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Morphological feedback effect on neurons of the nucl. arcuatus (sive infundibularis) and nucl. subventricularis hypothalami due to gonadal atrophy*

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Summary. We have correlated the light-microscopic features in the unmyelinated hypothalamus with gonadal atrophy in 15 women of 30–111 years of age and in 7 men between 29 and 82 years. In the postmenstrual cases there is a distinct concordance of gonadal atrophy and the manifestation of nucleolar changes (augmentation, multiplication and vacuolization) in many nerve cells of the arcuate and subventricular nuclei. In younger, still fertile women this nucleolar finding was seen only rarely and sporadically, was limited to the arcuate nucleus and was absent in the subventricular nucleus.

We interpret this nucleolar finding as a feedback effect. In man, too, this coincidence is obvious with age and in gonadal atrophy, with fewer nucleolar changes in old age than are seen in women. This difference is probably caused by a more rapid drop of the estradiol than of the testosterone level. – The hypertrophy of the subventricular nucleus (Sheehan and Kovacs) was also observed in our postmenstrual cases.

Key words: Feedback – Gonadal atrophy – Hypothalamus – Arcuate nucleus – Subventricular nucleus

Introduction

In a case of infantile haemochromatosis accompanied by hypogonadotropic hypogonadism due to siderosis of the hypophysis we recently found a marked increase, enlargement and vacuolization of neuronal nucleoli in the ncl. arcuatus hypothalami (Ule and Walter 1983). We interpreted these alterations as a morphological manifestation of a long-standing ineffective feedback effect caused by a complete loss of Leydig cells. As early as in 1966, Sheehan and Kovacs documented (but did not comment on) similar

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nucleolar changes in cases of postpartum hypopituitarism or during the menopause in their study dealing with "the hypertrophy of the nucleus subventricularis". We had the impression that our observation of nucleolar changes was comparable with the alterations in Sheehan's hypertrophy of the nucleus subventricularis. We examined 22 autopsy cases in order to correlate the light-microscopic feature in the unmyelinated hypothalamus to the gonadal findings.

Cases

A total of 22 cases was examined. They were classified in three groups, according to sex and age. Their clinical data and morphological findings are summarized in Tables 1–3: Table 1 contains the 10 cases of women from 62 to 111 years; in Table 2 the data of 5 women of the age of 30–56 years can be seen. In Table 3 the data of 7 men of 29–82 years are specified.

The *gonads* (and, if possible, the uterus) were weighed. Microscopical examination was carried out on routinely processed, HE-stained sections. The results are quoted in the Tables.

Table 1. Clinical and morphological findings of 10 women from 62 to 111 years

No.	Autopsy age	Clinical data	Ovary microscopy weight left/right	Uterus microscopy weight	Feedback-effect		
					Nucl. subventr.		Nucl. arcu-atus
					cent	lat. horn	
1	1043/82 68 years	Haemorrhagic necrotizing enterocolitis	No follicles; fibrous stroma Ø/4 g	Inactive endometrium; myomatous uterus 62 g	+	++	++
2	1051/82 69 years	Respiratory insufficiency	Senile involution; no follicles 4 g/3, 5 g	Inactive endometrium; dense stroma; cystic dilatation of the glands 62 g	+	+	++
3	1065/82 66 years	Chronic rheumatoid pancarditis; mitral insufficiency	Senile involution; no follicles 3 g/3 g	Inactive endometrium; dense stroma; cystic dilatation of the glands 20 g	(+)	(+)	(+)−+
4	27/83 81 years	Hypertonia; cardiac insufficiency	Ovarian cyst; senile involution; no follicles 4 g/2 g	Inactive endometrium; dense stroma 13 g	+	++	++

Table 1 (continued)

