Virchows Arch. Abt. A Path. Anat. 355, 105–122 (1972)

Adrenal Cytomegaly

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Received September 16, 1971

Summary. In the adrenal cortex of newborn infants there can occasionally be found distinct, large cells with large, prominent nuclei. This penomenon of "adrenal cytomegaly" has not been satisfactorily explained. In a series of 300 necropsies of infants up to 10 days of age "cytomegalic" cells were found in 16 instances. In 11 of these there was serological or pathological evidence of Rh-incompatibility, but in the remaining five instances no such evidence was found. There were, however, various congenital abnormalities. The morphological features of the large adrenal cells raised the possibility that these cells were polyploid. The suggestion is advanced that adrenal cytomegaly is a non-specific phenomenon, a response to intense, prolonged stimulation for a variety of reasons, which produces changes in the endocellular structure that manifest themselves as polyploidy. The hypothesis put forward here can account better for the occurrence of adrenal cytomegaly in a number of seemingly unrelated conditions than the other explanations of this phenomenon advanced so far.

The phenomenon of adrenal cytomegaly is well known, but poorly understood. In 1970 it remains as much of an "enigma" (Irving, 1967) and as "controversial" (Oppenheimer, 1970) as it has been since Kampmeier's (1927) description. Estimates of its incidence range from 6.5% of necropsies in the newborn (Craig and Landing, 1951) to about one in one thousand (Stowens, 1966) but, as Morison (1963) has pointed out, there are probably many cases with minimal changes. At necropsy of some newborn, occasionally premature, infants who have died for a variety of reasons, there are found in the adrenal cortex sometimes a few, sometimes many, cells which are considerably larger than the surrounding adrenocortical cells and which have distinctly large, even giant, nuclei. Their origin and significance have given rise to various opinions.

Thus Garneau (1957), who reported an instance of adrenal cytomegaly in an infant whose mother had mumps during the pregnancy, suggested a viral etiology for this striking cellular change. Adrenal cytomegaly has also been found on occasion in instances of the congenital rubella syndrome (Singer *et al.*, 1967; Oppenheimer, 1970). Morison (1963), who looked critically at the "puzzling conditions of adrenal cytomegaly", also thought that a virus infection was a possible explanation, but Potter (1961), Stowens (1966), Irving (1967) and Oppenheimer (1970) questioned this. Kampmeier (1927) and Uotila (1940), who studied the development of the adrenal in the embryo, reported cytomegaly in fetuses of varying ages, the inference being that adrenal cytomegaly can occur as part of the fetal development. Craig and Landing (1951), while confirming these findings, were impressed by the "anaplastic" features of the large cortical cells characteristic of adrenal cytomegaly and considered the possibility that these cells might be the precursors of some of the carcinomas of the adrenal cortex. Landing (1955) even spoke of "focal anaplasia or cytomegalia". A few years later Sherman, Bass, and Fetterman (1958) did indeed publish a case which, according to their interpretation, seemed to fit Craig's and Landing's (1951) concept in that an infant with

a metastasizing adrenal carcinoma presented cytomegaly also in the metastatic adrenocortical nodules. Stowens (1966) reported eight instances of "cytomegalic tumors of the adrenal", only one of which, however, proved malignant. The nature of these tumors is not clear, but it is well known that adrenocortical adenomas often present cells which fit the criteria of "cytomegaly" (Dhom and Städtler, 1968). Recently the suggestion has even been made that "cytomegaly may represent an *in situ* carcinoma of the fetal adrenal cortex" (Sotelo-Avila and Singer, 1970). Other workers, however, were not impressed by the suspected link between adrenal cytomegaly and tumorigenesis (Dhom, 1965) and looked for other explanations. Thus Beatty and Hawes (1955) tried to establish a connection between the presence of "congenital anomalies" and the occurrence of adrenal cytomegaly. Some years later Bayer, Židová, and Dušek (1962) arrived at a similar conclusion. Prompted by the case reported by Diamond, Anderson and McCreadie (1960) of an infant born to a mother treated with Myleran, they thought that an intrauterine toxic disturbance of the fetus stimulated the adrenal cortex to the development of giant cells. In support of their views they pointed to Selye's concept of the adaptation syndrome although, to our knowledge, Selye has not described adrenal cytomegaly as part of this syndrome (Selye and Stone, 1950). An association between adrenal cytomegaly and congenital anomalies has recently also been emphasized by writers on the "Beckwith Syndrome" (Sotelo-Avila and Singer, 1970; Beckwith, 1969). Other workers such as Potter (1961) or Kissane and Smith (1967) have, in view of the scanty knowledge of the factors leading to the development of this phenomenon, refrained from speculation and have merely recorded their findings, whereas yet others have ignored the problem altogether (Luse, 1967).

In view of these considerations it would seem that a re-examination of the question of the significance of adrenal cytomegaly can be defended on the grounds that the explanations advanced so far are merely divergent speculations which apparently do not possess any common features. In the paper presented here an admittedly speculative, but unifying concept is proposed, based on the thesis that adrenal cytomegaly is merely an indication of polyploidy of the adrenocortical cells which develops when, for a variety of reasons, the cells are intensely stimulated.

Material and Methods

The material on which this survey is based comprises a review of sections of the adrenal glands from 300 consecutive necropsies of newborns and infants up to the age of 10 days. Stillborn, but not severely macerated, infants were included in this series. The interval between death and necropsy ranged from two to eighteen hours. The necropsies were performed in the routine manner which, of course, includes the weighing and cutting of both adrenal glands, representative sections of which were fixed in buffered formalin solution, embedded in paraffin and cut at 6 μ. The sections were stained with hematoxylin-eosin, the periodic acid-Schiff (PAS) reagents and with Gomori's methenamine silver stain for reticulin. Selected sections were also stained with the Feulgen stain.

The following procedure of evaluating the material was adopted: First the sections of both adrenals from the 300 autopsies were studied for the presence or absence of cytomegalic cells. Since these cells have a striking appearance with a much larger body and nucleus than normal adrenocortical cells, no difficulty was encountered in reaching this decision. It should be emphasized, however, that the number of cells present in a given section did not influence this decision, so that on occasion a gland was classified as "cytomegalic" even if only a few such cells were found in the sections. Step-sectioning of the blocks from adrenal glands considered to be "negative" was not undertaken. It is possible that such a deliberate search for adrenocortical "megalocytes" might have increased the number of specimens considered to be positive. Following the preliminary screening of all sections the necropsy protocol and the clinical charts of the mother and infant were reviewed in those instances in whom cytomegalic cells were found in the adrenals, and any features considered significant were tabulated. It is, therefore, apparent that the diagnosis of cytomegaly had always been made before the clinical or pathological features of these cases had been studied.

