The ability of the epithelium of diencephalic origin to differentiate into cells of the ocular lens

GORDANA JURIĆ-LEKIĆ[1]*, FLORIANA BULIĆ-JAKUŠ[2], BORIS KABLAR[1] and ANTON ŠVAJGER[1]

[1]*Institute of Histology and Embryology and* [2]*Institute of Biology, Faculty of Medicine, University of Zagreb, Republic of Croatia, Yugoslavia*

ABSTRACT After the discovery that in adult salamanders following lentectomy a new, functional lens develops by transdifferentiation (cell-type conversion) of previously depigmented epithelial cells of the iris (Wolffian lens regeneration), this phenomenon has been intensively studied by various experimental approaches. During the last two decades it was shown that pleiomorphic aggregates of atypical lens cells (lentoids) differentiated in reaggregates of dissociated cells of the chick neural retina and in spread cell cultures of the pigmented epithelium of the iris and retina, of the neural retina and the pineal gland of the chick embryo. The neural retina of human fetuses and adults also displayed this capacity. We showed that lentoids developed at a low incidence in renal isografts of rat embryonic shields or isolated embryonic ectoderm and of lentectomized eyes of rat fetuses, as well as in organ cultures of rat embryonic shields in chemically defined media. The addition of transferrin significantly increased the incidence of differentiation of lentoids in explants. In both renal isografts and explants *in vitro* a continuous transformation of retinal epithelial cells into atypical lens cells was observed. In renal isografts lentoids were also observed to originate from the ependyma of the brain ventricle. All tissues having the capacity to convert into lens cells belong to the diencephalon in a broad sense. Evolutionary aspects of this feature are discussed.

KEY WORDS: *lens, lentoids, diencephalon, transdifferentiation, rat*

Introduction

The lens is a derivative of the epidermis. It develops from the lens placode which invaginates into the concavity of the optic cup, which is an outpocketing of the diencephalon. This is a classical example of epithelial-epithelial inductive interaction and therefore a classical experimental model for investigation of the nature and mechanisms of embryonic induction. However, several experiments have shown that many contradictions still exist in the interpretation of lens induction (Lopashov and Stroeva, 1964; Hoperskaya, 1972; Saha *et al.*, 1989).

The most fascinating result of the investigation of lens development has been the observation that in various experimental conditions lens cells can differentiate not only from their natural site of origin (surface ectoderm or epidermis) but also from neuroectodermal cells of diencephalic origin which are constituents of the embryonic or even adult retina (iris epithelium, pigmented epithelium of the retina or some cells of the neural retina). Following the observations of a few forerunners, Wolff made in the first years of this century a detailed analysis of the process by which, after removal of the lens from larval or adult newts, a new and functional lens develops from the inner, previously depigmented epithelial cells of the iris. This phenomenon is commonly known as *Wolffian lens regeneration* (for historical details see Yamada, 1977).

This discovery attracted the attention of many investigators whose work has been extended to other species by using various experimental approaches (Reyer, 1954; Yamada, 1977; Eguchi, 1979, 1986; Yamada and McDevitt, 1984).

The attractiveness of this phenomenon is based upon the fact that it represents an unique example of the switching of a differentiated cell phenotype into another one (cell-type conversion). It is commonly designated as transdifferentiation (for terminology see Yamada, 1977). This essential problem in developmental biology is nowadays being intensively studied, predominantly by the Japanese group guided by T.S. Okada (selected reviews of general aspects: Okada, 1980, 1983, 1986; Kondoh *et al.*, 1986).

In the present communication we will refer to some particular aspects of the general problems of atypical differentiation of lens cells, summarize our own results obtained by ectopic transplantation of mammalian embryonic and fetal tissues and point to some evolutionary aspects of the ability of the epithelial cells of diencephalic origin to differentiate into lens cells.

***Address for reprints**: Institute of Histology and Embryology, University of Zagreb, Salata 3, P.O. Box 166, 41001 Zagreb, Republic of Croatia, Yugoslavia. FAX: 38-41-424-001