No.	Autopsy age	Clinical data	Ovary microscopy weight left/right	Uterus microscopy weight	Feedback-effect		
					Nucl. subventr.		Nucl. arcu-atus
					cent	lat. horn	
5	30/83 62 years	Phenacetin – induced renal insufficiency	No follicles; senile involution 2 g/1,5 g	Proliferated endometrium; glandular-cystic hyperplasia 48 g	+	+	++
6	51/83 87 years	Marasmus	No follicles, some corpora albicantia; dense stroma; endometriosis externa 1,8 g/1,8 g	Inactive endometrium; dense stroma; cystic dilatation of the glands 58 g	++	++	++
7	55/83 80 years	Bodily trauma; cardiac insufficiency	n.d.[a]	n.d.	(+)	++	++
8	95 /83 92 years	Activated tuberculosis; cardiac insufficiency	No follicles; fibrous stroma, some corpora albicantia 1 g/1 g	Inactive endometrium; dense stroma 34 g	+	+	++
9	106/83 63 years	Surgery of breast cancer 5 years ptd[b] luteum hormone- and chemoherapy; marasmus	Some follicles; a few corpora albicantia; dense stroma 3 g/3 g	Myomatous uterus; inactive endometrium, dense stroma 94 g	(+)	(+)	++
10	123/79 111 years	Congophilic angiopathy of the parenchymatous organs and the brain; senile marasmus; cardiac insufficiency	Fibrous stroma; corpora albicantia n.d.	n.d.	+	+	++

[a] not determined [b] prior to death

Table 2. Clinical and morphological findings of 5 women of the age of 30–56 years

No.	Autopsy age	Clinical data	Ovary microscopy weight left/right	Uterus microscopy weight	Feedback-effect		
					Nucl. subventr.		Nucl. arcu-atus
					cent	lat. horn	
11	1048/82 51 years	Werlhof's disease; pulmonary embolism; pneumonia	Fibrous stroma; no follicles; corpora albicantia 4 g/4 g	n.d.[a] 42 g	(+)	(+)	(+)
12	4/83 39 years	Breast cancer with abundant metastases 5 years ptd; surgical and hormonal therapy; diencephalic metastases 6 mths ptd;[b] radiation to the brain	Bilateral ovariectomy 3 years ptd	Inactive endometrium; weakly proliferated glands 45 g	∅	∅	(+)
13	71/83 56 years	Ovarian cancer with metastases; urinary sepsis	Bilateral ovariectomy 5 years ptd	Exstirpation	∅	(+)	+
14	92/83 34 years	Atrophic myotonia; externe adiposis Morgagni-Stewart-Morel syndrome	Primary follicles 6 g/7,5 g	Proliferated endometrium; endometriosis interna; myomatous uterus 72 g	∅	∅	(+)
15	199/83 30 years	Cerebellar arteriovenous angioma; cerebellar hemorrhage	Numerous follicles of variant differentiation; dense stroma 9g/8 g	Low proliferation of the endometrium (genital cycle dependent) unremarkable myometrium 82 g	∅	∅	(+)

[a] not determined [b] prior to death

Table 3. Clinical and morphological findings of 7 men of 29–82 years

No.	Autopsy age	Clinical data	Testicle microscopy weight left/right	Feedback-effect		
				Nucl. subventr.		Nucl. arcu-atus
				cent	lat. horn	
16	1064/82 65 years	Haemolytic anemia; secondary hemosiderosis; cardiac insufficiency of the right ventricle	Thickening of the basement membrane; low spermatogenesis; mild interstitial fibrosis; some Leydig cells 26 g/23 g	(+)	(+)	+
17	52/83 31 years	Brain injury and traumatic myelomalacia; cross section palsy 11 years ptd;[b] secondary amyloidosis	Thickening of the basement membrane; missing spermatogenesis; mild interstitial fibrosis; numerous Leydig cells 15 g/10 g	(+)	(+)	(+)
18	59/83 80 years	Diabetes mellitus; arteriosclerosis; cardiac insufficiency	Thickening of the basement membrane; nearly complete loss of spermatogenesis; marked interstitial fibrosis; no Leydig cells 24 g/22 g	(+)		+
19	75/83 82 years	Diabetes mellitus; hypertonia; meta-static cancer of the tongue	n.d./n.d.[a] 29 g/32 g	(+)	(+)	+
20	119/83 74 years	Prostatectomy and bilateral orchiectomy 5 years ptd due the cancer; gynecomastia; female pubic hair; multiple metastases	bilateral orchiectomy	(+)		+
21	188/83 47 years	Chronic alcoholism; pancreatitis, hepatic steatosis; diabetes mellitus; pneumonia	Active spermatogenesis; mild interstitial fibrosis; distinctly reduced number of Leydig cells 10 g/12 g	(+)	(+)	+ to ++
22	213/83 29 years	Multiple traumatic injury	Active spermatogenesis; mild interstitial fibrosis; numerous Leydig cells 25 g/28 g			(+)

