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Comparative stereological study on zonation and cellular composition of adrenal glands of normal and anencephalic human fetuses. I. Zonation of the gland

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Summary. Comparative stereological studies were performed on zonation of adrenal glands in 10 anencephalic and 11 normal fetuses, aged between 24 and 39 postovulatory weeks.

The development of adrenal fetal zone (FZ) is the main factor responsible for normal adrenal gland growth during the intra-uterine life. At the end of fetal period volume of this zone attains over 8200 mm^3 and constitutes 69-70% of the total gland volume, while respective values for zona glomerulosa (ZG), zona fasciculata (ZF) and medulla (M) are 1665 mm^3 (14%), 833 mm^3 (7%) and 1071 mm^3 (9%). These data were in striking contrast with those found in adrenals of anencephalic fetuses, in which volume of the gland attained only ca 35% of that in normal fetuses. In the oldest anencephalic fetus studied (39 weeks) the volume of the fetal zone was 962 mm^3 (ca 8-folds lower than in normal fetus) comprising only 25% of the total gland volume. For the remaining zones the values were: ZG - 1501 mm^3 (ca 90% of the normal value and 39% of the total gland volume); ZF - 770 mm^3 (ca 92% and 20% respectively), and M - 539 mm^3 (ca 50% and 14%, respectively).

This stereological study provides the first systemic description of the development of adrenal gland of anencephalic fetuses in comparison with the normally developing gland. Moreover, evidence is given that the growth of adrenal medulla is also retarded in anencephaly while the growth of the zona glomerulosa remains rather unaffected.

Key words: Human fetal development, Adrenal gland, Zonation, Anencephaly

Introduction

Anencephaly, a neural tube defect incompatible with postnatal survival, according to Müller and O'Rahilly (1984, 1991) occurs in three phases: 1) cerebral dysraphia, which is the failure in closure of a neural tube, 2) exencephaly as an exposure of the open brain outside the skull, 3) anencephaly, resulting in exposure of the brain for a prolonged period to amniotic fluid and degeneration throughout the fetal period. In anencephaly the parts lacking are prosencephalon, the mesencephalon, and the anterior part of the rhombencephalon. Neural tissues are present in varying degree and small foci of histologically normal cerebral cortex or olfactory tract may persist.

In anencephaly the adenohypophysis may be deficient or may become detached from the brain. The anterior lobe is usually present and it may contain normal cells (Nakano, 1973). The pars intermedia and pars nervosa are often absent or markedly smaller (Covel, 1927; Angevine, 1938)

In this malformation the development of adrenal glands is of particular interest. It is well known that the adrenal gland in anencephalic fetuses in smaller than that in normal fetuses, and that fetal adrenals grow relatively normally during the first half of fetal period (Keene and Hewer, 1927; Bachman, 1954; Bernischke, 1956; Jost, 1966; Sucheston and Cannon, 1970; Dhom, 1981). However, Gray and Abramovich (1980) using histometric technique have revealed that between 11 and 21 weeks of gestation the volume of adrenal gland occupied by the fetal zone in anencephalic fetuses was 57.5%, in comparison to 76.5% in normal fetuses.

Scarcity of morphometric studies of adrenal gland in anencephalic fetuses encouraged us to perform the present investigations. The aim of the present study was to describe the development of adrenal zonation in anencephalic fetuses and to compare it with the development of adrenal zonation in normal human fetuses of the same age.

Materials and methods

Stereological studies were performed on 10 anencephalic fetuses collected from 1985 to 1995 in the Department of Clinical Pathomorphology, University School of Medical Sciences in Poznan. A few macerated

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Table 1. Crown-rump length (C.-R.L.), body and adrenal weight, age, sex and gross malformations of anencephalic fetuses.

No.	C.-R.L (mm)	BODY WEIGHT (g)	ADRENAL WEIGHT (mg)	AGE IN POST-OVULATORY WEEKS	SEX	OTHER GROSS MALFORMATIONS
1	228	800	1,800	24	F	Meningocele, haernia umbilicalis, palatoschisis, malformationes digitorum manus et pedis
2	231	960	2,100	25	M	Meningomyocele regionis cervicalis et thoracica
3	233	790	1,900	27	M	-
4	248	1,000	2,120	28	F	Rachischisis completa, malformation cordis (two vessels)
5	262	1,040	2,600	32	F	Hepatosplenomegalia
6	280	1,850	2,800	34	F	Gibbus columnae vertebralis regionis thoracalis, amyelia, aplasia columnae vertebralis regionis cervicalis, meningomyocele regionis cervicalis, atelectasis pulmonum completa
7	305	1,680	3,600	37	F	
8	318	2,100	4,000	39	F	Fibrosis nodularis endocardii
9*	290	2,120	5,010	34	F	Amyelia regionalis cervicalis, aplasia columnae vertebralis regiones cervicalis
10**	320	2,990	9,970	38	F	

*: anencephalic with hypophysis; **: anencephalic with hypophysis and hypothalamus.