0214-6282/91/$03.00

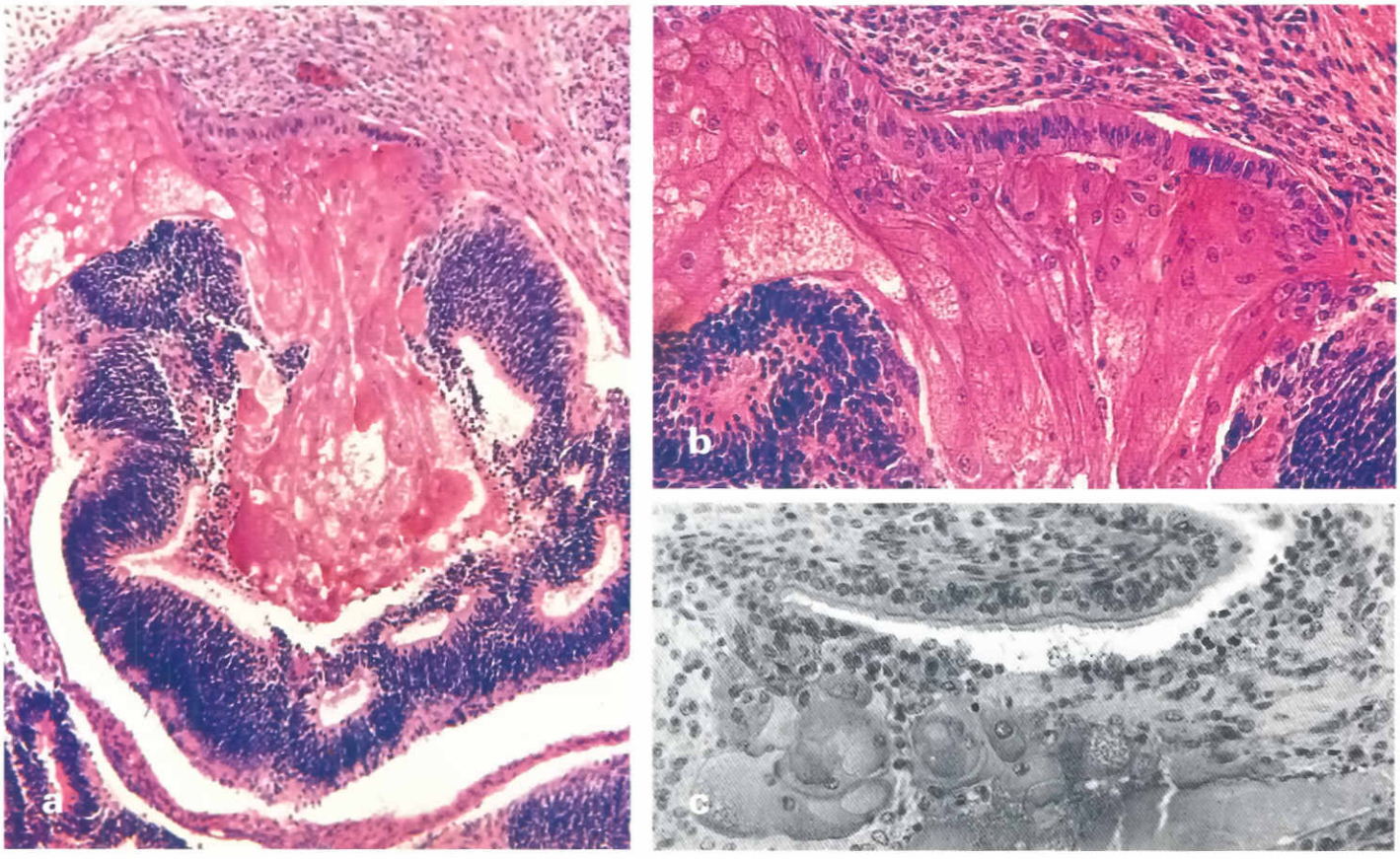

Fig.1. Lentoids in an experimental embryonic teratoma. (a) *A large mass of tissue of the neural retina resembling by its shape the optic cup. Note the massive invasion of lentoid cells from the retinal epithelium into the interior of the cup and partly over its left edge.* **(b)** *Detail of the top (a). The transitional zone between the retinal epithelium and the lentoid cells is in the left upper corner.* **(c)** *Part of a brain ventricle lined by ependymal cells from which lentoid cells arise (bottom). Hemalaun and eosin. (a) x50, (b) x120, (c) x450.*

General remarks

In order to investigate the potency of a cell population to differentiate into cells with characteristics of lens cells several approaches are possible. Recently the spreading cell culture seems to have become the method of choice. It has been recently combined with methods of immunohistochemistry, immunofluorescence, electrophoresis and molecular genetics. By using these methods, low levels of lens-specific protein crystallins were also detected in various embryonic tissues (extralenticular crystallins) such as adenohypophysis, central nervous system, limb bud etc., predominantly at early stages of differentiation (see reviews by Clayton *et al.*, 1986; Kondoh and Okada, 1986). However, in interpreting such results one has to consider the most recent discovery that crystallins also show various enzymatic activities so that they represent proteins with two entirely different functions, encoded by the same gene («gene sharing»). Therefore, the expression of a crystallin gene in a population of embryonic cells does not necessarily mean their ability to differentiate into lens cells (Piatigorsky and Wistow, 1989). The formation of groups of atypical lens cells (lentoids) in an experimental system, combined with other methods listed above, can therefore be considered as the only exact criterion for the lens-forming capacity of a cell population under appropriate environmental conditions.

Lentoids

Lentoids (lentoid bodies, lens-like bodies) are pleiomorphic aggregates of cells which bear tinctorial, ultrastructural and immunohistochemical characteristics of lens fibers. The size and shape of particular cells as well as of whole lentoids is variable. Cells are characterized by large, sometimes even giant size. Their shape can be round, elongated or flask-like. The cytoplasm is eosinophilic and diffusely fine granular without any other structural details visible by light microscopy. As a rule, the nuclei are euchromatic with prominent nucleolus. Ultrastructurally they resemble lens cells (fibers) within the normal lens *in situ* (Ophir *et al.*, 1985). Lentoids sometimes appear as single cells or as small aggregates of them, but more frequently as large aggregates of circular shape.