Results

Cytomegalic cells in the adrenal cortex were found in 16 out of 300 necropsies reviewed. As can be seen from Table 1, five of these infants were stillborn, four died 1 day, three 2 days, three 3 days and one 4 days after birth. In 11 out of 16 cases there was present serological and/or pathological evidence of Rh-incompatibility. Of the remaining 5 instances one case had the malformation of anencephaly and presented the typical hypoplasia of the adrenal glands found in this malformation. Another infant was born with a large omphalocele for which an operation was attempted, but the child died shortly afterwards. One infant

Table 1. *Clinical data of authors' series of necropsies with adrenal cytomegaly*

Age	Sex	Estimated gestation (weeks)	Weight (g)	Diagnosis
Stillborn	♂	25	700	Rh incompatibility
Stillborn	♂	22	750	Immaturity
Stillborn	♀	28	1175	Anencephaly
Stillborn	♀	34	1350	Rh incompatibility hydrops. Intrauterine transfusion
Stillborn	♀	28	1175	Rh incompatibility Hydrops
1 day	♂	30	1715	Omphalocele, operation
1 day	♀	Dates not available	3250	Rh incompatibility Icterus
1 day	♀	38	3525	Rh incompatibility Icterus
1 day	♂	37	3730	Rh incompatibility Hydrops
2 days	♂	30	1025	Prematurity, Single umbilical artery Abruptio placentae
2 days	♂	35	2150	Rh incompatibility Hydrops
2 days	♀	35	2825	Rh incompatibility Icterus
3 days	♂	36	1600	Rh incompatibility Icterus
3 days	♂	35	2800	Rh incompatibility
3 days	♀	40	3100	Adrenogenital syndrome with external appearance of female pseudohermaphrodite. Transposition of great vessels
4 days	♀	32	1800	Rh incompatibility Icterus

Table 2. *Clinical data of present series of infants with erythroblastosis fetalis showing adrenal cytomegaly*

Age	Sex	Gestation (weeks)	Weight (g)	Maternal history	Antibody titre	Infants history, condition
Stillborn						
1	♂	25	700	G6,P5. All children jaundiced (4 with exchange transfusions). Amniocentesis. Intrauterine death 2nd day after intrauterine transfusion	1:1280	Maceration, moderate
2	♀	34	1350	G3,P2 (2nd with exchange transfusion). Intrauterine transfusion	1:160	Hydrops
3	♀	28	1775	G4,P3 (1st normal, 2nd and 3rd stillbirths); mild toxemia	1:5120	Hydrops
1 day						
4	♀	Dates not available	3250	G5,P4	1:320	Hb-11.4 g-%. Icterus, Coombs test +, Anasarca
5	♀	38	3525	G7,P6 (6 with exchange transfusions). Refused induced labor	not recorded	Hb—8 g-%. Exchange transfusion; death 3 hours after exchange transfusion
6	♂	37	3730	G5,P4 (3 with exchange transfusions). C/S for abruptio placentae	1:16 incomplete	Hb—8 g-%. Hydrops, Bilirubin—10 mg% at birth. Coombs test +. Exchange transfusion.
2 days						
7	♂	35	2150	G4,P3 (no history). Induced labor	1:160	Hb—4.4 g-%. Icterus 36 hours of age. Death 1 hour after exchange transfusion
8	♀	35	2825	G8,P7 (3 normal, 1 miscarriage by first husband; 2 normal. One stillborn by 2nd husband)	1:180, 3 mo. before delivery	Hb—4 g-%. Hydrops. Exchange transfusion, 27 hours of age
3 days						
19	♂	36	1600	G6,P5 (3 normal, 1 exchange transfusion, 1 stillborn)	1:128	Icterus. Exchange transfusion immediately after birth and at 54 hours of age
10	♂	35	2800	G3,P2 (1 exchange transfusion) Amniocentesis. Induced labor	1:5120	Hb—4 g-%. Icterus, exchange transfusion.
4 days						
11	♀	32	1800	G6,P5 (2 normal, 2 stillborn, 1 jaundiced)	1:640	Icterus, Hydrops, Coombs test + Exchange transfusion

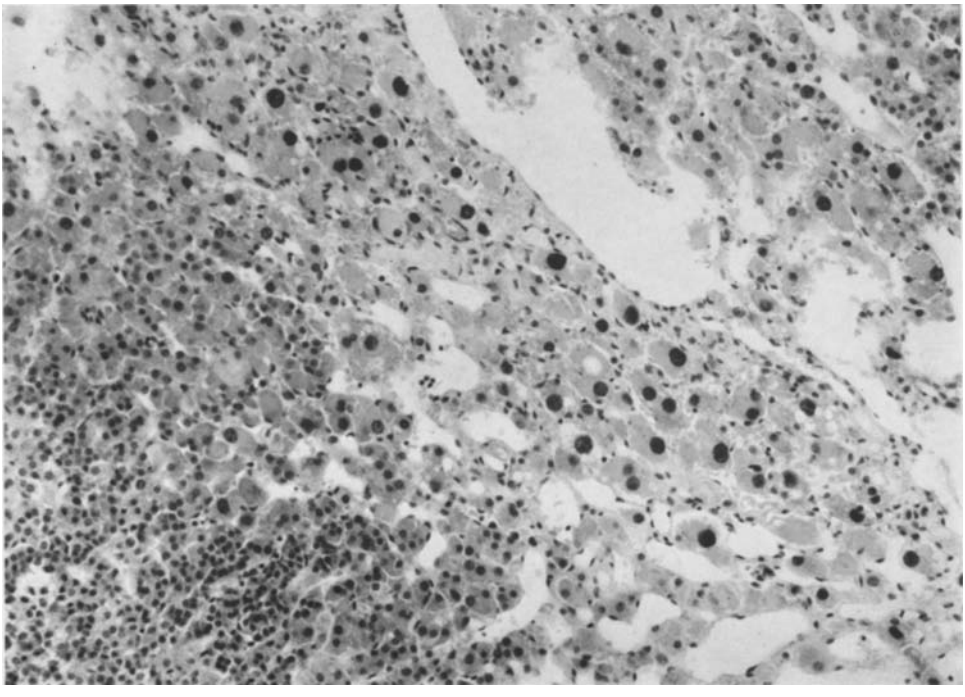

Fig. 1. Microphotograph of adrenal cortex of an infant with erythroblastosis fetalis to show the distribution of the adrenal "megalocytes".—Note the large, prominent nuclei. H.E., 6 μ. ×100

died 3 days after birth with the clinical diagnosis of the adrenogenital syndrome with the external appearance of a female hermaphrodite. At necropsy very large adrenal glands as well as a transposition of the large vessels of the heart were found. Another infant died after the mother had developed an abruptio placentae; necropsy showed multiple hemorrhages, and only two vessels in the umbilical cord. Finally, in one of the stillborn infants in whom Rh-incompatibility could not be invoked, the only diagnosis made at necropsy was that of "immaturity". These data are presented in Table 1.

In Table 2 are shown some of the pertinent data concerning the eleven cases in whom a clinical diagnosis of erythroblastosis fetalis was made, and confirmed by necropsy. It is not possible to correlate either the distribution of the cytomegalic cells or their numbers with the clinical picture. Of interest in this context is the fact that in our series of 300 necropsies 20 cases were considered to be erythroblastotic, but adrenal cytomegaly was found in only eleven of them The question again arises whether a systematic search for adrenal cytomegalic cells by step-sectioning of the adrenal glands would have altered this proportion which, incidentally, is considerably higher than that reported by other workers (Oppenheimer, 1970; Craig and Landing, 1951; Bayer *et al.*, 1962). To some extent this high incidence can be attributed to the fact that adrenal glands