[a] not determined [b] prior to death

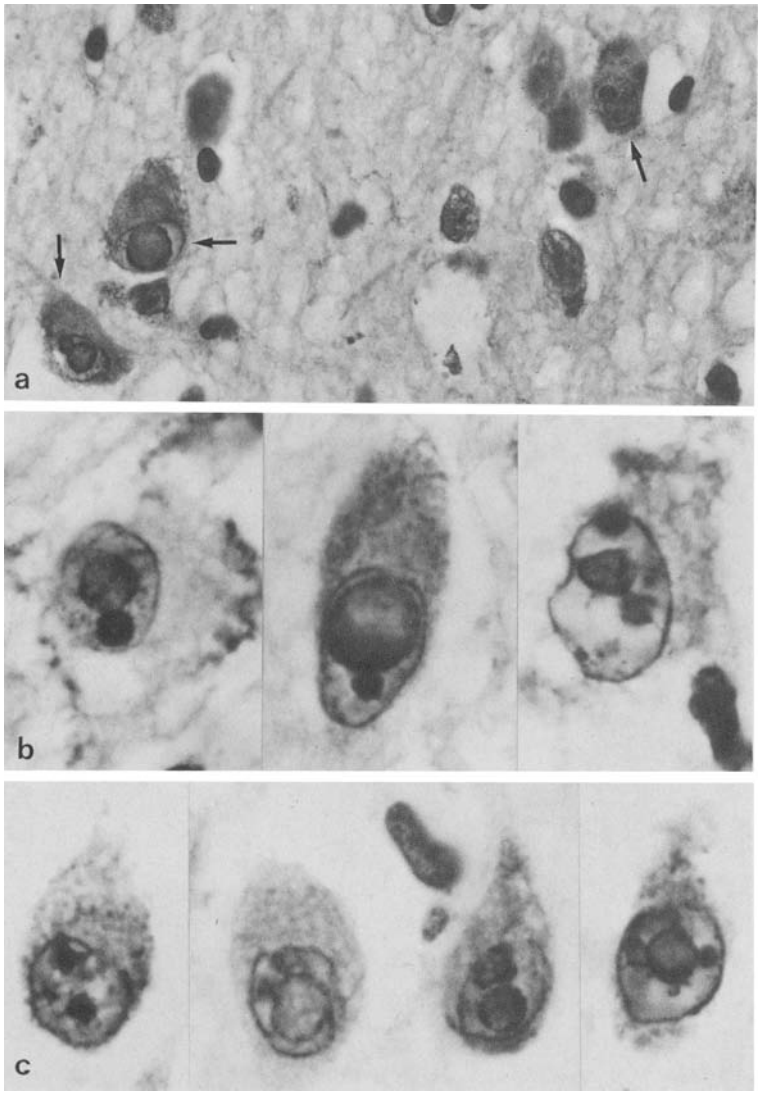

Fig. 1 a–c. Nuclear-nucleolar changes in neurons of the nucl. arcuatus in postmenstrual cases. **a** Case no. 9. Nissl, ×600, **b** Case no. 7. Nissl, ×1,500, **c** Case no. 6. Nissl, ×1,500

Serial sections (6 μm; distance 150–200 μm) were performed on formalin-fixed and paraffin-enbedded *unmyelinated hypothalamus* and stained according to the Nissl' procedure. The neurons of the infundibular and subventricular nuclei were selectively examined with respect to nuclear activation. Special attention was given to enlarge, multiplied and vacuolized nucleoli (Figs. 1 and 2). Intranuclear membrane foldings mimicked the feature of vacuolization, and for this reason differentiation was often difficult. Occasionally, nucleolar vacuolization might be due to intranuclear cytoplasmic invagination. As these invaginations, however, express activated nuclear metabolism, they were regarded as positive findings. Other structural changes due to membrane folding were not taken into consideration.

