Sex steroid hormones in the pathogenesis of chronic subdural haematoma

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Summary

In order to verify whether hormonal factors could be involved in the pathogenesis of Chronic Subdural Haematoma (CSDH), based on clinical and epidemiological demonstration of higher incidence of this disease in male patients and particularly in those with high urinary estrogen values, Estradiol (ER) and Progesterone (PR) Receptors were studied in the Haematoma External Membrane (HEM) in 18 male and 7 female CSDH patients. The observed higher incidence of ER and PR in male rather than in female patients (73% vs 27% and 72% vs 28% for male and female patients respectively), and the higher concentration of ER in the HEM of male rather then female patients (55 ± 15 S. E. vs 13 ± 7 S.E. fmol/mg protein) suggest that this pathological process, which affects individuals whose gonadal activity is quiescent, is mainly dependent upon hormonal local effect played by estrogen compounds on the HEM of the male patients. In this sex, in fact, whose tissues are not usually adapted to an estrogen action, the effect of estrogens on a responsive tissue such as the newly-vascularized HEM could lead to an increased formation of tissue Plasminogen Activator (t-PA), a compound that, escaping into the subdural collection, could maintain a local hyperfibrinolysis with formation of Fibrinogen Degradation Products (FDP). Therefore local hyperfibrinolysis enhanced by steroid hormones and the subsequent CSDH may perhaps be influenced by the prophylactic or adjuvant treatment with inhibitors either of the aromatase activity or of the estrogen action at receptor level.

Key-Words

Subdural haematoma – Steroids – Estradiol – Progesterone – t-PA – Hyperfibrinolysis

Sexualhormone in der Pathogenese des chronischen subduralen Hämatoms

Um nachzuweisen, daß hormonale Faktoren bei der Pathogenese des chronischen subduralen Hämatoms (CSDH) eine Rolle spielen – zu vermuten durch den klinischen und epidemiologischen Hinweis der höheren Inzidenz bei Männern, insbesondere bei denen mit höheren Östrogen-Werten im Urin – wurden bei 18 männlichen und 7 weiblichen Patienten mit CSDH die Östrogen (ER)- und die Progesteron (PR)-Rezeptoren in der äußeren Hämatom-Kapsel (HEM) bestimmt. Die gefundene höhere Rate von ER und PR bei männlichen gegenüber den weiblichen Patienten (73% vs 27% und 72% vs 28%) und die höhere Konzentration von ER in der HEM (55 ± 15 vs 13 ± 7 fmol/mg Protein) lassen vermuten, daß dieser pathologische Prozeß, der Individuen mit ruhender Gonadenfunktion befällt, hauptsächlich von einem lokalen hormonalen Effekt östrogener Verbindungen in der HEM der männlichen Patienten abhängig ist. Bei Männern ist das Gewebe gewöhnlich nicht an die Östrogen-Effekte adaptiert und bei der Reaktion von Gewebe wie der neuvaskularisierten HEM kann es zur erhöhten Bildung von Plasminogen-Faktor (t-PA) kommen. Diese Substanz kann beim Übertritt in das Hämatom zu einer lokalen Hyperfibrinolyse mit gleichzeitiger Erniedrigung von Fibrinogenen (FDP) führen. Daher kann die lokale Hyperfibrinolyse – verstärkt durch Steroid-Hormone und nachfolgendes CSDH – vielleicht durch die prophylaktische oder zusätzliche Behandlung mit Antiöstrogenen auf dem Rezeptorenniveau oder mit Antiaromatasesubstanzen.

Chronic subdural haematoma (CSDH) has a higher incidence in male than in female patients (18), affects male patients with urinary estrogens higher than those found in normal subjects (28); it is more common during the 7th decade of life (6, 18, 19) i.e. during a period of quiescent gonadal activity.

Inside the newly-formed haematoma external membrane (HEM) the ectatic capillaries (10, 15, 26, 32) are believed to be similar to the vascular network of skin in alcoholic cirrhotic patients with elevated estrogen levels (28). Furthermore the fibrinolytic activity and tissue plasminogen activator (t-PA) of the haematoma fluid and the HEM respectively can be compared to those of menstrual bleeding and uterine mucosa (13).

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Table I Concentration of Estrogen receptors (ER) and Progesterone receptors (PR) in Normal Dura Mater.