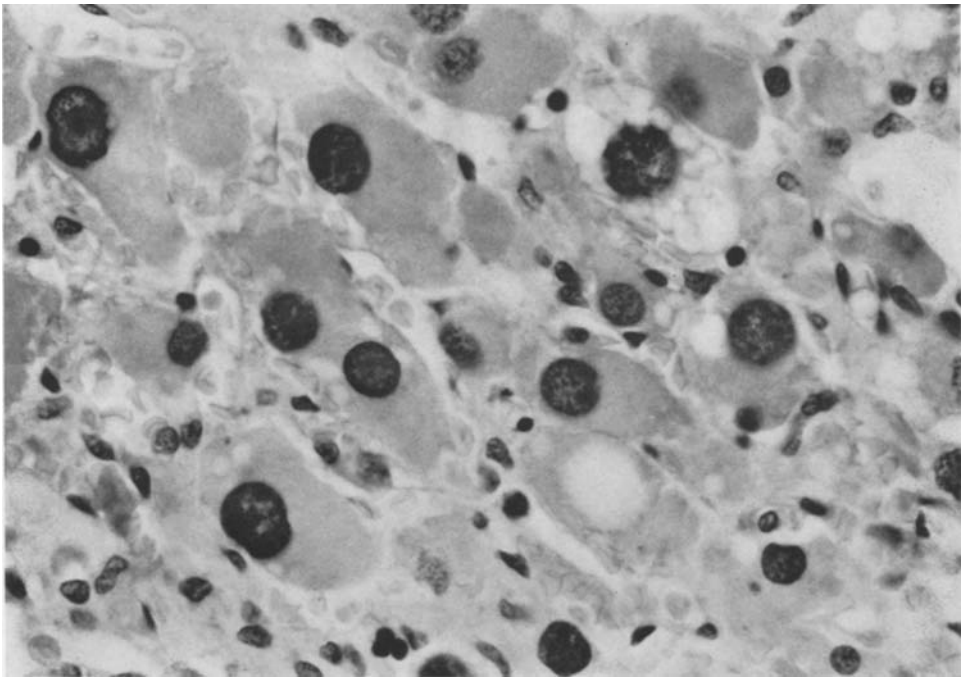

Fig. 2

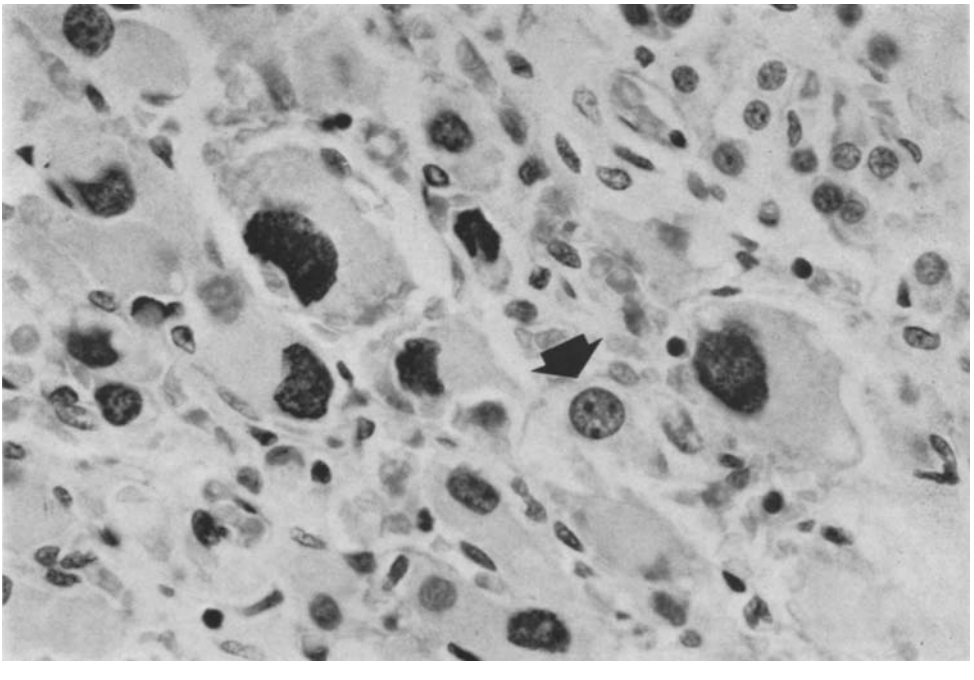

Fig. 3

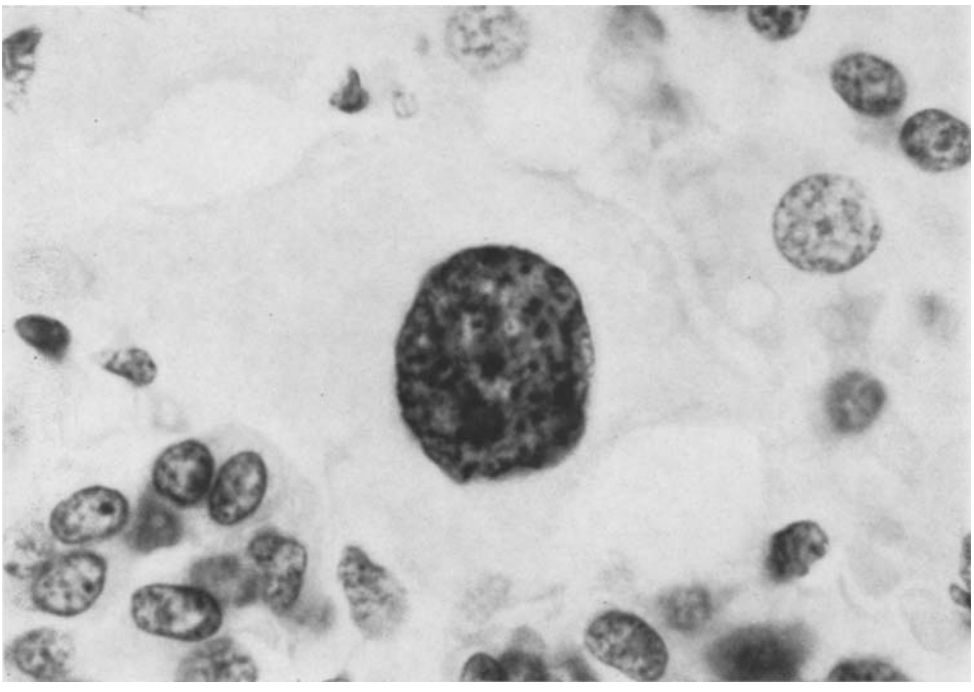

Fig. 4. High magnification of a megalocytic nucleus from the adrenals of an erythroblastotic infant to show the characteristic nuclear structure. A nucleolus is faintly visible surrounded by clumps of chromatin (same section as Fig. 2) $\times$ 1200

containing only a few adrenal "megalocytes"[1] were included in the series as being "cytomegalic". The number of these cells varied considerably from case to case; on occasion they were quite numerous (Fig. 1). In the series presented here they were only seen in the "fetal" cortex, and although they tended to be present mainly in the inner third, this was by no means invariably so. The cells were easily recognized by their large size and, above all, by their prominent

1 The terms "megalocyte" and "megalocytosis" have been used by Bull (1954) and by Jago (1969) to describe the significant increase in the size of the liver cells and their nuclei in experimental animals under the chronic influence of pyrrolizidine alkaloids. These purely descriptive terms are used here as synonyms for "cytomegaly". The "megalocytes" of the adrenal cortex are, of course, unrelated to the well-known "megalocytes" of the red blood cell series. There is no reason why this convenient descriptive term should be restricted to erythrocytes.