Table 2. Crown-rump length (C.-R.L.), body and adrenal weight, sex and age of normal fetuses.

No	C.-R.L (mm)	BODY WEIGHT (mg)	ADRENAL WEIGHT (mg)	AGE IN POSTOVULATORY WEEKS	SEX
1	230	850	3,737	24	F
2	231	810	3,602	25	F
3	244	910	4,850	26	M
4	266	1,090	5,840	29	M
5	284	1,500	7,030	31	F
6	291	2,320	7,890	32	F
7	298	2,600	8.020	33	F
8	305	2,850	8,120	35	M
9	312	3,100	9,800	36	F
10	316	3,210	12,100	38	M
11	328	3,550	12,360	39	F

specimens were excluded from the study. In 2 cases only right adrenal gland was found. The sex, crown-rump length (C.-R.L), body weight, and gross malformations of each fetus are shown in Table 1. According to autopsy protocols of anencephalic fetuses in one of them the hypothalamus and hypophysis was found, in one fetus only hypophysis (both lobes) was present, and in the remaining fetuses rudimentary adenohypophysis was observed or the gland was lacking.

Comparative stereological studies were performed on 11 pairs of adrenal glands of human fetuses obtained from the collection of the Department of Anatomy. The collection consists of normal fetuses from spontaneous abortions (Table 2). The permission of the Ethic Committee was obtained.

The age of fetuses (in postovulatory weeks) was calculated on the basis of body weight, C.-R.L., and foot length.

Adrenals were carefully excised together with the connective tissue capsule and weighed to the nearest 10 mg for anencephalic glands or 1 mg for normal fetuses. After fixation in Bouin's solution, and embedding in paraplast, glands were sectioned at 5-6 μm. Transverse sections of the middle part of the gland were stained with haematoxylin and eosin.

Stereological studies were performed according to Weibel (1979). Using a magnification of about x100 and a square lattice test system of type A (Weibel, 1979), the volume densities of the adrenocortical zones (zona glomerulosa - ZG; zona fasciculata - ZF; fetal zone - FZ), medulla (M) and capsule were evaluated. The measurements were made on 5-10 paraplast sections of the gland. The volume of the adrenal gland was calculated from its weight, by assuming that the average specific gravity of the gland was 1.039 mg/mm^3 (Swinyard, 1938). Results were expressed per adrenal pair (where applicable).

The results were processed statistically and graphically by Origin programme for IBM PC.

Results

In both normal and anencephalic fetuses, a linear increase in the adrenal gland weight and volume took place; however, the rate of increase was markedly lower in the latter (Fig. 1). The adrenal volume of anencephalic fetus possessing well developed hypophysis reached intermediate value, while in anencephalic specimen with hypothalamus and hypophysis volume of the gland did not differ from normal fetal glands.

Stereological analysis of the developing adrenal gland of all investigated fetuses revealed the linear increase in the volume of the ZG (Fig. 2). Of interest was the lack of statistically significant difference between the rate of ZG growth in normal and anencephalic fetuses. On the contrary, ZF volume in normal fetuses exhibited an exponential increase, while in anencephalic fetuses it remained rather constant (Fig. 3). The high rate of increase in adrenal volume in normal fetuses depended mainly on enlargement of the fetal zone (Fig. 4). On the contrary, in anencephalic fetuses volume of the FZ increased slightly. Exponential increase in the volume of adrenal medulla of normal fetuses contrasted with a slow linear growth of that zone in anencephalic fetuses (Fig. 5). As evidenced by Figs. 4, 5, in normal fetuses FZ and M enlarged rapidly after 31st week of intra-uterine life.

In anencephalic fetus with hypothalamus and hypophysis (C.-R.L. 320 mm, week 38 of intra-uterine

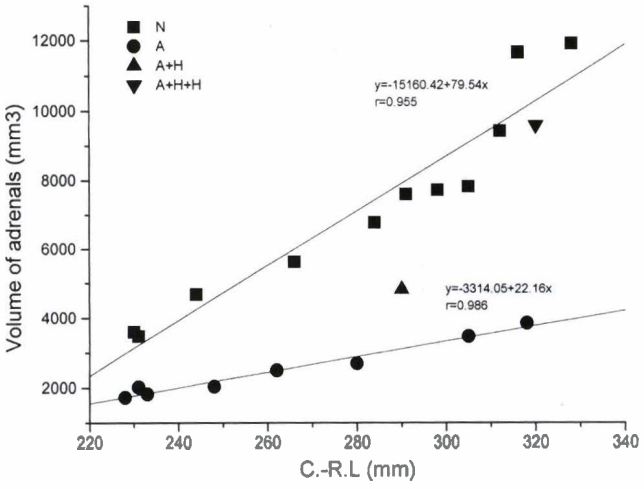

Fig. 1. Adrenal gland volume of normal and anencephalic human fetuses as plotted against the crown-rump length (C.-R.L). Each point presents one case.