Lentoids show a positive immunofluorescent reaction when treated with anti-crystallin antibodies. In our experiments, however,

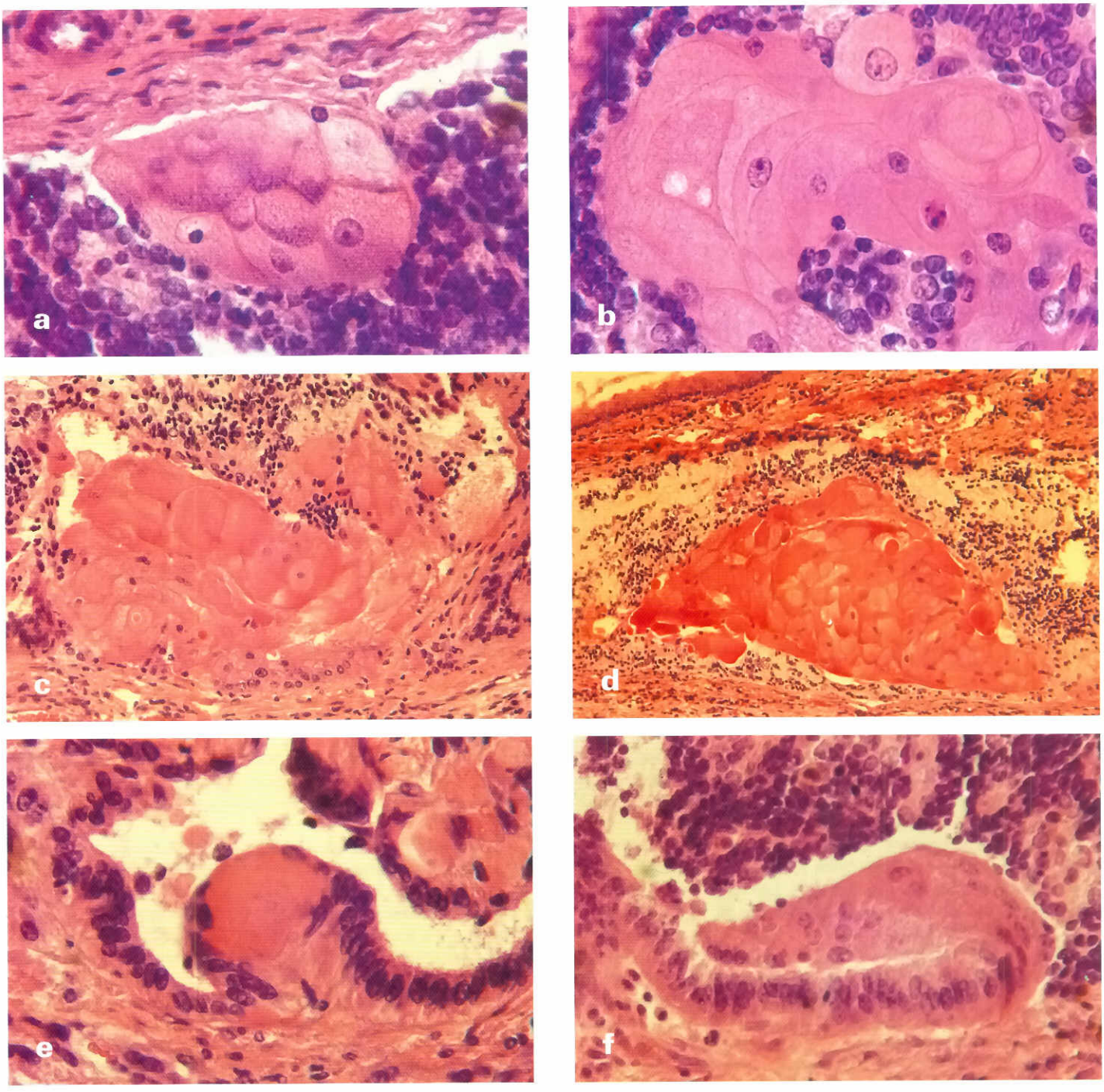

Fig. 2. Lentoids in grafts of lentectomized eyes. *Grafts were recovered after 5 days* **(f)**, *11 days* **(a,b)** *and 42 days* **(c,d,e)**. *Note that lentoids are in close spatial relationship with the neural retina. (e) Part of an irregularly outlined cyst lined by the retinal epithelium. A protrusion into the cavity contains a single large lentoid cell. (f9 The outset of the conversion of the retinal epithelium into lentoid cells with the clearly visible transitional zone (right). Hemalaun and eosin. (a,b,e) x320, (c,f) x200, (d) x150.*

they appeared at a low frequency, as accidental findings in some sections of serially sectioned large grafts. We therefore had no other choice than to rely on the distinctive histological features for their detection and characterization.