Fig. 2. Higher magnification of a field of Fig. 1.—Note the eccentric position of some of the nuclei, and the vacuolated cytoplasm of the cell in the upper right hand corner. H.E., 6 μ. $\times$ 500

Fig. 3. Microphotograph to show the characteristic nuclear appearance of "cytomegaly" in the adrenals of an erythroblastotic infant.—Compare these nuclei with the normal nucleus (arrow), and note the deformed shape of the nuclei of the "megalocytes". H.E., 6 μ. $\times$ 500

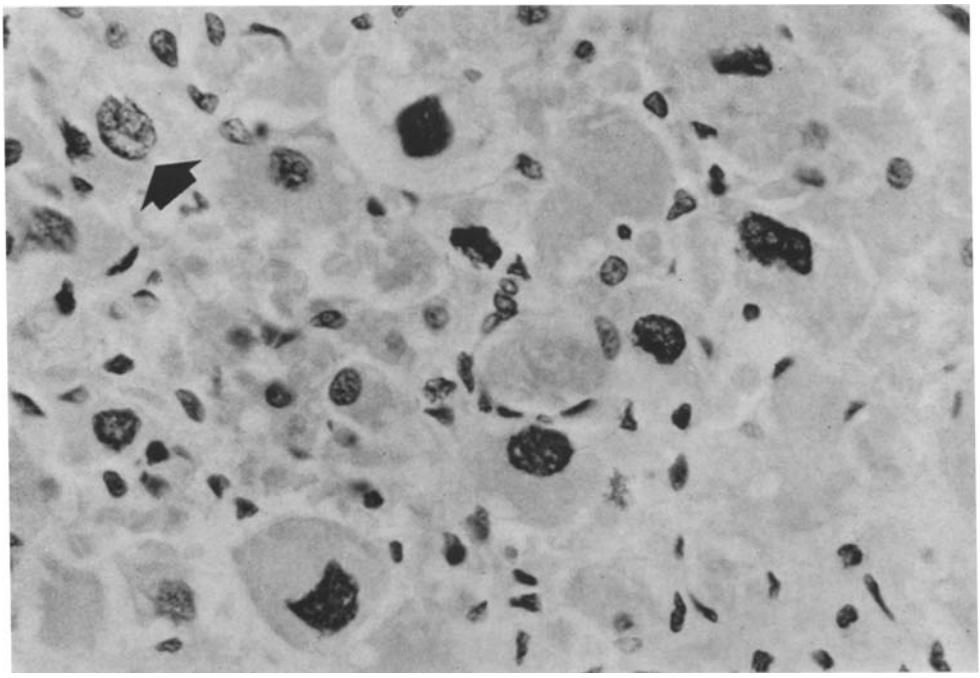

Fig. 5. Microphotograph to show the cytomegaly in the adrenal cortex of an infant with erythroblastosis fetalis.—Note the prominence of the nuclei in sections stained by the Feulgen stain. ×500. (Arrow =normal nucleus)

and very large nuclei. It is the appearance of the nucleus that is so characteristic for this cellular change (Figs. 2, 3). The nucleus can occupy the center of the cell, but can also be found shifted towards one pole of the cell. Its prominence is due not only to its size, but also to an increase in the amount of chromatin. Often the nucleus appears dense, so that not much detail may be recognized but if the interval between death and fixation is short and fixation is good, a prominent chromatin network (Fig. 4) is seen in the nucleus, consisting not only of more, but also of rather coarser threads and aggregates than are found in the normal nucleus. The appearance is faintly suggestive of the "Chinese Scroll work" of the megaloblast nucleus of the red cell series. While this can be seen quite well in sections stained with hematoxylin-eosin, it is accentuated by the Feulgen stain (Fig. 5). Nucleoli may or may not be present, but regressive nuclear and cytoplasmic changes are common. In the eosinophilic cytoplasm they take the form either of an ill-defined area of pinker staining, homogeneous

Fig. 6. Another microphotograph to show regressive nuclear changes in the form of vacuolation.—Note the chromatin network. 6 μ, Feulgen stain. ×340

Fig. 7. Microphotograph of cytomegalic cells from hyperplastic adrenal glands of adrenogenital syndrome to show nuclear pyknosis and dissolution. H.E., 6 μ. ×500