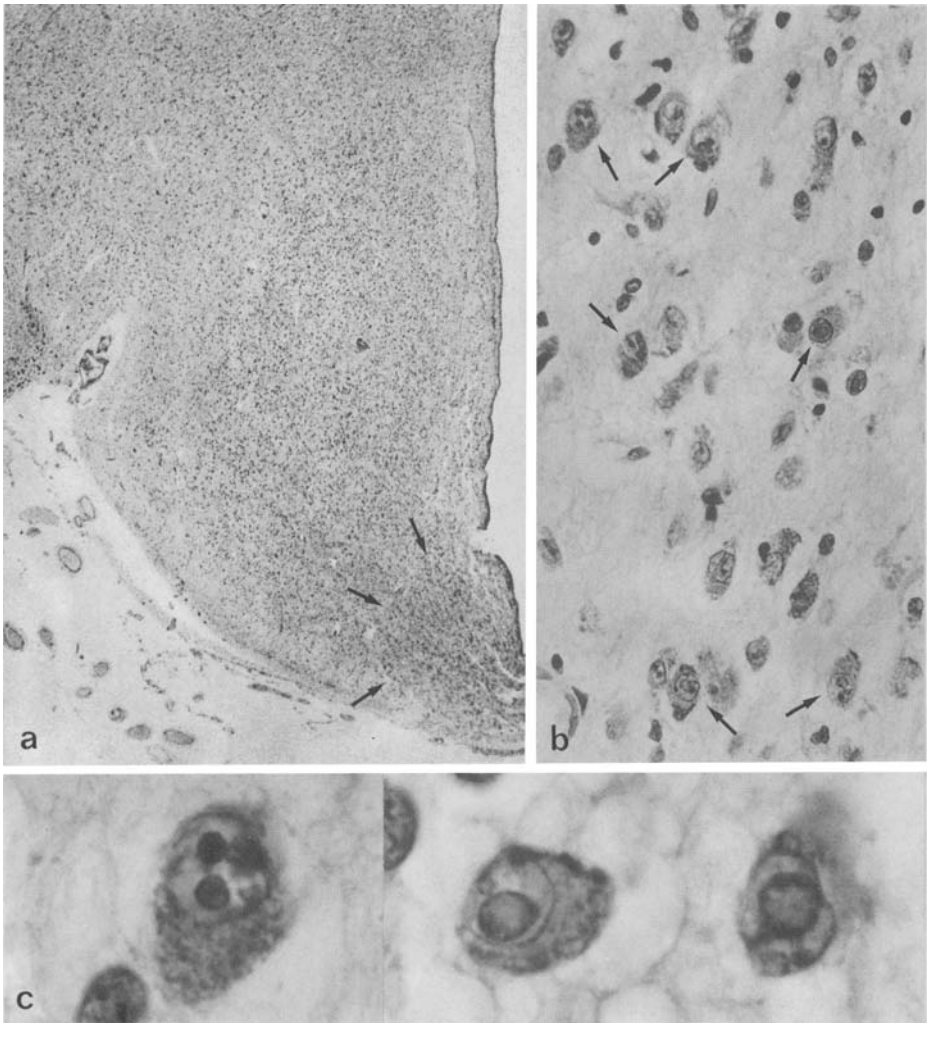

Fig. 2a–c. Feedback effect in a 111-year-old woman (case no. 10). **a** Hypothalamus with nucl. arcuatus. – broderline to the dorso-lateral adjoining nucl. ventro-medialis. Nissl, ×40. **b** Detail of **a** with many alterated nucleoli – Nissl, ×380. **c** Alterated nucleoli. Nissl, ×1,500

A gradual differentiation between isolated, numerous or very many nuclear or nucleolar changes was made and symbolized in the Tables as (+), + and + +, respectively. The distribution of the alterations in the two nuclei was more often focal than diffuse. In the infundibular nucleus, nuclear-nucleolar changes were more marked at the periphery. Large neurons as well as neurons of types 3 and 4 in Sheehan and Kovacs' (1966) scale were involved, even though the large neurons were more affected.

It must be emphasized, however, that the most severe nuclear changes (graded as + +) of the present cases never reached the quantitative extent of the case quoted above of a young man suffering from infantile haemochromatosis accompanied by hypogonadotropic hypogonadism. This difference is obviously explained by greater susceptibility of this feedback mechanism in infancy.