Lentoids in various experimental systems

There are many reports concerning differentiation of lentoids in grafts, organ and cell cultures of amphibian and chick embryonic tissues, predominantly of retinal or head ectodermal origin (see

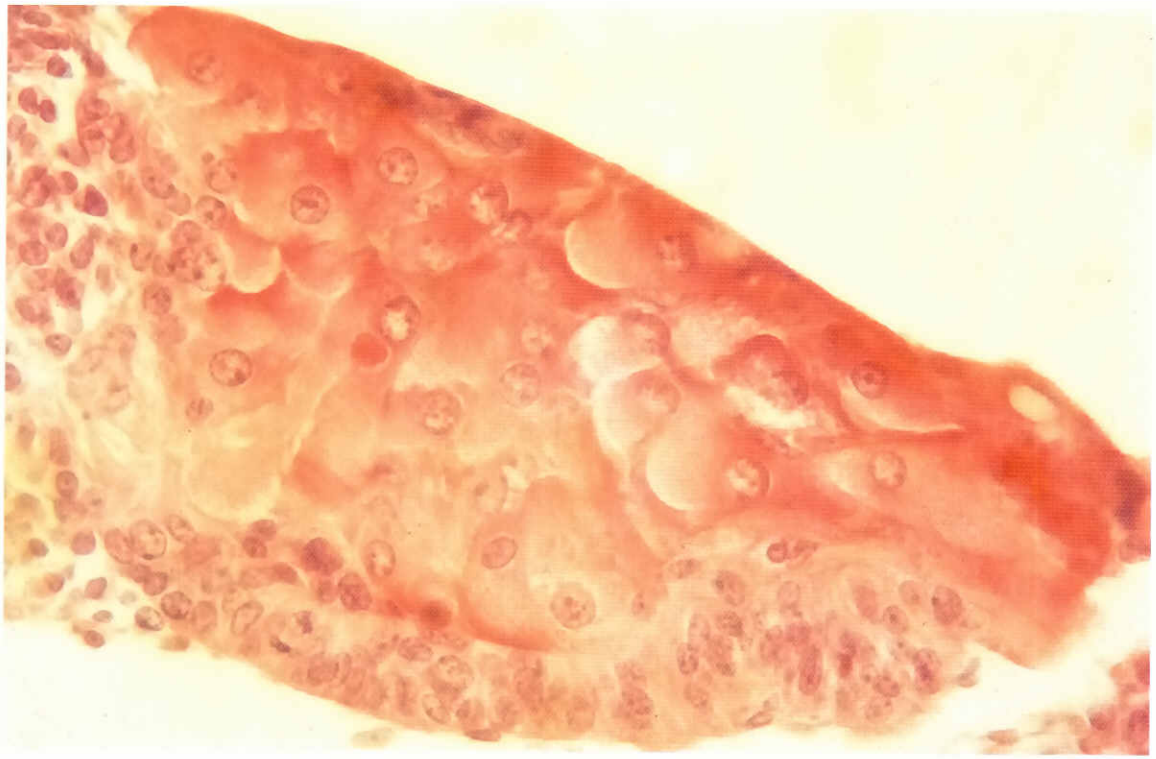

Fig. 3. Lentoid in an explant of the rat embryonic shield cultivated in Eagle's MEM supplemented with transferrin. *Note the retinal epithelium (bottom) with the transitional zone (right). Hemalaun and eosin. x150.*

Yamada, 1977; Eguchi, 1979 and Okada, 1980 for review). Some of them will be referred to in the following chapters of this review. Here we will mention only the ones most relevant to the main subject of our interest.

The essential problem of lentoid-progenitor cells within the neural retina seems to have been resolved by Moscona and his group. He first showed that lentoids appear in reaggregates of dissociated cells of the chick embryo neural retina (Moscona, 1957), In a series of further experiments it came out that the lens cells do not differentiate from neurons but from Müller glia cells (reviewed by Moscona, 1986). Another relevant finding was that lentoids can form in cultures of cells derived from the area comprising the chick embryo telencephalon and diencephalon with the rudiment of the pineal gland (Nomura, 1982).

In mammals, the ability of retinal cells to transform into lens cells was as yet confirmed only in human fetuses. Lentoids developed in cell cultures of iris and retinal pigment epithelial cells of 12-week fetuses (Yasuda *et al.*, 1978), of neural retina cell from 9-and 15-week fetuses (Okada *et al.*, 1977) and of retinal pigment epithelial cells from an 80 year-old person (Eguchi, 1988). Here we show that in some experimental conditions lentoids can arise from cells of diencephalic origin in rodents as well.

Lentoids in experimental embryonic teratomas

Experimental embryonic teratomas are tumors which develop from early mammalian embryos transplanted to an ectopic (extrauterine) site (usually under the capsule of the testis or the kidney). They consist of a multitude of mature tissues in a chaotic arrangement (for details see Levak-Svajger *et al.*, 1991 in this issue).