Patient	Sex	Age (Yr)	ERc (fmol/mg protein)	ERn (fmol/mg DNA)	PRc (fmol/mg protein)
1	f	65	33	neg.	43
2	f	32	n.d.	56	75
3	m	41	12	76	neg.
4	m	54	50	150	9

C = cytosol; N = nuclear; N.D. = not determined; neg. = negative

Table 2 Presence of Estrogen receptors (ER) and Progesterone Receptors (PR) in the cytosol and nuclei of HEM of CSDH patients.

	total	male	female
ER_C	15/20 (75%)	11/15 (73%)	4/15 (27%)
PR_C	18/23 (78%)	13/18 (72%)	5/18 (28%)
ER_N	15/22 (68%)	10/15 (67%)	5/15 (33%)
PR_N	5/8 (63%)	3/5 (60%)	2/5 (40%)

Table 3 Concentration of cytosol ER and PR in the HEM of CSDH patients.

	male	female
	(fmol/mg of protein ± S.E.)	
ER_C	55 ± 15	13 ± 7
PR_C	37 ± 7	35 ± 21

S.E. = standard error

In attempt to advancing our knowledge in the pathogenesis of CSDH, estrogen receptor (ER) and progesterone receptor (PR) were measured in the HEM of CSDH.

Materials and Methods

The HEM was removed at surgery under local anesthesia from twentyfive CSDH patients, eighteen male and seven female (mean ages 69 ± 7 years old and 71 ± 13 years old for male and female respectively). Specimens of dura mater lying above the CSDH were simultaneously removed from four patients (3 male and 1 female patient). Four dura mater specimens from glioma patients (2 male and 2 female) were also studied for comparison. Specimens of HEM and dura mater were stored at −70 °C until processed. Cytosol fractions and nuclear extracts were then obtained with differential centrifugation. Methodological details have been published elsewhere (8, 9).

Total cytosol ER (ER_c) was measured in 20 specimens and total nuclear ER (ER_n) in 22 specimens of HEM using the method of *Sica* et al. (27). Total cytosol PR (PR_c) was measured in 23 specimens and total nuclear PR (PR_n) in 8 specimens of HEM using low and high salt concentration of sodium molybdate respectively (23). Only single saturation dose analysis could be performed on account of the small size (80–100 mg) of available HEM fragments.

After separation of free and bound ligands, radioactivity was measured on a Packard 460 CD scintillation spectrometer, protein concentration by the *Bradford* method (5) and DNA content by *Burton* procedure (7).

The threshold values were fixed at 3 fentomoles (fmol) per milligram (mg) of protein for cytosol receptors and at 50 fmol/mg DNA for nuclear receptors.

Results

Steroid receptors in normal dura mater

The concentration of ER_c and ER_n were similar in the normal dura mater taken from male and female patients. The PR_c content, on the contrary, was higher in normal dura mater of the women than in that of the men (Table 1).

Steroid receptors in HEM

Incidence of cytosol and nuclear steroid receptors

The incidence of ER_c was 75% (15/20) and that of PR_c 78% (18/23). Both ER_c and PR_c in HEM were more frequent in male rather than in female patients (73% vs 27%; 72% vs 28% for ER_c and PR_c respectively). The incidence of ER_n was 68% (15/22) and that of PR_n 63% (5/8). ER_n was more frequent in the specimens taken from male rather than from female patients (67% vs 33%) (Table 2).

The five PR_n were found in three male and two female patients.

Concentration of cytosol steroid receptors

The concentration of ER_c in the HEM of the male patients showed mean value of 55 ± 15 S. E. fmol/mg protein with a range from 3 to 104 fmol/mg protein. Therefore the ER_c content was slightly higher than that found in the normal dura mater taken from male patients (Table 3).

Furthermore the ER_c values in two specimens of dura mater taken from CSDH patients were different from those found in the HEM of the corresponding patients.

The ER_c content in the HEM of the four female patients (13 ± 7 S. E. fmol/mg protein) was slightly lower than that found in the normal dura mater taken from a female patient (33 fmol/mg protein). Furthermore in one female patient the amount of ER_c in the HEM was found comparable to that measured in the dura mater laying above the CSDH.

The concentration of PR_c in the HEM of the male patients showed a mean value of 37 ± 7 S. E. fmol/mg protein with range from 5 to 85 fmol/mg protein. Therefore, as for ER_c, also the mean value of PR_c in HEM was higher than that found in normal dura mater taken from male patients (Table 3). Two out of the three dura mater specimens lying above the CSDH had PR_c values different from those found in HEM of the corresponding patients.

The concentration of PR_c in the HEM of the female patients showed a mean value of 35 ± 21 S. E. fmol/mg protein with a range from 9 to 91 fmol/mg protein. Therefore the ER_c value in HEM was lower than that found in normal dura mater taken from female patients.

