoradiography as previously described[6]. The animals were kept in ambient

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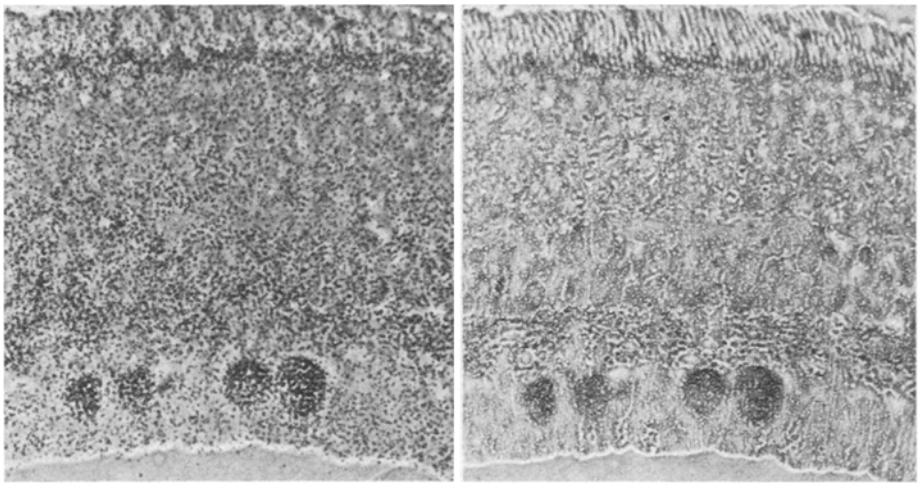

Autoradiograph of rabbit retina, 4 h after the intravitreal injection of 25 μCi ^{3}H-serine (generally labelled). There is radioactivity in 4 ganglion cells, in a layer near the external limiting membrane and for the rest rather diffusely throughout the retina. Left, focus on the grains; right, focus on the section. Phase contrast micrograph. ×450.

artificial laboratory illumination. Other rabbit retinas were incubated (15 min) as previously described[4] with the tritiated serine, 1 μCi/ml before being processed. Exposure times of the autoradiographs were 1 to 3 months.

No convincing selective labelling of retinal amacrine neurons was seen. The radioactivity had apparently accumulated (Figure) to a large extent in glial cells (Müller cells) and in the ganglion cells. Particularly after 24 h there was also a strongly radioactive zone in the photoreceptors. The pattern is very similar to what is seen with a number of other amino acids not suspected of being neurotransmitters or neurotransmitter precursors[6]. By waiting for 24 h after the intravitreal injection, it was hoped to reveal accumulation into the neuronal pool with slow turnover, but no such effect was evident.

Thus, it is not possible to tag the presumed glycinergic neurons with the precursor serine. This is in contrast with what is seen in, e.g., catecholaminergic neurons where dopa is readily converted to dopamine or noradrenaline. The reason for the failure with serine is not apparent, but one possibility is that the enzyme converting serine to glycine is being kept inactive in the cells under the conditions of the experiment resulting in no or little de novo synthesis of transmitter glycine. By comparison the rate limiting enzymes in the catecholamine synthesis are similarly subject to regulation in the neurons[9].

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Penetration of Melanocytes from Embryonic Japanese Quail Peritoneum into Associated Embryonic Avian Gonads, Grown on Chicken Chorioallantoic Membrane

M. Callebaut[1]
Laboratorium voor Anatomie en Embryologie, Rijksuniversitair Centrum, Groenenborgerlaan 171, B–2020 Antwerpen (Belgium), 4 June 1976.

Summary. After association on chorioallantoic membrane (CAM) of an embryonic bird testis with pigmented peritoneum from a Japanese quail embryo, numerous melanocytes penetrate in its interstitial tissue. If, instead of a testis, an ovary is transplanted under similar conditions, then the melanocytes may be found in the medulla or between the secondary sex cords at the rim of the ovary.

After a period of elective chimiotactic attraction for germ cells[2], the surface epithelial cells of the gonad penetrate together with these germ cells into the underlying stroma and form sex cords. In the present work, we have studied the behaviour of extragonadal pigmented coelomic epithelium (from quail embryos) when placed in contact with avian gonads, just after their sexual differentation.

Material and methods. The left gonad from 9- to 11-day-old Japanese quail embryos, or from 8-day-old chick embryos, was placed on the CAM of 7- to 9-day-old chick embryos according to the technique of Harris[3]. A sheet of pigmented parietal peritoneum from the infero-lateral part of the abdomen (either from female or male 10-day-old quail embryos) was placed over these gonads. Then, 6 to 9 days later, the transplants were excised and fixed in acetic-alcohol (1:3$_v$). After dehydration and embedding in paraffin, the transplants were sectioned at 7 μm thickness. For each transplant the sex of the host on which it developed was noted. The sections were stained with toluidine blue, PAS or Feulgen.

Results and discussion. In the 10-day-old Japanese quail embryo (wild type) pigmented coelomic cells can usually be seen under the stereomicroscope (X120). Their number increases in older embryos and after hatching. They are most numerous on the inferior and posterior walls of the peritoneal cavity.

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