We examined teratomas derived from whole *pre-primitive* streak and early primitive streak rat embryos and of the primitive embryonic ectoderm isolated from them, transplanted under the kidney capsule of syngeneic adult male rats for 15-30 days (Svajger *et al.*, 1987). Lentoids were found in 11 out of 207 tumors (5.3%). In some of them a direct relationship to the immediately neighboring structures could be observed: a) to the neural retina. Lentoids are regularly, at least partially, surrounded by the immature tissue of the neural retina, and, in most typical cases they are in direct cellular continuity with the retinal epithelium. In these cases all transitional forms exist between the cuboidal epithelium and the large, atypical lens cells (Fig. 1a,b); b) to ependyma of the brain ventricle. A brain ventricle lined by a layer of cuboidal ependymal cells, into which a well-developed choroid plexus protrudes, is often found in embryonic teratomas. In one teratoma lentoids were found whose cells were continuous with the ependymal layer and the whole lentoid protruded into the surrounding brain tissue without any spatial relationship to the neural retina (Fig. 1c).

No surface ectodermal structures (skin) have ever been observed in the proximity of lentoids. This, as well as the cellular continuity, strongly suggest the origin of lentoids from the epithelium of diencephalic origin. Lentoids were also found in teratomas derived

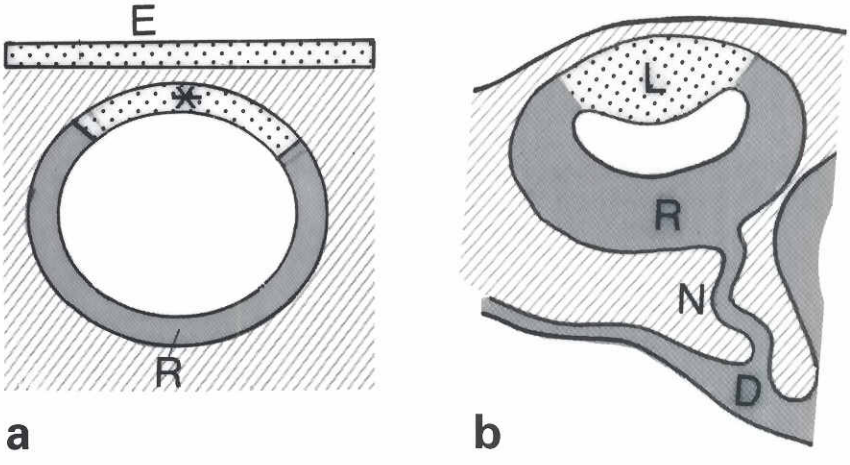

Fig. 4. The vesicular eye (a) and the pineal organ of a lizard embryo (b). *(D) roof of the diencephalon, (E) epidermis, (L) lens, (N) parietal nerve, (R) retina, (*) undifferentiated part of the optic vesicle.*

from mouse embryos (Diwan and Stevens, 1976; Bennett *et al.*, 1977). We found a few lentoid cells within the sacrococcygeal teratoma of a newborn (unpublished).

Lentoids in grafts of lentectomized eyes

In order to test the ability of the rat fetal iris epithelium to differentiate into lens cells, the eyeballs of 14- and 18-day rat fetuses were enucleated, lentectomized and transplanted under the kidney capsule of adult syngeneic rats for a period of 5-66 days (Juric-Lekic and Svajger, 1989). The histological examination of grafts showed that their internal structure (topography of tissues) was disturbed to a great extent, thus giving the tumors a teratoma-like appearance. The neural retina and the epithelium of the retina were present in all grafts. Lentoids were absent in grafts of 14-day fetuses (23 grafts) and only 4 lentoids developed in grafts of 18-day fetuses (41 grafts). The distinctive features of these lentoids were: a)they did not need a long period of time to appear (one of them was found only 5 days after cultivation); b) they were always found in close proximity to the neural retina (Fig. 2a-f); c) in favorable sections one could observe, as in experimental teratomas, a cellular continuity with transitional forms between the epithelium of the retina and the lentoid cells (Fig. 2f).

Teratomas obtained from renal isografts of the 15- and 16-day mouse fetuses (experimental period 5-33 days) continued the neural retina and the pigmented retinal epithelium but lentoids did not develop (Juric-Lekic, 1991).

These observations lead to the conclusion that epithelium of the retina of the rodent fetus has the capacity to differentiate into lens cells, but due to a presumable multitude and complexity of yet unknown intrinsic environmental factors the mechanism of cell-type conversion works with a low efficiency.

Lentoids in organ culture of rat embryonic shields

When rat embryonic shields (egg cylinders) are grown in a modified organ culture (Skreb and Svajger, 1973) in various serum-supplemented or serum-free media, lentoids sometimes develop in explants. In a recent experiment rat embryonic shields at the primitive streak stage were cultivated for 2 weeks in a chemically defined medium to determine the influence of human transferrin on the terminal tissue differentiation (Bulic-Jakus *et al.*, 1990). The essential results relevant to the present problem were the following: a)lentoid cells were continuous with a cuboidal epithelium characterized by pale cytoplasm, reminiscent of the epithelium of the retina found in teratomas; b)in media which contained transferrin, the incidence of lentoid formation was strikingly higher than in Eagle's MEM supplemented with rat serum or in MEM alone (33%, 2% and 1% respectively, Fig.3).

The incidence of lentoid formation from rat embryonic shields cultivated *in vitro* in the presence of transferrin in the medium is 6 times higher than in teratomas derived from embryos and ectoderms transplanted under the kidney capsule at the same developmental stage (Svajger *et al.*, 1987). However, the true nature of the triggering mechanism of transferrin remains obscure.