Concentration of nuclear steroid receptor

The mean value of ER_n content measured in the HEM of 10 male patients was 77 fmol/mg DNA, a value comparable with that obtained in 2 normal dura mater, whilst ER_n was found only in 1 out of the 3 samples of dura mater laying above CSDH. However the data obtained in these 3 dura mater samples were concordant with those measured in the 3 HEM specimens of the corresponding patients. Furthermore it is important to underline that negative results were obtained in 7 out of the 22 specimens examined: 86% (6/7) of this ER_n negative HEM specimens were removed from male patients.

The mean value of ER_n content measured in the HEM of 5 female patients was 99 fmol/mg DNA, higher than that found in one normal dura mater specimen. The dura mater removed from one female CSDH patient was ER_n negative as the corresponding HEM taken from the same patient.

The measurement of PR_n was performed only in 8 HEM specimens, five of which were provided with nuclear PR. The PR_n measured in the dura mater specimens taken from two CSDH patients was found negative as the PR_n in the HEM of the corresponding patients.

The comparison of the receptor content in the sample of HEM of male and female CSDH patients showed that the values of cytosol ER were higher in HEM of male than female patients (Table 3); on the contrary, those of nuclear ER were higher in HEM of female than male patients. The values of cytosol PR in the HEM of both sexes were similar.

Discussion

It is well accepted that the primary etiological factor responsible for CSDH is the tearing of cortico-dural bridging veins (31). The bleeding usually occurs in either a virtual space (25) or a cellular compartment between the dura and the arachnoid membranes (10), mostly in elderly people because of concomitant cortical atrophy (6).

This extravascular blood in some cases can be easily reabsorbed, whilst in other cases is responsible for a fibroblastic reaction leading to the formation of a highly vascularized subdural membrane. Experimental evidence is in favour of a stimulating action on the inner surface of the dura mater by the blood fibrin (1). This neomembrane may be found unchanged at autopsy, whilst in some patients a local hyperfibrinolysis that interferes with haemostatic mechanisms, is responsible for the formation of a bloody collection between the dura and the arachnoid membranes: the CSDH (14, 17).

It is interesting to remind also the reported formation of a meningioma (2) or of a sarcoma (16) inside the HEM of CSDH patients, as well as the association of a meningioma with a newly-formed subdural membrane without a collection of blood (22).

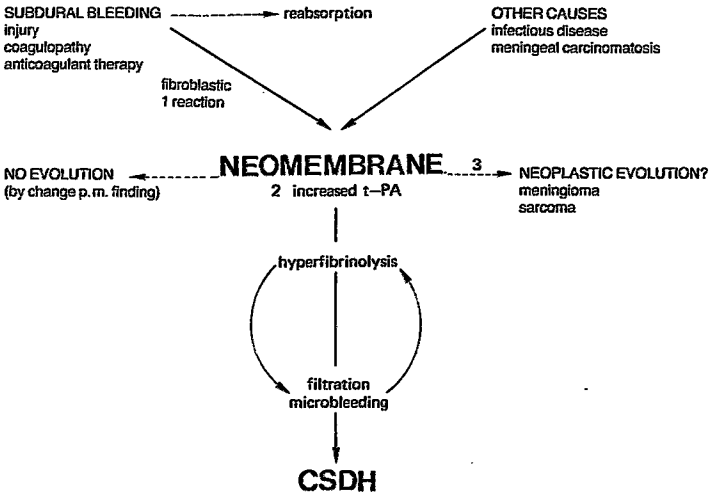

Fig. 1 Sites of possible steroid hormones action in the pathogenesis of CSDH.

Studies performed since 1970 demonstrated that the pathogenetic mechanism of a CSDH is represented by the high content of tissue plasminogen activator (t-PA) in the ectatic capillaries of HEM, which exudes into the fluid collection, transforming plasminogen into plasmin. This active compound breaks down fibrin with formation of fibrinogen degradation products (FDP) and induces continuous microbleeding (13, 30).

The t-PA has the highest value in the HEM (followed by the dura mater and arachnoid), whilst the haematoma content lacks fibrinogen and plasminogen in spite of an extraordinary large amount of FDP. This situation is similar to that found in the menstrual blood and uterine mucosa (the former being rich in FDP and the latter rich in t-PA) which are affected by hormonal modifications during the menstrual cycle. In this respect it has been demonstrated that the production of t-PA in vitro is stimulated by physiological levels of estrogens and further potentiated by progesterone (21). It is known that these steroids act through their own receptors.