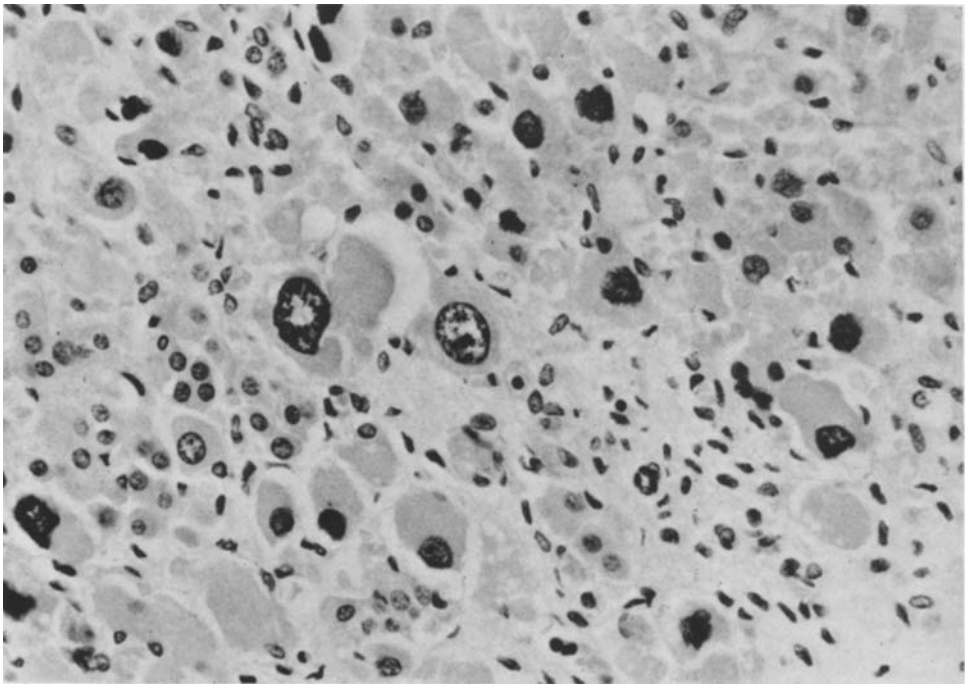

Fig. 6

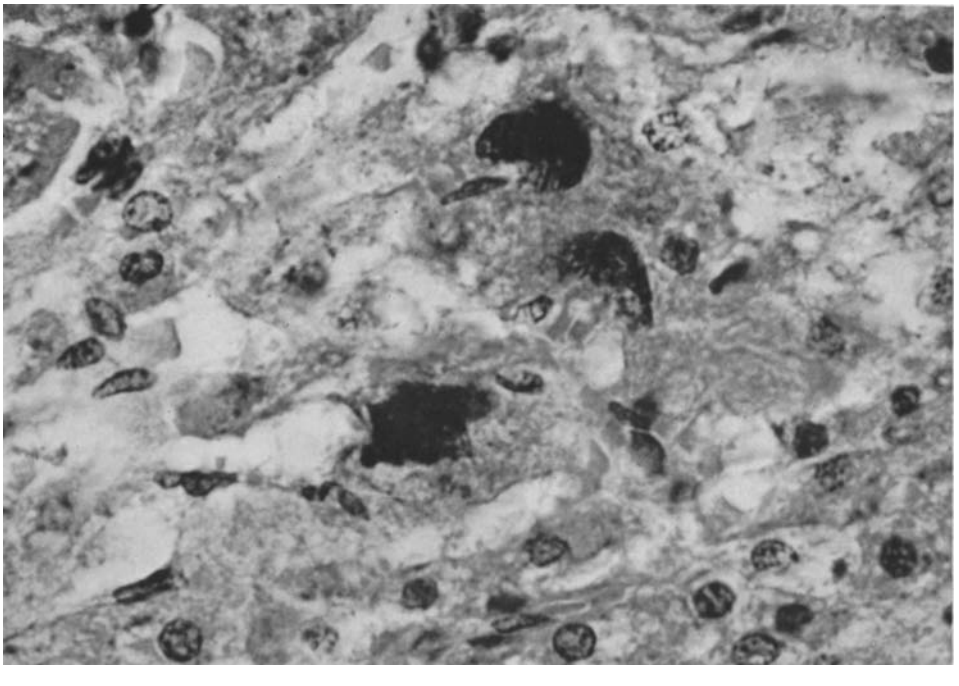

Fig. 7

condensation, or of irregular vacuolation (Fig. 2); in the nucleus they are shown by condensation and "pyknosis" of the chromatin, formation of empty (Fig. 6) or of eosinophilic nuclear inclusions of variable size and, above all, deformities of the nuclear outline so that the shape of the nucleus becomes quite bizarre. In the end nuclear fragmentation and dissolution may follow (Fig. 7). While most of these observations are similar to the descriptions given by some other workers (Craig and Landing, 1951; Morison, 1963; Potter, 1961; Beatty and Hawes, 1955; Diamond *et al.*, 1960; Kissane and Smith, 1967), the frequency with which regressive nuclear changes are found should be emphasized. It is unlikely that these changes are simply an expression of the normal involution of the fetal adrenal cortex in which the "megalocytes" participate. The question may well be asked whether these alterations are not rather an indication of an increased vulnerability of the enlarged cells to a variety of stimuli which must, of course, also include the normal process of involution of the adrenal cortex. The fact that these large cells may participate in the physiological involution of the fetal cortex should not be taken to imply that the phenomenon of cytomegaly itself is an expression of this involution. This has been emphasized by Potter (1961). One final point should be stressed. It was not possible to differentiate between various types of "cytomegalic cells". Thus the cells found in the cortex of the hyperplastic glands of the adrenogenital syndrome closely resembled those of the hypoplastic cortex of the anencephalic infant of our series, and these in turn were similar to the cells found in the adrenals of infants dying with erythroblastosis fetalis.

Discussion

The findings presented here suggest that in the newborn infant there exists an association between adrenocortical cytomegaly and erythroblastosis fetalis. No significant association between certain signs of erythroblastosis fetalis which might reflect the severity of this disorder, such as intensity of jaundice, the presence of Kernicterus, hydrops, etc., could be established. The only salient feature appears to be the fact that a high percentage of infants with adrenal cytomegaly also present clinical and histological evidence of blood group incompatibility. This observation is by no means new.

Craig and Landing (1951) had noted that in about 30% of their instances of adrenal cytomegaly erythroblastosis fetalis was also present. The higher incidence found in our series is probably due to the inclusion of minor degrees of cytomegaly. Dhom (1965) and Bayer *et al.* (1962) also pointed to the possible association between cytomegaly and erythroblastosis fetalis and, in a somewhat different context, Gasser (1954) reported an instance of adrenal cytomegaly in an infant with hemolytic anemia. There have, however, been published reports of the occurrence of adrenal cytomegaly in instances where a blood dyscrasia has not been associated. Five such cases were also found in our series. Attention has already been drawn to the paper by Beatty and Hawes (1955) who noted the occurrence of congenital anomalies in instances of adrenal cytomegaly and who, therefore, thought that there must exist a connection between these changes. Close examination of their data, however, shows that the "congenital anomalies" listed comprise a rather heterogenous group. It contains obviously serious disorders such as fibrocystic disease of the pancreas or malformations of the urinary tract, as well as anomalies of questionable significance such as Meckel's diverticulum. Since no further details of these anomalies are given, the data of Beatty and Hawes (1955) are difficult to evaluate. Eight years later Bayer *et al.* (1962) arrived at con-

clusions essentially similar to those of Beatty and Hawes (1955). These authors point to the high percentage of cases with adrenal cytomegaly who also had other anomalies. More recently Beckwith (1969), Sotilo-Avila and Singer (1970) and Oppenheimer (1970) have also commented on the presence of an assortment of congenital anomalies in infants with adrenal cytomegaly—the "Beckwith Syndrome". Hence it is pertinent to state that an occasional case in our series also presented additional congenital malformations such as omphalocele or anencephaly. While the occurrence of an omphalocele will obviously interest the upholders of the "Beckwith Syndrome", we would like to emphasize at this stage the cytomegaly in the anencephalic. Some years ago Kerenyi (1961) suggested a division of the rare cases of congenital adrenal hypoplasia into a primary or "cytomegalic", and a secondary or "anencephalic" type. Bayer *et al.* (1962), however, reported an instance of cytomegaly in the hypoplastic adrenals of an anencephalic infant and pointed out that, therefore, this division into an anencephalic as opposed to a cytomegalic type of hypoplasia cannot be maintained. It should be noted that in the series presented here we also found one instance of an anencephalic monster whose hypoplastic adrenals contained adrenocortical "megalocytes", so that this aspect of the paper by Bayer *et al.* (1962) can be confirmed. But these instances apparently are rare. While we would agree that cytomegaly alone can no longer be considered as a criterion for the classification of adrenal hypoplasias, a division of the congenital hypoplastic adrenals into a secondary, "anencephalic", and into a primary, or "non-anencephalic" type would, on morphological grounds, still appear to be justified, particularly if the frequency with which cytomegaly has been encountered in the "primary" type (Šikl, 1948; Deamer and Silver, 1950; Harlem and Myhre, 1957; MacMahon *et al.*, 1957; Mitchell and Rhaney, 1959) is kept in mind. MacMahon *et al.* (1957) realized this difference when they stated that "we are dealing here with a very small adrenal gland composed for the most part of very large cells, a picture that is quite different from the miniature adrenals that one so commonly associates with the anencephalic state". The occurrence of adrenocortical "megalocytes" in the suprarenals of the anencephalic is, however, also of great interest from another point of view. It presents a striking counterpart to the occasional occurrence of cytomegaly in the huge cortex of the adrenocortical hyperplasia of the congenital adrenogenital syndrome. This was reported by Craig and Landing (1951) and by Oppenheimer (1970) in one instance each and was also seen in one of our cases presented here.

We are, therefore, faced with the fact that cytomegaly can be encountered in the hypoplastic as well as in the hyperplastic adrenal gland—a confusing situation which, however, can be resolved by our postulate that in both instances adrenocortical cells are, presumably, intensely stimulated and adapt to the demands made on them by developing an appropriate cellular machinery in the form of nuclear and cytoplasmic enlargement, i.e., polyploidy.

This suggestion is based entirely on the appearance of the adrenal "megalocytes" in sections stained with such routine dyes as hematoxyline-eosin, or with the Feulgen reagents. It may, therefore, be appropriate to draw here attention to Geitler's (1953) treatise on endomitotic polyploidy. According to him a nucleus can be considered polyploid if it is not only considerably enlarged, but if it also shows a considerable increase in the amount of chromatin. Nuclear size alone is not enough; there must also be present certain structural changes in the nucleus. While, as has been pointed out earlier, these nuclear structural characteristics may not always be clearly visible because of regressive changes taking place in the nucleus for a variety of reasons not necessarily related to the stimulus giving rise to polyploidy, they should, nevertheless, be firmly kept in mind. Recently, for instance, de Baker *et al.* (1967) reported the case of a young child with hypoplastic adrenals which were also stated to be "cytomegalic". The cells, however, were described as large with nuclei of normal size. They, therefore, seem to differ from those discussed in the present paper. Cellular

enlargement can take place for a variety of reasons, but does not necessarily merit the designation of "cytomegaly" as used here or by other workers quoted in the present review. It is, therefore, proposed to limit the term "adrenal cytomegaly" to those cells that present certain morphological characteristics, such as significant nuclear enlargement and increased amounts and prominence of chromatin in the form of thick threads and clumps. These changes, in turn, are suggestive of polyploidy.