Lentoids in cultures of pinealocytes

It was recently shown that in cultures of dissociated cells of quail embryo pineal glands differentiation of pigment cells and crystallin-containing lentoids occurred («oculopotency» of embryonic avian pineals; Watanabe *et al.*, 1985).

In higher vertebrates the pineal gland is not a photoreceptive organ and it is commonly considered as a vestige of the median (parietal or third) eye of lower vertebrates (Gladstone and Wakeley, 1940; Duke-Elder, 1958; Eakin, 1970). This is a dorsal outpocketing of the roof of the diencephalon, the third brain vesicle which through a hole in the skull approaches a translucid area of the skin and expands into a vesicle. This contains photoreceptors, except in the distal area which represents the lens and contains crystallins (McDevitt, 1972). It is developed in primitive fishes (Cyclostomes) but photoreceptors showing the same ultrastructure as the visual cone cells of the eye retina can also be found within the pineal gland of teleosteans such as the trout, carp, and mackerel (Hibiya, 1982). A parietal eye also exists in primitive reptiles such as Sphenodon and Chameleon (Duke-Elder, 1958) and in some others as a transient embryonic structure (Fig. 4).

In a preliminary experiment we isolated rudiments of the pineal gland with the immediately surrounding brain tissue of 12-, 13- and

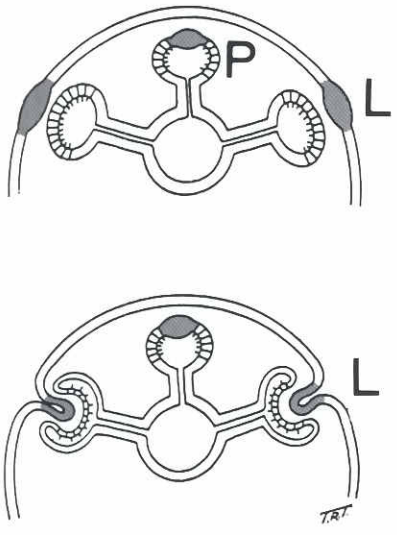

Fig. 5. Origin of the lens in the parietal eye. *Note that the lens of the lateral eyes (L) forms by invagination of the thickened surface ectoderm (lens placode) while the lens of the parietal eye (P) is a part of the vesicular eye which originates from the diencephalon. Modified from Duke-Elder (1958).*

15-day rat embryos and grafted it under the kidney capsule for 30 days. Only brain tissue and no other diencephalic structures developed in grafts.

The problem of causal factors

Such a unique and unexpected phenomenon as is lentoidogenesis from epithelial cells of diencephalic origin rather than from its natural source (surface ectoderm), requires a causal explanation, *i.e.* revealing the mechanism which triggers the cell-type conversion. There have been numberless attempts to attack this problem (for information see Zalokar, 1944; Yamada, 1977; Eguchi, 1979; Okada, 1980; Moscona, 1986). It is beyond the scope of this review to discuss this intricate matter full of controversies. We may only mention some factors which have been considered to influence the atypical lentoidogenesis either by promoting or by inhibiting it: species-species or genetic differences, origin of the epithelium from different segments of the iris, traumatism of the tissue or disruption of cell-to-cell contacts, the presence of retina, lens, other tissues and substances, developmental stage, culture conditions etc. The enhancement of lentoid formation *in vitro* by transferrin (Bulic-Jakus *et al.*, 1990) is a promising finding but it is still hardly possible to interpret it.

Evolutionary aspects

The evolution of the eye is very complex, discontinuous and characterized by many variations in structural pattern and origin during development. There is however, with very few exceptions, one general rule: photosensitive elements always have an adjoining structure of various origin and composition, which is comparable or analogous to the ocular lens. (All data referred to in this section are quoted from Duke-Elder, 1958.)

Already in some unicellular organisms (Flagellates) the photosensitive, pigment-containing organelle, the eye-spot or stigma is attached to a refractile structure which has been considered as the primitive lens. In evertebrates the entire eye originates from the surface ectoderm and photosensitive cells are subsequently connected with the nervous system. Even in some very primitive evertebrates, in which the eye exists only as a pit in the epidermis, as well as in compound eyes of insects a refractile lens-like structure covers the photosensitive cells. It may be cellular or acellular and originate from the cuticle, the hypodermal cells or from a gelatinous secretion of epidermal, hypodermal or retinal cells.

The next step in the evolution of the eye is the *vesicular eye,* characteristic particularly for gastropods. It develops by a complete pinching-off of the photosensitive area of the epidermis into the underlying mesenchyme. The major proximal part of the vesicle contains photoreceptors and represents the retina, while the distal part, underneath the epidermis, consists of relatively undifferentiated cells (Fig. 4a).

This part is homologous to the lens of the parietal eye in some fishes and reptiles (Fig. 4b).