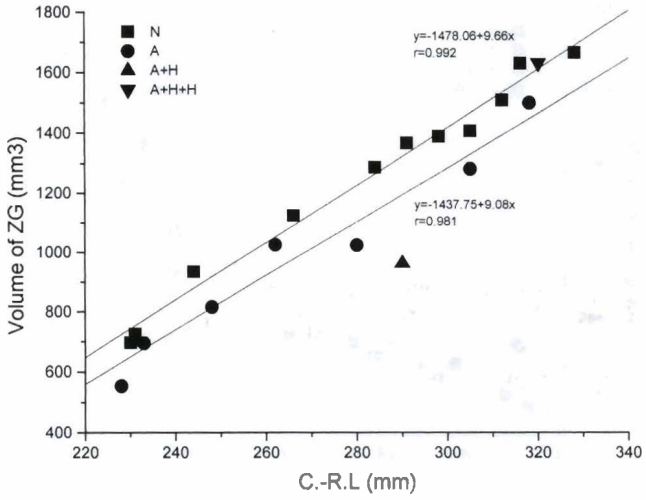

Fig. 2. Zona glomerulosa (ZG) volume of normal and anencephalic human fetuses as plotted against the crown-rump length (C.-R.L.). Each point presents one case.

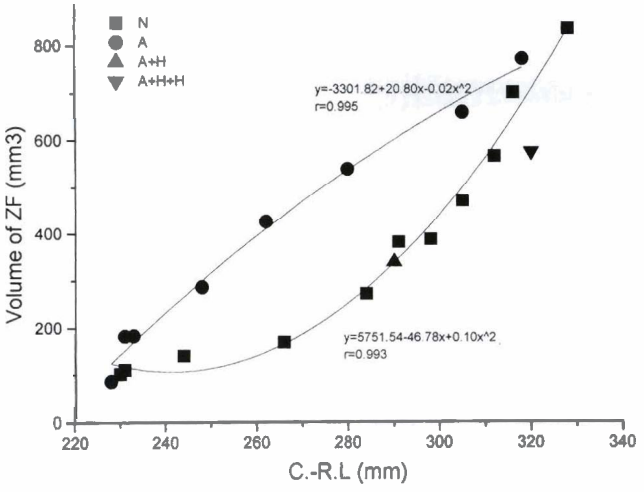

Fig. 3. Zona fasciculata (ZF) volume of normal and anencephalic human fetuses as plotted against the crown-rump length (C.-R.L.). Each point presents one case.

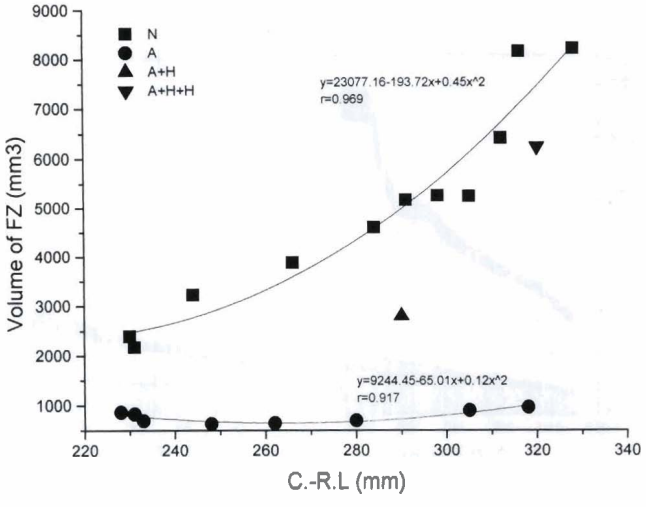

Fig. 4. Fetal zone (FZ) volume of normal and anencephalic human fetuses as plotted against the crown-rump length (C.-R.L.). Each point presents one case.

life) the growth of the entire adrenal gland and its zones was similar to that observed in normal fetuses. In case of younger fetus (C.-R.L. 290 mm, week 34 of intra-uterine life) in which pituitary gland but not hypothalamus was present, the growth of the adrenal gland was retarded in comparison with normal fetuses but higher than in anencephalic fetuses without adenohypophysis.