The possibility that adrenal cytomegaly might represent polyploidy had already occurred to Craig and Landing (1951) and is also mentioned by Dhom (1965), but they did not follow up this lead. Applying Geitler's (1953) criteria of polyploidy, however, to the material reviewed in the present series, the possibility, that the changes seen in the adrenocortical cells did indeed represent polyploidy, strongly suggested itself. Although we have not undertaken quantitative measurements, slides stained with the Feulgen reagent are well in keeping with this interpretation of the morphology of adrenal cytomegaly, and the recent studies of Borit and Kosek (1969) bear this out. Craig and Landing (1951) were struck by the similarity of these cells to adrenocortical cancer cells. Cancer cells can, of course, frequently be polyploid: "It has been seen that they often have an abnormally large nucleus, and cytology has shown that this property corresponds to a polyploid or heteroploid condition of the chromosomes" (Le Breton and Moule, 1961). It is, therefore, understandable that Craig and Landing (1951) and, later on, Sherman *et al.* (1958) should have been impressed with the "anaplastic" aspects of these cells. It is, however, less easy to understand why Sotelo-Avila and Singer (1970) should have separated these "anaplastic" cells in the fetal cortex—to them evidence of cancer *in situ!*—from similar cells in the adrenal cortex of the adult or, for that matter, of the infant suffering from conditions other than the "Beckwith Syndrome". For polyploidy ". . . cannot be considered as a specific characteristic inseparable from carcinogenesis" (Le Breton and Moule, 1961), and the question arises as to the significance of polyploidy in non-neoplastic conditions.

Information on this point is rather scanty. According to Gabe and Arvy (1961) ". . . it is difficult to give a functional interpretation of the relation of the activity of certain tissues and endomitosis"; they quote Geitler (1953) as having pointed out that "in all probability endomitosis creates a lesser functional disturbance than mitosis; this would explain the functional significance of endomitosis in especially active gland cells". Although Rather (1958) considers this a questionable assumption, he states elsewhere that ". . . in comparison with mitotic division, endopolyploidization appears to be a shorter process, and one less apt to interfere with the normal activity of the cell". Similarly Bungenberg de Jong (1957) also states that "if cells are implicated in an important functional process in an early stage, this function should not be allowed to be interrupted for the sake of growth". Since the process of mitosis involves the whole cell, endomitosis would render possible growth without the function of the cell being unduly disturbed. Furthermore, "Endopolyploidy or polyteny represent one of the two possible ways of increasing, in the cell, the quantity of material directly concerned with the intracellular synthesis of proteins" (Le Breton and Moule, 1961), and thus, presumably, also of other substances which result from the increased function of the cells. The findings of Borit and Kosek (1969) are well in keeping with this interpretation of the functional significance of polyploidy. While this, admittedly teleological, interpretation at the moment can only be viewed as a working hypothesis, it can account for the otherwise "puzzling" (Morison, 1963) fact that adrenal cytomegaly has been found in a variety of conditions which apparently are unrelated. "No common feature is present to explain the

significance of the change" (Morison, 1963). Potter (1961) similarly has pointed out that she could not find any "recognizable similarity in maternal conditions or in the causes of the infants' death".

The difficulty in understanding this phenomenon seems to have resided mainly in the efforts of some workers to ascribe the development of the large cells of the adrenal cortex exclusively to one factor or to another. We prefer, in keeping with the observations of Potter (1961) and of Morison (1963), quoted above, to view adrenal cytomegaly merely as an expression of the demands made by a variety of factors on the functioning adrenocortical cells. How can one otherwise explain the frequency of cytomegaly in erythroblastosis fetalis—a protracted intrauterine "stress" which clearly must considerably tax the mechanisms of adaptation—or its association with congenital malformations, usually of a serious nature? How is one to account for the presence of cytomegaly in the hypoplastic adrenals of the anencephalic monster as well as in the hyperplastic gland of the infant with adrenogenital syndrome? The stimulus need not necessarily be a pathological one. The claims of Kampmeier (1927), Uotila (1940), Craig and Landing (1951) and Bachmann (1954) of having encountered adrenal cytomegaly in some fetuses, and the presence of a rare cytomegalic cell in the adrenal cortex of an embryo of 5 cm's (crown-rump) length from a spontaneous abortion in our own material, not included here, suggest that even seemingly normal, "physiological", demands made on the adrenals at a time of rapid growth may lead to the development of adrenocortical polyploidy, perhaps because the polyploid cell may be better equipped to meet these demands under the prevailing conditions.

Some additional pertinent points should be mentioned here. Cytomegaly is not a feature which is limited to the adrenals and, therefore, characteristic for them.

Diamond *et al.* (1960), for instance, have described the very interesting case of an infant born to a leukemic mother treated with Myleran. At necropsy the infant showed extensive cytomegaly of the adrenals, but a similar cytomegalic change was also seen in several other organs. Cytomegalic cells in the testis have been found in one infant with adrenal cytomegaly by Sotelo-Avila and Singer (1970). Cytomegaly has also been described in the hyperfunctioning follicular cells of the human thyroid gland in hyperthyroidism (Wägelin, 1926) and, under varying conditions, particularly in the liver of experimental animals (Jago, 1969; Bull and Dick, 1959; Schoental and Magee, 1959; Schoental and Bensted, 1963). Widespread cytomegalic change in a variety of organs, including the adrenals, has recently been described in cases of ataxia teleangiectasia (Aguilar *et al.*, 1968). These observations support the view that under certain poorly understood conditions many cell types can become cytomegalic or polyploid, perhaps because normal mitosis is inhibited or because they can better function under these conditions. Another point of interest is the frequency with which regressive nuclear changes are found in cytomegalic adrenocortical cells. No attempts, to our knowledge, have been made to explain this change in the adrenal, but de Jong (1957) has drawn attention to the nuclear distortions frequently found in polyploid cells. It may well be that the advantage of the postulated heightened, or more effective, functioning of the polyploid cell is also accompanied by its increased vulnerability or by a shortened life span. This possibility is in keeping with the tentative conclusion of Bull and Dick (1959) who also thought that the "very accelerated polyploid change" seen in the liver cells of experimental animals treated with pyrrolizidine alkaloids "carries with it a precocious senility or a high susceptibility to certain injurious substances or both".

Finally, and this point fits well with the hypothesis suggested here, adrenal cytomegaly is not a feature of the fetal adrenal cortex only. Although adrenal cytomegaly is most frequently seen, and hence best known, in the cortex of

the premature or full-term neonate, it is by no means limited to this age group. Large cells with, often, giant nuclei have been found quite frequently in the adrenals of patients with Addison's disease (Kissane and Smith, 1967; Dietrich and Siegmund, 1926; Steinbiss, 1926; Guttman, 1930; Wells, 1930; Russfield, 1955). Fig. 4 of Guttman's (1930) paper, for instance, depicting a cytomegalic cell from the adrenal cortex of an adult patient with Addison's disease, closely resembles the cells seen in the adrenal cytomegaly of infants. Sotelo-Avila and Singer (1970) are merely confusing the issue when they caution that "these enlarged cells in the cortex of adults should not be confused on morphological grounds", without adducing one single criterion by means of which these cells can be distinguished. Wells (1930) viewed these large cells as representing "undoubtedly a process of regeneration or compensatory hypertrophy". The occurrence of adrenal cytomegaly in adult Addison's disease is of interest in view of the presence of adrenal megalocytes in the "primary" type of congenital adrenal hypoplasia in infants which frequently also produces signs of adrenal hypofunction; the occasional finding of adrenocortical giant cells also in the "anencephalic" type has already been alluded to. Adrenal cytomegaly has also been demonstrated in various other pathological conditions involving the adrenals, such as Cushing's disease or aldosteronism (Bayer *et al.*, 1962; Therien *et al.*, 1959; Kracht and Tamm, 1960; Cohen *et al.*, 1959; Chute *et al.*, 1949). These instances of adrenal overactivity again find a parallel in the occasional case of congenital adrenal hyperplasia with cytomegaly in the infant.