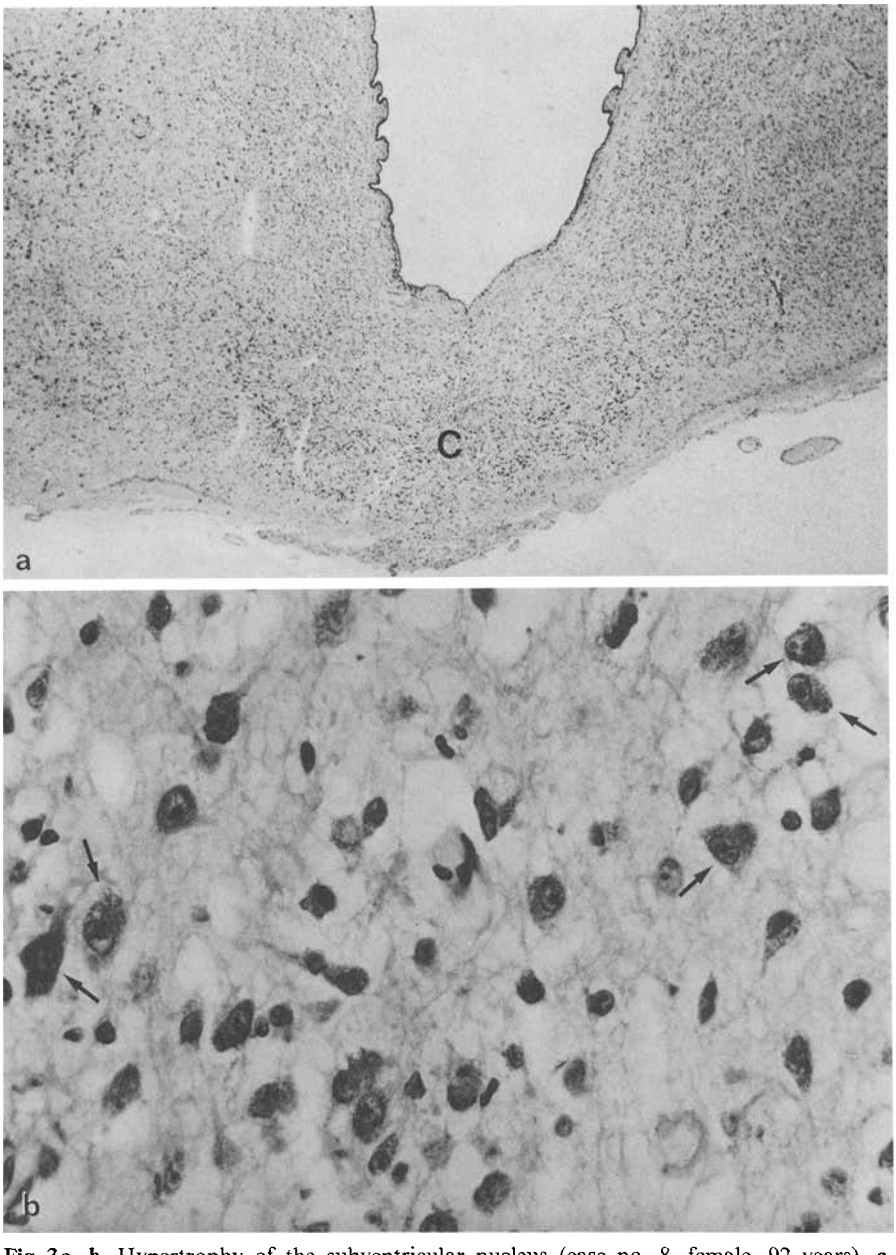

Fig. 3a, b. Hypertrophy of the subventricular nucleus (case no. 8, female, 92 years). **a** In the parvocellular part "cent" (*C*) many large neurons are present. They are different from the neurons of the posteromedial tuberal nucleus (Sheehan and Kovacs 1966) and of the tuberomammilar nucleus, respectively. Nissl, ×15. **b** Detail of **a** Activated nucleoli. Nissl, ×380

Discussion

Except for case 3 in Table 1, all women older than 62 showed an increased number of enlarged and vacuolated nucleoli in the above-mentioned regions of the unmyelinated hypothalamus. The alterations are accentuated in the arcuate nucleus and less marked in the region forming the subventricular nucleus. In other hypothalamic nuclei the alterations described were isolated and rare.