Analysis of the growth of glands in normal fetuses showed that the volume of the ZG increased from 725 to 1665 mm^3, constituting from 20.9 to 14.0% of the total volume of adrenals, respectively (Figs. 6, 7). In anencephalic fetuses the volume of the ZG increased from 554 to 1501 mm^3 while fraction of the gland occupied by this zone increased from 32 to 39%. During the studied period of intra-uterine life the volume of the ZF in normal fetuses increased from 111 to 883 mm^3 while in anencephalic fetuses from 87 to 770 mm^3, comprising 3.2-7.0% and 5.0-20.1% of the total gland volume, respectively. The high rate of increase in adrenal volume in normal fetuses depended mainly on enlargement of FZ, from 2170 to 8208 mm^3 and in the youngest fetus studied the fetal zone comprised about 63% of the total gland volume, while in the oldest one the respective value was 69%. On the contrary, in anencephalic fetuses volume of the fetal zone increased slightly (866 to 962 mm^3), constituting from 50 to 25% of the total gland volume. The volume of adrenal medulla in normal fetuses increased from 329 to 1071 mm^3 (9.5-9.0% of the gland volume), while in adrenals of anencephalic fetuses respective values were 173-539 mm^3 and 10-14%.

General structure of adrenal gland of normal and anencephalic fetuses at 37-38 weeks of development is shown in Figs. 8, 9.

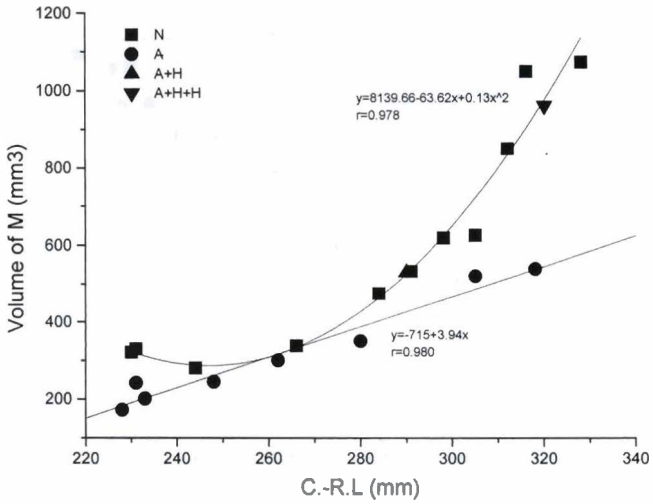

Fig. 5. Medulla (M) volume of normal and anencephalic human fetuses as plotted against the crown-rump length (C.-R.L.). Each point presents one case.

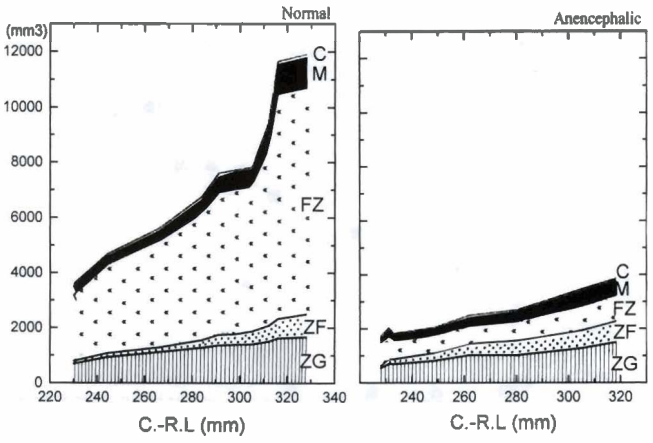

Fig. 6. Volume of adrenal zones and capsule (mm^3) of normal and anencephalic human fetuses from 228 to 328 mm C.-R.L. (24-39 postovulatory weeks). ZG: zona glomerulosa; ZF: zona fasciculata; FZ: fetal zone; M: medulla; C: capsule.

Discussion

Most aspects of anencephaly, which has been produced experimentally (Giround and Martinet, 1960), are still not well understood and its etiology is considered to be usually polygenic or multifactorial. Mechanical factors, such as fetal adhesion to the placenta or amniotic bands, also chromosomal abnormalities seem to be important (Hurwitz, 1955; Branchaud and Murphy, 1992). Apart from this, geographic factors seem to be significant because for example in Ireland and Scotland incidence is 2 to 4 per 1000 births (Elwood, 1970), which is 10 times higher than the overall incidence. Valuable information relating to anencephaly are available in the interesting paper by Müller and O'Rahilly (1991).