Adrenal cytomegaly, therefore, is not a characteristic of the fetal adrenal cortex only, but seems to be one of the possible responses of adrenocortical cells to stimuli, whose nature can vary, but whose effect must be an activation of the endocellular mechanisms concerned with the heightened or altered function of those cells that are responsive. Hence the surviving cells in Addison's disease as well as the stimulated cells of the hyperplastic cortex or the neoplastic cells of adrenal tumors can present the same cytomegalic characteristics leading to the appearance of polyploidy. The occurrence of adrenal "megalocytes" in erythroblastosis or in "stressful" congenital abnormalities also falls into this category of cells with an activated endocellular machinery. We have so far avoided the use of the terminology of "stress", although clearly most of the features of the problem presented here could easily be interpreted in these terms. But it seems that the acute stimulation of the adrenal cortex by ACTH does not produce fully developed polyploidy, only nuclear enlargement (Stark *et al.*, 1965; Pehlemann and Hanke, 1968) and/or mitosis, unless perhaps—as in all instances encountered here—the stimulation is protracted and not "sudden". This question had already been raised in a modified form by Tonutti, Bayer, and Spiegelhoff (1960/61) in their discussion of the adrenogenital syndrome. Attention should be drawn in this context to an interesting parallel from the field of experimental pathology of the liver. Schoental and Magee (1959), Bull and Dick (1959), and Jago (1969) noted that only the prolonged effect of hepatotoxins led to the induction of hepatic megalocytosis. Rather (1958), on the other hand, stated that polyploidy is frequently encountered in glands characterized by intensive, but transitory states of hyperfunction. The tacit assumption of the involvement of the pituitary-adrenal axis in "stress" would at first thought

seem to be contradicted by the occasional occurrence of cytomegaly in the hypoplastic adrenals of the anencephalic infant with a small adenohypophysis. A moment's consideration, however, will show that this objection is not really pertinent to the problem of cytomegaly or polyploidy, the conditions for whose development are unknown. We can merely infer that the cytomegaly of the adrenal cortex occurs under conditions which must rather severely tax the cortical cells, so that some cells—their number may perhaps depend on the severity of the stimulus—become polyploid, because this cytological change enables them to meet the demands placed upon them. While it is difficult to envisage such a change in the absence of ACTH, there is no convincing evidence in the human that it must be ACTH-dependent. It is, however, interesting to note that the presence of giant cells in the adrenals has been interpreted by earlier workers as evidence of pituitary activity (Russfield, 1955), and Beckwith (1969) states that "cytomegaly in endocrine tissues is in general a response to overstimulation". We should, therefore, like to conclude by summarizing our views to the effect that in our opinion cytomegaly of the adrenal cortex is merely an example of a cellular phenomenon which can be seen in other cells as well, which can occur, as far as the adrenals are concerned, under conditions which call not necessarily for the increased function of the whole adrenal cortex, but merely for the increased function of some cells, and which is accompanied in the responsive cells by changes interpreted as polyploidy. The morphological expression of this response is the familiar picture of the large cells with the characteristic large nucleus known as "adrenal cytomegaly".

One final point should be made. Recently Beckwith (1969) and others (Irving, 1967) described a combination of certain features which is now being known as "Beckwith's Syndrome" (Sotelo-Avila and Singer, 1970; Borit and Kosek, 1969). It is characterized by "omphalocele, macroglossia, adrenal cortical cytomegaly, hyperplasia of gonadal interstitial cells, renal medullary dysplasia and hyperplastic visceromegaly" (Sotelo-Avila and Singer, 1970). A glance at Table 3 of Sotelo-Avila and Singer's (1970) paper, as well as Irving's (1967) and Oppenheimer's (1970) remarks, show that the manifestations of this syndrome are rather variable. Here we are only concerned with its adrenal manifestations which are stated to be a) adrenal cortical cysts and b) adrenal cortical cytomegaly. Cystic changes in the adrenal cortex are seen so frequently in the fetus and newborn that their significance as part of any syndrome is more than questionable. Beckwith (1969) himself expressed reservations on this point. Adrenal cytomegaly, on the other hand, is considered by Sotelo-Avila and Singer (1970) to be "the most consistent endocrine gland abnormality in patients with the syndrome of hyperplastic fetal visceromegaly". Irving (1967) and Beckwith (1969) also view it as a component of the syndrome. If the hypothesis advanced here is correct, then the frequent occurrence of adrenal cytomegaly in patients with markedly enlarged viscera, including the pancreas, and other taxing anomalies should occasion no surprise. One may, however, well question whether the inclusion of this *non-specific reaction* as a "most consistent" feature of this, or any other syndrome for that matter, is justified.

Acknowledgements. Photographs were taken by the Audio-Visual Department of the Faculty of Medicine of Dalhousie University to whom our appreciation is expressed.

References

Aguilar, M. J., Kamoshita, S., Landing, B. H., Boder, E., Sedgwick, R. P.: Pathological observations in ataxia-telangiectasia. A report on five cases. J. Neuropath. exp. Neurol. **27**, 659–676 (1968).

Bachmann, R.: Die Nebenniere. In: Handbuch der mikroskopischen Anatomie des Menschen, ed.: W. Bargmann, vol. V, Die Nebenniere. Berlin-Göttingen-Heidelberg: Springer 1954.

Baker, de C., Wise, G., Mezger, M. L.: Cytomegalic adrenal hypoplasia in a $4^1/_2$-year-old boy. Amer. J. Dis. Child. **114**, 180–185 (1967).

Bayer, A., Židová, L., Dušek, J.: Zum Vorkommen von Riesenzellen in den Kindernebennieren. Zbl. allg. Path. path. Anat. **104**, 27–34 (1962).

Beatty, E., Hawes, C. R.: Cytomegaly of the adrenal gland. Amer. J. Dis. Child. **89**, 463–467 (1955).

Beckwith, B. J.: Macroglossia, omphalocele, adrenal cytomegaly, gigantism and hyperplastic visceromegaly. Birth Defects, Original Article Series, 5, No 2, 188–196, February (1969).

Borit, A., Kosek, J.: Cytomegaly of the adrenal cortex. Electron microscopy in Beckwith's syndrome. Arch. Path. 88, 58–64 (1969).

Bull, L. B.: The histological evidence of liver damage from pyrrolizidine alkaloids: Megalocytosis of the liver cells and inclusion globules. Aust. vet. J. **30**, No 1, 33–40, January (1954).

— Dick, A. T.: The chronic pathological effects on the liver of the rat of the pyrrolizidine alkaloids heliotrine, lasiocarpine and their n-oxides. J. Path. Bact. **78**, 482–502 (1959).

Chute, A. L., Robinson, G. C., Donahue, W. L.: Cushing's syndrome in children. J. Pediat. **34**, 20–39 (1949).

Cohen, R. B., Chapman, W. B., Castleman, B. J.: Hyperadrenocorticism (Cushing's disease). A study of surgically resected adrenal glands. Amer. J. Path. **35**, 5237–561 (1959).

Craig, J. M., and Landing, B. H.: Anaplastic cells of fetal adrenal cortex. Amer. J. clin. Path. **21**, 940–949 (1951).

Deamer, W. C., Silver, H. K.: Abnormalities in the secretion of the adrenal cortex during early life. J. Pediat. **37**, 490–516 (1950).

de Jong, B.: Polyploidy in animals. Bibliographia Genet. **92**, 111–228 (1957).

Dhom, G.: Die Nebennierenrinde im Kindesalter. Orthologie und Pathologie. Berlin-Heidelberg-New York: Springer 1965.

— Städtler, F.: Morphologische Befunde an der Nebennierenrinde bei primärem Aldosteronismus. Virchows Arch. Abt. A **345**, 176–199 (1968).