When the nuclear changes were distinct, the gonads showed progressive involution with a loss of follicles. This finding express a decreased production of sexual hormones. The weight of the uterus ranged from 13 to 94 g. The endometrium was inactive.

As far as the postmenstrual cases are concerned, it seems that an augmented size and number of the nucleoli and their vacuolization indicates an ineffective and so-to-speak "frozen to death" activation of the arcuate and subventricular nucleus. It is regarded as an ineffective feedback regulation due to hormonal insufficiency in gonadal atrophy. It is very remarkable that the feedback mechanism is still active, independent of the patients' advanced age (see Fig. 2).

It is not clear why the nucleolar vacuolization in the hypothalamic neurons of case 3 was moderate despite of the fact that progressive ovarian involution and an atrophic uterus (20 g) were noted. The possible interference of hormonal therapy could not be determined, but must be kept in mind. In this context, it might be of interest that Sheehan (1971) recorded a similar case of extreme atrophy of the uterus which was not correlated with the "hypertrophy of subventricular nucleus."

If the above-mentioned interpretation of a positive feedback effect is correct, nucleolar changes in the hypothalamic nuclei should not or should at best be existent sporadically in younger, still fertile women. In order to corroborate this hypothesis, 6 women no older than 56 years were examined (Table 2). In fact, nucleolar changes in a young woman of 30 suddenly dying due to cerebellar haemorrhage (case 15, Table 2) were absent in the subventricular nucleus and rare in the arcuate nucleus. This is also correct in the case of a 34 years-old woman (case 14) and a woman of 39 (case 12). In the latter case, a distinct increase of nucleolar alteration was expected because of a bilateral ovariectomy 3 years prior to death. Hormonal therapy after surgery and radiation of the diencephalic metastases might explain this unexpected result. At autopsy calcified necroses as well as large carcinomatous infiltrates were detected in the thalamus and dorsal parts of the hypothalamus.

As far as sex-related differences in the course of gonadal involution are concerned, hypothalamic alterations in men are of special interest. A 29-year-old man who died after massive trauma (case 22, Table 3) showed a age-related gonadal function, no nucleolar alterations in the subventricular nucleus and only isolated changes in the arcuate nucleus. In another case of a 31-year-old man (case 17, Table 3), hypothalamic nucleolar alterations were expected but rare, despite gonadal insufficiency and inguinal testis

on the right side caused by traumatic paraplegia 11 years prior to death. Nonexistent spermatogenesis, but persistent Leydig cells accompanied by only mild interstitial fibrosis explain the unexpected result. In addition, a small cystic area with glial proliferation in the cinerian tuber reaching the borderline between arcuate nucleus and ventro-medial nucleus was noted and correlated to extensive trauma 11 years prior to death with distinct fronto-basal and temporal contusions. Another case of a 47-year-old man suffering from chronic alcoholism (case 21, Table 3) is a unique case in this group. Nucleolar changes in the arcuate nucleus are numerous. Even though the spermatogenesis was active and the Leydig cells are only reduced in number, the gonadal atrophy, the enhanced conversion of testosterone to estradiol and the reduction of free plasma testosterone by an increase of sex-hormone-binding globulin in liver diseases (Gersthövel 1979) may explain this surprising fact.[1] Ethylism also led to mammallian pseudomalacia in the hypothalamus (Neubuerger 1931), which is regarded as a form of Wernicke encephalopathy. In the other cases of this group of males whose ages ranges from 65 to 82 years, the arcuate nucleus demonstrates a certain increase in nucleolar changes. Quantitatively, however, they do not reach such a great extent as in the group of senile women.

In this context we have to comment on the term "hypertrophy of the subventricular nucleus". Sheehan and Kovacs (1966) and Sheehan (1967 and 1971) emphasized the enlargement of individual neurons in this hypothalamic area. In younger people, this nucleus consists of poorly differentiated, isomorphous and small neurons. Subsequent publications (Christ 1969; Hayward 1977) do not mention the subventricular nucleus. Sheehan and Kovacs (1966) stress the opinion that this region might not be identical with the infundibular or arcuate nucleus. This statement was questioned by Oksche (1971) and Orthner (1971). The subventricular nucleus contains a considerable number of large neurons in postpartual hypopituitarism and – in accordance with our findings – in the postmenstral period.