Literature data also indicate the higher frequency of anencephaly in females than in males, with a ratio of 2.3 to 1.0. Moreover, the pituitary gland is absent in up to 53% of cases (Angevine, 1938; Bernischke, 1956;

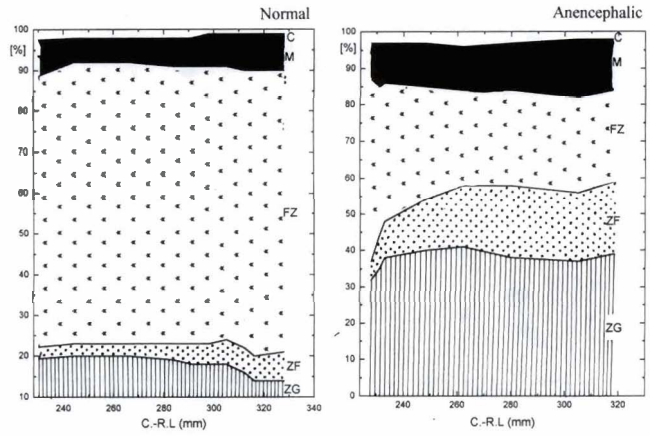

Fig. 7. Fractions (%) of adrenal volume of normal and anencephalic human fetuses occupied by adrenocortical zones, medulla and capsule. Explanations as in Fig. 6.

Sucheston and Cannon, 1970). In our material the ratio of anencephalic females to males was higher (4:1) and only 2 cases (out of 10) possessed well developed hypophysis.

As far as adrenal gland weight and volume are concerned, there is general agreement that development of the anencephalic adrenals is quite normal up to the fifth month of gestation. After that time a notable difference in the weight of the gland in comparison with normal fetuses are noted, an effect caused by a high rate of growth in normal fetuses and only an insignificant one in anencephalic fetuses (Angevine, 1938; Bernischke, 1956; Sucheston and Canon, 1970). Jost (1975) suggests that the weight of the fetal adrenal in anencephaly changes little from 15 to 20 weeks onward, being usually 1 g or less at birth (per one gland). In the report of Kenny et al. (1966) two live-born anencephalic infants (one died at 72 h, the other at 98 h) had adrenal glands of approximately 0.8 and 0.6 g.

In our material the weight and volume of adrenals from normal fetuses are similar to that described earlier (Bocian-Sobkowska et al., 1993). In anencephalic fetuses studied, the weight of adrenals was 1.80 g in the youngest case, 4.02 g in the oldest, 5.01 in fetus with hypophysis, and 9.97 g in anencephalic fetus with hypothalamus and hypophysis. Thus at week 24 of intrauterine life adrenals of normal fetuses are 2-fold heavier than those of anencephalic fetuses while at the 39th week the difference is even higher (ca 4-fold). These observations suggest that differences in adrenal weight and volume between normal and anencephalic fetuses have appeared before the mid-fetal period. This contention is supported by data of Gray and Abramovich (1980), who observed smaller adrenals in anencephalic fetuses at the 14th week of gestation. However, non-availability of so young anencephalic fetuses does not allow to perform us comprehensive longitudinal studies on this subject.

To our knowledge, the present report provides the first systematic description of the development of adrenal zonation in human anencephalic fetuses. Stereological data reported here are burdened with an error resulting from imperfect sampling of the adrenal gland. Counting performed only on transverse sections of the middle part of the gland certainly overestimates the volume of the adrenal medulla (Weibel, 1979). However, since comparable sections were studied from both anencephalic and normal glands, they may represent well the overall changes occurring during the adrenal development. Additional error may arise from

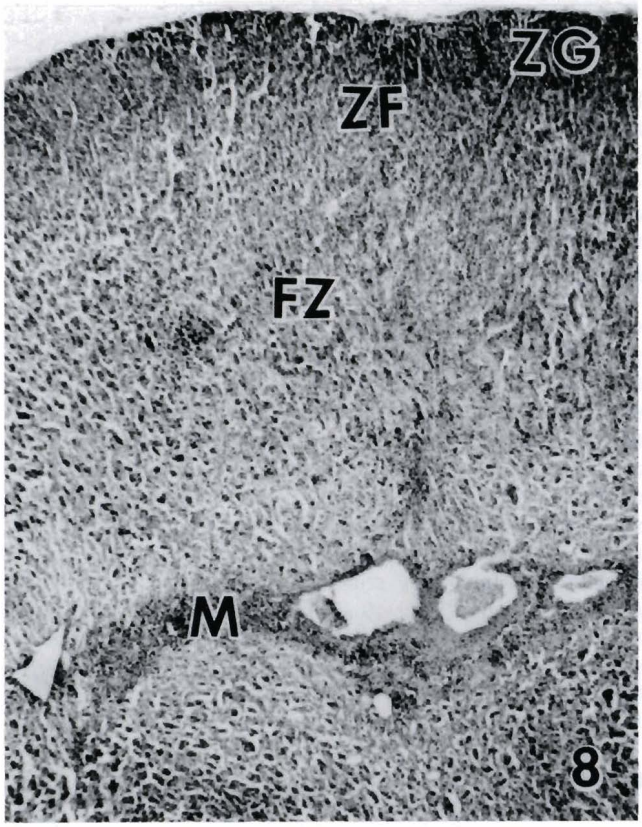

Fig. 8. Adrenal gland of normal 38-week-old human fetus - case No 10. The wide adrenal cortex contains well developed fetal zone surrounding abundant medullary tissue. Haematoxylin and eosin. x 30