In vertebrates a sudden change occurs: the compound eye with the inserted retina arises and a cellular lens develops by invagination of the surface ectoderm. The only exception is the parietal eye (an «evolutionary by-way») which has the structure of the primitive vesicular eye from which it differs by the origin from the diencephalon instead from the epidermis and by the differentiation of its distal part into a functional lens (Fig. 5).

To sum up, the lens has a long evolutionary history, during which it has had various origins and compositions. It shares with the neural retina the surface ectodermal evolutionary origin which other sensory organs (olfactory, taste, auditory, vestibular) have conserved during evolution. The retina, however, became an integral part of the central nervous system (diencephalon) but it still has retained some dormant developmental capacities reminiscent of its ancestral epidermal origin. The same seems to hold true for the ependyma of the diencephalon and for the pineal gland. The expression of this «phylogenic memory» is the capacity of tissues of diencephalic origin to differentiate into the lens, which is a typical epidermal tissue. The capacity to form integumental structures was also observed in the human parotid gland (which is a derivative of the surface ectoderm) in which a metaplasia of the duct epithelium into sebaceous glands can occur (Meza-Chàvez, 1949).

Concluding remarks

The data presented in this review open many questions which are difficult or even impossible to answer. It seems obvious that the capacity of some tissues to differentiate into lens cells depends on both the imprinting in the genome and the extrinsic factors created by the experimental approach. It is especially difficult to disclose what is actually going on during tissue differentiation in experimental embryonic teratomas. We have no evidence about the state of commitment of the neuroectodermal cells at the time of their engagement in lens cell formation and we are therefore not able to decide whether we are dealing with a true transdifferentiation or with a transdetermination. Other unanswered questions are the low incidence of lentoids in renal explants and the apparent species-

dependence of the lens-forming capacity (absence of lentoids in teratomas derived from mouse embryos and in those derived from rudiments of the pineal glands of rat embryos). Inductive mechanisms which bring about the differentiation of lens cells from atypical progenitor cells are completely unknown.

What seems to be certain is the following: a) the retinal epithelium and the ependyma of the diencephalon of the rat embryo have the capacity to differentiate into lens cells and b) in all experimental systems in which lentoids develop, their progenitor cells originate from the ependymal wall of the diencephalon. We can hope that the molecular genetic approach will shed more light on the essential problem of causal factors involved in this intricate developmental process.

Acknowledgments

The authors gratefully acknowledge the technical assistance of Durdica Cesar and Radmila Delas. The original work was supported by grants of the Scientific fund of the Republic of Croatia, through funds made available to the USA-Yugoslav Joint Board on Scientific and Technological Cooperation (No. 02-094-N) and by the Small Supplies Program of the WHO, Geneva.

References

BENNETT, D., ARTZT, K., MAGNUSON, T. and SPIEGELMAN, M. (1977). Developmental interactions studied with experimental teratomas derived from mutants of the T/t locus in the mouse. In *Cell Interactions in Differentiation* (Eds. M. Karkinen-Jääskeläinen, L. Saxén and L. Weiss). Academic Press, New York, pp. 389-398.

BULIC-JAKUS, F., SKREB, N., JURIC-LEKIC, G. and SVAJGER A. (1990). Transferrin enhances lentoid differentiation in rat egg cylinders cultivated in a chemically defined medium. *Int. J. Dev. Biol. 34:* 275-279.

CLAYTON, R.M., JEANNY, J-C., BOWER, D.J. and ERRINGTON, L.H. (1986). The presence of extralenticular crystallins and its relationship with transdifferentiation to lens. *Curr. Top. Dev. Biol. 20:* 137-151.

DIWAN, S.B. and STEVENS, L.C. (1976). Development of teratomas from the ectoderm of mouse egg cylinders. *J. Natl. Cancer Inst. 57:* 937-939.

DUKE-ELDER, Sir S. (1958). The eye in evolution. In *System of Ophthalmology* (Ed. Sir S. Duke-Elder). Henry Kimpton, London, Vol.I, pp. 125-177, 233-248, 711-720.

EAKIN, R.M. (1970). A third eye. *Am. Sci. 58:* 73-79.

EGUCHI, G. (1979). «Transdifferentiation» in pigmented epithelial cells of vertebrate eyes *in vitro*. In *Mechanisms of Cell Change* (Eds. J.D. Ebert and T.S. Okada). John Wiley & Sons, New York, pp. 273-291.

EGUCHI, G. (1986). Instability in cell commitment of vertebrate pigmente epithelial cells and their transdifferentiation into lens cells. *Curr. Top. Dev. Biol. 20:* 21-37.

EGUCHI, G. (1988). Cellular and molecular background of Wolffian lens regeneration. *Cell. Differ. Dev. 25 (Suppl.):* 147-158.

GLADSTONE, R.J. and WAKELEY, C.P.G. (1940). *The Pineal Organ*. Bailliere, Findal and Cox, London, pp. 240-258.

HIBIYA, T. (1982). *An Atlas of Fish Histology*. Gustav Fischer-Verlag, Stuttgart, pp. 116-119.

HOPERSKAYA, O.A. (1972). Differentiation of lens- and iris-like tissue in explants of the anterior part of the frog neural plate. *W. Roux Arch. Entw. Mech. Org. 171:* 1-16.