Morphometric analyses have still to be done. However, the 22 cases studied, where alternating cuts were performed (at intervals of 150 to maximally 200 μm), allow us to make the following valid statement: the formation of the median group of the parvocellular hypothalamic nuclei (Christ 1969) demonstrates a considerable variability which is not exclusively related to age (childhood, puberty, fertile age, senile involution). The borders of the parvocellular regions are not well defined. Their cytoarchitecture is not completely homogenous and isomorphous. This may be in part explained by the events in human development where the phenomenona of the rotation of the basal cortex, the intussusception of the diencephalon and the rotation of the neurohypophyseal axis in the sense of Spatz (Diepen 1962) are accompanied and followed by remarkable morphokinetic shifts, particularly in the hypothalamus (Diepen 1962). Additionally the level of sectioning deter-

1 After ending of this series we have found the same condition in a recent case of gin-drinkers liver (SNr.: 294/83): atrophy of the testicles, persistent Leydig cells and a positive feedback effect in the arcuate nucleus.

mines the expansion of the nuclei (Christ 1951 and 1969). This explains the differences in definition and demarcation of the various centres in this area. According to our experience both nuclei were often a coherent complex. The caudal part is called subventricular nucleus and the rostral part is defined as infundibular nucleus. In the caudal region the neurons are small and poorly differentiated until midlife whereas the rostral parts demonstrate a circumscribed and earlier differentiation. Only in later life did a part of the aforementioned parvocellular neurons become enlarged. In addition, this process was accelerated in postmenstral patients. This phenomenon, which was called "hypertrophy" by Sheehan and Kovacs (1966), was also observed in our postmenstral cases. Even though in our series the continuity of the subventricular to the infundibular nucleus was occasionally interrupted, the following factor speaks in favor of a unity between the caudal and the rostral part, namely, the accumulation of nuclear and nucleolar alterations which is considered a feedback phenomenon due to gonadal atrophy is found both in the rostral (infundibular nucleus) and in the caudal (subventricular nucleus) part. In other regions of the unmyelinated hypothalamus accumulation of the nucleolar changes was not noted[2].

The results of our study confirm our interpretation concerning the feedback character of the nuclear and nucleolar changes described in the arcuate and subventricular nucleus. It must be emphasized that they do not indicate a hypothalamus-induced endocrinopathy. Obviously, these alterations can be present in fertile individuals, even though sparsely. Wasting, however, is strictly correlated to gonadal atrophy, i.e. a loss of Leydig cells in men and of granulosa cells in women. After a certain interval, surgical exstirpation of the gonads has the same effect. If a positive feedback effect did not occur or was weakly developed, additional factors like hormonal therapy, cerebral radiation may be responsible, amoung other factors. The accentuation in senile women found less intensively in men is probably caused by a more rapid drop of estradiol than testosterone level.

Finally, many questions remain to be answered, for example:

- the effect of the long-standing "frozen to death" activated nucleoli with regard to the cellular metabolism;
- the fact that only a part of the neurons in these nuclei are involved;
- the fundamental (patho-) physiological mechanism leading to both nucleolar activation and cellular differentiation or hypertrophy, and
- the time necessary for their development.

Further research in this field seems to be promising. Recent experimental studies using immunocytochemical, electron-microscopic and morphometric methods (Hoffman et al. 1978; Merchenthaler et al. 1980; Nozaki et al.

2 Later on, we have sporadically seen these nuclear-nucleolar findings in the lamina terminalis, too. However, they are more numerous in the arcuate and subventricular nuclei which conspiciously play a dominant role with respect to this feedback mechanism. Their manifestation is a reference to a persistent response of these cells even in the senium, visible in the elevated gonadotropic hormone levels in the postmenstrua and senium, too (Ule, G.: Zur Frage des Feedback-Effektes an Neuronen der Lamina terminalis bei Involution der Keimdrüsen. Der Pathologe: in press).

1980; Zimmermann et al. 1982) have yielded contradictory findings and have produced no concept of the significance of the arcuate nucleus.

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