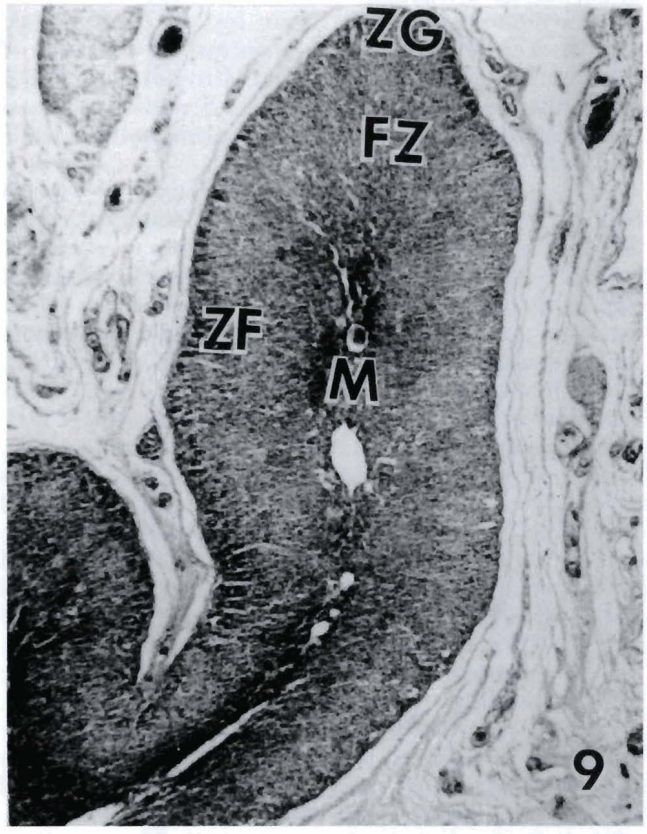

Fig. 9. Adrenal gland of anencephalic 37-week-old human fetus - case No. 7. The narrow adrenal cortex surrounds only small islets of chromaffin cells. Haematoxylin and eosin. x 30

accuracy of weighing of the adrenal gland which for anencephalic fetuses was 10 mg and for normal samples 1 mg.

Earlier data on zonation of the adrenal gland are reviewed by Bachman (1954) and more recently in our earlier reports (Bocian-Sobkowska et al., 1993, 1996). There is general agreement that the development of adrenal fetal zone is the main factor responsible for proper adrenal gland growth during the intra-uterine life. At the end of fetal period volume of this zone attains over 8200 mm^3 and constitutes 69-70% of the total gland volume, while respective values for ZG, ZF and M are 1665 (14%), 833 (7%) and 1071 mm^3 (9%). The above data are in striking contrast to those found in adrenals of anencephalic fetuses, in which volume of the gland attains only ca 35% of that seen in normal specimens. In the oldest anencephalic specimen studied (39th week of intrauterine period) volume of the fetal zone was 962 mm^3 (ca 8-folds lower than in normal fetus) comprising only 25% of the total gland weight. For the remaining zones the values were: ZG - 1501 mm^3 (ca 90% of the normal value and 39% of the total gland volume); ZF - 770 mm^3 (ca 92% and 20%, respectively), and M - 539 mm^3 (ca 50% and 14%, respectively).

Earlier reports estimated that at birth the fetal zone of anencephalic adrenals is reduced to about 25% of the gland volume (Bernischke, 1956; Jost, 1975; Van Hale and Turkel, 1979). On the contrary, by means of histometric technique Gray and Abramovich (1980) found that at gestational age of 17.5 weeks, the fetal zone comprises 58+3% of the total gland volume while in normal fetuses 77+2%.