JURIC-LEKIC, G. (1991). Retinal epithelium in the enucleated and lentectomized eye of the mouse fetus grafted under the kidney capsule. *Rad. Med. Fak. Zagrebu 32:*73-78.

JURIC-LEKIC, G. and SVAJGER, A. (1989). Lentoid formation in ectopic grafts of lentectomized eyes of rat fetuses. *Cell. Differ. Dev. 27:* 225-232.

KONDOH, H., HAYASHI, S., TAKAHASHI,Y. and OKADA, T.S. (1986). Regulation of δ-crystallin expression: an investigation by transfer of the chicken gene into mouse cells. *Cell. Differ. 19:* 151-160.

KONDOH, H. and OKADA, T.S. (1986). Dual regulation of expression of exogenous δ-crystallin gene in mammalian cells: a search for molecular background of instability in differentiation. *Curr. Top. Dev. Biol. 20:* 153-164.

LEVAK-SVAJGER, B., KNEZEVIC, V. and SVAJGER, A. (1991). Development of separated germ layers of rodent embryos on ectopic sites: a reappraisal. *Int. J. Dev. Biol. 35* (In this issue).

LOPASHOV, G.V. and STROEVA, O.G. (1964). Development of the Eye. Experimental Studies. Israeli Program for Scientific Translations. Jerusalem.

MCDEVITT, D.S. (1972). Presence of lateral eye lens crystallins in the median eye of the American chameleon. *Science 175:* 763-764.

MEZA-CHÀVEZ, L. (1949). Sebaceous glands in normal and neoplastic parotid glands. *Am. J. Pathol. 25:* 627-645.

MOSCONA, A.A. (1957). Formation of lentoids by dissociated retinal cells of the chick embryo. *Science. 125:* 598-599.

MOSCONA, A.A. (1986). Conversion of retina glia cells into lenslike phenotype following disruption of normal cell contacts. *Curr. Top. Dev. Biol. 20:* 1-19.

NOMURA, K. (1982). Differentiation of lens and pigment cells in cultures of brain cells of chick embryos. *Differentiation 22:* 179- 184.

OKADA, T.S. (1980). Cellular metaplasia or transdifferentiation as a model for retinal cell differentiation. *Curr. Top. Dev. Biol. 16:* 349-380.

OKADA, T.S. (1983). Recent progress in studies of the transdifferentiation of eye tissue *in vitro*. *Cell. Differ. 13:* 177-183.

OKADA, T.S. (1986). Transdifferentiation in animal cells: fact or artifact? *Dev. Growth Differ. 28:* 213-221.

OKADA, T.S., YASUDA, K., HAYASHI, M., HAMADA, Y. and EGUCHI, G.(1977). Lens differentiation in cultures of neural retinal cells of human fetuses. *Dev. Biol. 60:* 305-309.

OPHIR, I., MOSCONA, A.A., LOYA, N. and BEN-SHAUL, Y. (1985). Formation of lentoids from retina gliocytes: ultrastructural study. *Cell. Differ. 17:* 149-157.

PIATIGORSKY, J. and WISTOW, G. Y. (1989). Enzyme/crystallins. gene sharing as an evolutionary strategy. *Cell 57:* 197-199.

REYER, R.W. (1954). Regeneration of the lens in the amphibian eye. *Q. Rev. Biol. 29:* 1-46.

SAHA, M.L., SPANN, C.L. and GRAINGER, R.M. (1989). Embryonic lens induction: more than meets the optic vesicle. *Cell. Differ. Dev. 28:* 153-172.

SKREB, N. and SVAJGER, A. (1973). Histogenetic capacity of rat and mouse embryonic shields cultivated *in vitro*. *W. Roux Arch. Entw. Mech. Org. 173:* 228-234.

SVAJGER, A., LEVAK-SVAJGER, B. and JURIC-LEKIC, G. (1987). Origin of lentoids in experimental teratomas derived from early postimplantation rat embryos. *Roux Arch. Dev. Biol. 196:* 385-387.

WATANABE, K., AOYAMA, H., TAMAMAKI, N., YASUIMA, M., NOJYO, Y., UEDA, U. and OKADA, T.S. (1985). Oculopotency of embryonic quail pineals as revealed by cell culture studies. *Cell. Differ. 16:* 251-257.

YAMADA, T. (1977). *Control Mechanisms in Cell-Type Conversion in Newt Lens Regeneration* (Ed. A. Wolsky). Monograph in Developmental Biology. Vol. 13. S. Karger, Basel, pp. 92-97.

YAMADA, T. and MCDEVITT, D. (1984). Conversion of iris epithelial cells as a model of differentiation control. *Differentiation 27:* 1-12.

YASUDA, K., OKADA, T.S., EGUCHI, G. and HAYASHI, M. (1978). A demonstration of a switch of cell type in human fetal eye tissues *in vitro*: pigmented cells of the iris or the retina can transdifferentiate into lens. *Exp. Eye Res. 26:* 591-595.

ZALOKAR, M. (1944). Contribution à l'étude de la régénération du cristallin chez le Triton. *Rev. Suisse Zool. 51:* 443-521.