Diamond, I., Anderson, M., McCreadie, S. R.: Transplacental transmission of busulfan (myleran) in a mother with leukemia. Production of fetal malformation and cytomegaly. Pediat. **25**, 85–90 (1960).

Dietrich, A., Siegmund, H.: Die Nebenniere und das chromaffine System (Paraganglien, Karotisdrüse, Steißdrüse). In: Handbuch der speziellen pathologischen Anatomie und Histologie, ed.: F. Henke and O. Lubarsch, vol. 8, Drüsen mit innerer Sekretion. Berlin: Springer 1926.

Gabe, M., Arvy, L.: Gland cells. In: The cell, ed.: Brachet and Mirsky, vol. 5. New York: Academic Press 1961.

Garneau, R.: La cytomégalie des glandes surrénales. Maladie à virus. Ann. Anat. path. **2**, 104–108 (1957).

Gasser, C.: Hemolytic anemia in premature babies with spontaneous appearance of Heinz bodies. Quart. Rev. Pediat. **9**, No 1, 12–13 (1954).

Geitler, L.: In: Protoplasmatologie, Handbuch der Protoplasmaforschung, ed.: L. V. Heilbrunn and F. Weber, VI—C Wien: Springer 1953.

Guttman, P. H.: Addison's disease. A statistical analysis of 566 cases and a study of the pathology. Arch. Path. **10**, 742–785 (1930).

Harlem, O. K., Myhre, E.: Congenital adrenal hypoplasia. Amer. J. Dis. Child. **94**, 696–701 (1957).

Irving, I. M.: Exomphalos with macroglossia. A study of eleven cases. J. pediat. Surg. **2**, No 6, 499–507 (1967).

Jago, M. V.: The development of the hepatic megalocytosis of chronic pyrrolizidine alkaloid poisoning. Amer. J. Path. **56**, 405–522 (1969).

Kampmeier, O. F.: Giant epithelial cells of the human fetal adrenal. Anat. Rec. **37**, 95–102 (1927).

Kerenyi, N.: Congenital adrenal hypoplasia. Report of a case with extreme adrenal hypoplasia and neurohypophyseal aplasia, drawing attention to certain aspects of etiology and classification. Arch. Path. **71**, 336–343 (1961).

Kiefer, H.: Addisonsche Erkrankung infolge chronischer Nebennierendystrophie mit adenomartigen Regeneraten. Virchows Arch. path. Anat. **265**, 472–489 (1927).

Kissane, J. M., Smith, M. G.: Pathology of infancy and childhood. St. Louis: C. V. Mosby 1967.

Kracht, J., Tamm, J.: Bilaterale kleinknotige Adenomatosis der Nebennierenrinde bei Cushing-Syndrom. Virchows Arch. path. Anat. **333**, 1–9 (1960).

Landing, B. H.: Studies on the anatomy of the human adrenal cortex in various functional states. In: The human adrenal cortex. Ciba Foundation Colloquia on Endocrinology, vol. 8, p. 52–66 (1955). London: J. & A. Churchill Limited 1955.

Le Breton, E., Moule, Y.: Biochemistry and physiology of the cancer cell. In: The cell, ed.: Brachet and Mirsky, vol. 5. New York: Academic Press 1961.

Luse, S.: Fine structure of adrenal cortex. In: The adrenal cortex, ed.: Eisenstein, A. B. Boston: Little, Brown and Co. 1967.

MacMahon, H. E., Wagner, R., Weiner, D. B.: Acute adrenal insufficiency due to congenital defect. Amer. J. Dis. Child. **94**, 282–285 (1957).

Mitchell, R. G., Rhaney, K.: Congenital adrenal hypoplasia in siblings. Lancet **I**, 488–492 (1959).

Morison, J. E.: Foetal and neonatal pathology. Washington: Butterworths 1963.

Oppenheimer, E. H.: Adrenal cytomegaly studies by light and electron microscopy. Comparison with the adrenal in Beckwith's and virilism syndromes. Arch. Path. **90**, No 1, 57–64 (1970).

Pehlemann, R. W., Hanke, W.: Funktionsmorphologie des Interrenalorgans von *Rana temporaria* L. Z. Zellforsch. **89**, 281–302 (1968).

Potter, E. L.: Pathology of the fetus and the newborn, second ed. Chicago: Year Book Medical Publications Inc. 1961.

Rather, L. J.: The significance of nuclear size in physiological and pathological processes. Ergebn. allg. Path. path. Anat. **38**, 127–199 (1958).

Russfield, A. B.: The endocrine glands after bilateral adrenalectomy compared with those in spontaneous adrenal insufficiency. Cancer (Philad.) **8**, 523–537 (1955).

Schoental, R., Bensted, J. P. M.: Effects of whole body irradiation and of partial hepatectomy on the liver lesions induced in rats by a single dose of retrorsine, a pyrrolizidine (Senecio) alkaloid. Brit. J. Cancer **17**, 242–251 (1963).

— Magee, P. N.: Further observations on the subacute and chronic liver changes in rats after a single dose of various pyrrolizidine (Senecio) alkaloids. J. Path. Bact. **78**, 471–482 (1959).

Selye, H., Stone, H.: On the experimental morphology of the adrenal cortex. American Lecture Series. Springfield, Ill.: Charles C. Thomas 1950.

Sherman, F. E., Bass, L. W., Fetterman, G. H.: Congenital metastasizing adrenal cortical carcinoma associated with cytomegaly of the fetal adrenal cortex. Amer. J. clin. Path. **30**, 439–446 (1958).

Šikl, H.: Addison's disease due to congenital hypoplasia of adrenals in an infant aged 33 days. J. Path. Bact. **60**, 323–324 (1948).

Singer, D. B., Rudolph, A. J., Rosenberg, H. S., Rawls, W. E., Boniuk, M.: Pathology of the congenital rubella syndrome. J. Pediat. **71**, No 5, 665–675 (1967).

Sotelo-Avila, S., Singer, D. B.: Syndrome of hyperplastic fetal visceromegaly and neonatal hypoglycemia (Beckwith's syndrome). Pediatrics. **46**, No 2, 240–251 (1970).

Stark, E., Palkovits, M., Fachet, J., Hajtman, B.: Adrenocortical nuclear volume and adrenocortical function. Acta med. Acad. Sci. hung. **21**, 263–269 (1965).

Steinbiss, W.: Über eine eigenartige Degeneration der Nebennieren bei Addisonscher Krankheit. Virchows Arch. path. Anat. **262**, 286–297 (1926).

Stowens, D.: Pediatric pathology, second ed. Baltimore: Williams & Wilkins Co. 1966.

Therien, B., Mellinger, R. C., Caldwell, J. R., Howard, P. J.: Primary aldosteronism due to adrenal hyperplasia. Occurrence in a boy aged 10 years. Amer. J. Dis. Child. **98**, 90–99 (1959).

Tonutti, E., Bayer, J. M., Spiegelhoff, W.: Beitrag zur Kenntnis der Struktur der Nebennierenrinde beim connatalen adrenogenitalen Syndrom. Endokrinologie **40/41**, 310–335 (1960/61).

Uotila, U. U.: The early embryological development of the fetal and permanent adrenal cortex in man. Anat. Rec. **96**, 183–203 (1940).

Wägelin, C.: Schilddrüse. Drüsen mit innerer Sekretion. In: Handbuch der speziellen pathologischen Anatomie und Histologie, ed.: F. Henke and O. Lubarsch. Berlin: Springer 1926.

Wells, H. G.: Addison's disease with selective destruction of suprarenal cortex (suprarenal cortex atrophy). Arch. Path. **10**, 499–523 (1930).

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