It is generally accepted that until the fifth month of fetal period the development of the fetal adrenal cortex depends on maternal placental factors, presumably human chorionic gonadotropin (hCG) and/or placental adrenocorticotropin (ACTH). However, after 10-15 weeks of gestation the fetal pituitary ACTH is the main factor responsible for the growth of the gland (Jost, 1966; Fisher, 1986; Mesiano and Jafe, 1992). This statement is supported by studies in which ACTH was administered to human anencephalic fetuses in utero. Such treatment restores adrenals of anencephalic fetuses to the size of normal newborn with a well developed fetal zone (Johannisson, 1968; Honnebier and Swaab, 1973). Thus, it seems apparent that the impaired development of fetal and fasciculata zones of the adrenal cortex of anencephalic fetuses is caused by a lack of fetal ACTH secretion. However, as in the present study, the development of zona glomerulosa in anencephalic fetuses is similar to that observed in normal adrenals. During the whole studied period of intra-uterine life the volume of this zone increases linearly; however, due to marked atrophy of all inner zones in anencephalic gland this zone comprises about 40% of total gland volume, while in the normal gland it is only 14%. These findings suggest that during intra-uterine life the growth of zona glomerulosa is rather autonomous, and does not depend on the presence of fetal ACTH. Similar separate regulatory mechanisms for control of the zona glomerulosa and zona fasciculata/reticularis growth are present in other mammalian species (for review, see Nussdorfer, 1986; Vinson et al., 1994).

It is well known that in anencephaly there is a partial absence of the brain or its subdivisions. The adenohypophysis may be deficient or may become detached from the brain. According to Jirásek (1980) in anencephalic the adenohypophysis should always be present and it was found in 7 of 14 cases described in the literature (Müller and O'Rahilly, 1991). Data of Allen et al. (1974) indicated that the adenohypophysis of anencephalic fetuses contained a readily dischargeable pool of ACTH. But endogenous secretion or exogenous administration of this tropic hormone did not result in the expected acute secretory response by its target organ, presumably due to functional atrophy of the latter. These findings could explain our results in one anencephalic fetus which had well developed pituitary gland and demonstrated a lack of the hypothalamus. It showed middle values of investigated parameters between normal and anencephalic fetuses. It is probably due to the lack of a releasing factor from the hypothalamus, because adrenals of anencephalic fetus possessing hypophysis and hypothalamus do not differ from normal glands.

Present studies revealed a marked increase in the volume of adrenal medulla of normal fetuses in contrast with a slow linear growth of that zone in anencephalic fetuses. At the 24th week of fetal life volume of the adrenal medulla in normal versus anencephalic adrenals is 329 and 173 mm^3 while at the 39th week respective values are 1071 and 539 mm^3. The adrenal medulla is derived from sympathoadrenal neural crest lineage that migrate to adrenal primordium during the embryonic period (Landis and Patterson, 1981; Anderson and Axel, 1986; Unsicker et al., 1989). Glucocorticoids are essential for differentiation of sympathoadrenal progenitors into catecholamine-producing cells and for the induction of phenylethanolamine-n-metyltransferase (Michelsohn and Anderson, 1992; Anderson, 1993). In the anencephalic fetuses, with deficient provisional cortex and insufficient glucocorticoid production, this differentiation of neuroblasts is probably retarded. The relative increase in the neuroblastic component of the adrenal glands of anencephalic fetuses as compared with controls reflects the relative lack of cortex in these glands rather than an absolute increase in neural elements. The lack of sufficient glucocorticoid production may be related to the incidental neuroblastoma found int he adrenal gland of an anencephalic fetus (Van Hale and Turkel, 1979).

References

Allen J.P., Greer M.A., McGilvra R., Castro A. and Fisher D.A. (1974). Endocrine function in an anecephalic infant. J. Clin. Endocrinol. Metabol. 38, 94-98.

Anderson D.J. (1993). Molecular control of cells fate in the neural crest: the sympathoadrenal lienage. Annu. Rev. Neurosci. 16, 129-158.

Anderson D.J. and Axel R. (1986). A bipotential neuroendocrine precursor whose choice of cell fate is determined by NGF and glucocorticoids. Cell 47, 1079-1090.

Angevine D.M. (1938). Pathologic anatomy of hypophysis and adrenals in anencephaly. Arch. Pathol. 26, 507-528.

Bachman R. (1954). Die Niebenniere. In: Handbuch der mikroskopischen Anatomie des Menschen. Bargmann W. (ed). Bd 6/5. Springer. Berlin, Gottingen, Heidelberg. pp 1-952.

Bernischke K. (1956). Adrenals in anencephaly and hydrocephaly. Obstet. Gynecol. 8, 412-425.

Bocian-Sobkowska J., Malendowicz L.K. and Woźniak W. (1993). Cytological aspects of the human adrenal cortex development in the course of intra-uterine life. Histol. Histopathol. 8, 725-730.

Bocian-Sobkowska J., Wozniak W., Malendowicz L.K. and Ginda W. (1996). Stereology of human fetal adrenal medulla. Histol. Histopathol. 11, 389-393.

Branchaud C.L. and Murphy B.E.P. (1992). Physiopathology of the fetal adrenal gland. In: Hormone and fetal pathophysiology. Pasqualini J.R. and Choller R. (eds). Marcel Dekker Publ. New York, Basel, Hong Kong. pp 53-85.

Covelli W.P. (1927). A quantitative study of the hypophysis of the human anencephalic fetus. Am. J. Pathol. 3, 17-28.

Dhom G. (1981). Die Niebennierenrinde. In: Pathologie der endocrinen Organe II. Doerr W. and Seifert G. (eds). Springer-Verlag. Berlin, Heidelberg. New York. pp 794-797.

Elwood J.H. (1970). Anencephalus in the British Islets. Dev. Med. Child. Neurol. 12, 582-591.

Fisher D.A. (1986). The unique endocrine mileu of the fetus. J. Clin. Invest. 78, 603-611.

Giround D. and Martinet A. (1960). Anencéphalié expérimetale chez la souris et comparisons avec l'anéncephalié chez l´homme. Acta Anat. 43, 358-370.

Gray E.S. and Abramovich D.R. (1980). Morphologic features of the anencephalic adrenal gland in early pregnancy. Am. J. Obstet. Gynecol. 137, 491-495.

Honnebier W.J. and Swaab D.F. (1973). The influence of anencephaly upon intrauterine growth of fetus and placenta and upon gestation length. J. Obstet. Gyneacol. Br. Commonw. 80, 577-588.

Hurwitz C.H. (1955). Anencephalus. Obstet. Gynecol. 6, 303-308.

Jirásek J.E. (1980). Human fetal endocrines. Nijhoff. The Hague.

Johannisson E. (1968). The fetal adrenal cortex in the human. Acta Endocrinol. 130 (suppl.), 1-107.

Jost A. (1966). Problems of fetal endocrinology: the adrenal glands. Rec. Progr. Hormone. Res. 22, 541-574.

Jost A. (1975). The fetal adrenal cortex. In: Handbook of Physiology. Adrenal gland. Vol. VI. Greep R.O. and Estwood E.B. (eds). American Physiological Society. Washington DC. pp 107-115.

Keen M.F. and Hewer E.E. (1927). Observations on the development of the human suprarenal gland. J. Anat. 61, 302-324.

Kenny F.M., Predyasombat C., Spaulding J.S. and Migeon C.J. (1966). Cortisol production rates. IV. Infants born of steorid-treated mothers and of diabetic mothers. Infants with trisomy syndrome and with anencephaly. Pediatrics 37, 960-966.

Landis S.C. and Patterson P.H. (1981). Neural crest cell lineages. Trends Neurosci. 4, 172-175.

Mesiano S. and Jaffe R.B. (1992). Regulation of growth and differentiated function in the human fetal adrenal. In: Cellular and molecular biology of the adrenal cortex. Sach J.M., Brownie A.C., Capponi A., Chambaz E.M. and Mantero F. (eds). Colloque INSERM. Vol. 222. pp 235-245.

Michelsohn A.M. and Anderson D.J. (1992). Changes in competence determine the timing of two sequential glucocorticoid effect on sympathoadrenal progenitors. Neuron 8, 589-604.

Müller and O'Rahilly R. (1984). Cerebral disraphia (future anencephaly) in a human twin embryo at stage 13. Teratology 30, 167-177.

Müller F. and O'Rahilly R. (1991). Development of anencephaly and its variants. Am. J. Anat. 190, 193-218.

Nakano K.K. (1973). Anencephaly: a review. Dev. Med. Child. Neurol. 15, 383-400.

Nussdorfer G.G. (1986). Cytophysiology of the adrenal cortex. Int. Rev. Cytol. 98, 1-405.

Sucheston M.E. and Cannon M.S. (1970). Microscopic comparison of the normal and anencephalic human adrenal gland with emphasis on the transient zone. Obstet. Gynecol. 25, 544-557.

Swinyard C.A. (1938). Methods for volumetric determination of fresh endocrine glands. Anat. Rec. 74, 71-78.

Unsicker K., Seidl K. and Hofmann H.D. (1989). The neuro-endocrine ambiguity of sympathoadrenal cells. Int. J. Dev. Neurosci. 7, 413 417.

Van Hale H.M. and Turkel S.B. (1979). Neuroblastoma and adrenal morphologic features in anencephalic infants. Arch. Pathol. Lab. Med. 103, 183-203.

Vinson G.P., Hinson J.P. and Toth I.E. (1994). The neuroendocrinology of the adrenal cortex. J. Neuroendocrinol. 6, 235-246.

Weibel E.R. (1979). Stereologic methods. Vol. 1. Practical methods for biological morphometry. Academic Press. London. pp 1-415.

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