For this reason, based on the above mentioned epidemiological and clinical data, we decided to seek for the presence of ER and PR in the HEM of CSDH. The results of the present investigation support the hypothesized pathogenetic role played by the steroid hormones in the formation of CSDH.

The presence of both ER and PR in the cytosol and nuclear fractions of the HEM studied is accompained by a preferential distribution of steroid receptors in the HEM of male rather than female CSDH patients (ER_c 73% vs 27%; PR_c 72% vs 28% for male and female respectively). Furthermore the higher content of cytosol ER in the HEM of male rather than female CSDH patients (55 vs 13 fmol/mg protein) could perhaps account for the sex related higher incidence of CSDH in male rather than female patients.

The prevalence of steroid receptors and the higher content of ER in the male rather than female patients cannot be explained on the basis of the stimulation of recep-

tors by circulating hormones. CSDH, in fact, is a disease of elderly patients, whose gonadal activity is usually quiescent. However it must be taken into consideration that some enzyme activities could be responsible for the formation of active metabolites from adrenal and gonadal precursors.

This concept is supported by the increased aromatase activity reported in elderly men, which can be responsible for the formation of estrogens from testosterone and androstenedione. It is known that this enzyme activity plays an important role in the pathogenesis of human benign prostatic hyperplasia, which affects elderly men. Therefore the reported higher incidence and concentration of ER in the HEM of male CSDH patients could perhaps be dependent upon the conversion of circulating androgens into estrogens by the high aromatase activity in the male patients.

Furthermore the tissue response to hormonal stimulation and mainly to estrogens is more pronounced in male than in female vascular network, not because of a lack of progesterone and PR, which appear to be uniformly distributed in the two sexes, but because of the different thresholds of response. In women, in fact, vessels are usually adapted to high estrogen values. This abnormal tissue response to estrogens in male patients may account for the values of both ER_c and PR_c of male CSDH patients higher than those measured in the normal dura mater taken from male glioma patients usually of younger age than the CSDH patients. On the contrary, in the female patients whose tissue are usually adapted to estrogen stimulation, the values of both ER_c and PR_c in the HEM are lower than those measured in the normal dura mater taken from female younger glioma patients.

As far as the role played by steroid hormones on the pathogenetic mechanism of CSDH (Fig. 1), estrogens could be involved in the formation of the newly-vascularized external membrane, for their action on the neoangiogenic process (12) and on the mesenchymal cells (24). In this respect it is important to note that other steroid hormones such as glucocorticoids are known to be related to fibroblastic reactions (29). In addition to this direct effect, steroid action could also be dependent upon a receptor mediated mechanism based on genomic expression of hormonal stimulation. This action could lead to the proliferation of the innermost layer of the dura mater and particularly to the mesenchymal proliferation of the HEM.

Moreover ER and PR have been reported in cranial and spinal meningiomas (4, 9, 20). The association of a meningeal tumor with a subdural haematoma (2, 16) and that of a meningioma with a subdural neomembrane without bloody collection (22) have been reported too. It is tempting to speculate a possible influence of steroid hormones on both the lesions.

Furthermore the effects of estrogens and progesterone are more likely to be predominant in maintaining local hyperfibrinolysis through their action on the levels of t-PA. In this respect the presence of ER and PR in the HEM can be considered in favour of an hormonal action on this tissue: the steroid hormones could play at level of the HEM the same action on t-PA they play at endometrial mucosa level.

The receptor mediated effects of steroids could also explain the higher values of t-PA found in the HEM than in the dura mater on account of the higher content of steroid receptors in the former rather than in the latter. The hormonal effect on t-PA, although not the primary event leading to the CSDH, can be considered as one of the main factors responsible for the fluid collection.

The proposed hormonal pathogenetic mechanism of CSDH could lead to a medical approach to CSDH patients, different from those already proposed (3, 11). In fact, the availability of some drugs with antiaromatase activity or with antiestrogen action (able to block the interconversion of androgens into estrogens or to interfere with estrogens at receptor level) allows to attempt a prophylactic therapeutic regimen of patients who experienced a skull injury, in order to inhibit the local hyperfibrinolysis induced by the hormones, and eventually to reduce the incidence of formation of CSDH. This medical treatment, however, can also be taken into consideration as adjuvant of CSDH patients who underwent surgery.

In conclusion, the results of the present study seem to support the hypothesized hormonal effects in the pathogenesis of a CSDH. The biochemical data sustain the clinical and epidemiological investigations quoted in the literature. The action of estrogens and progesterone, through their own receptors, seems to be particularly relevant at the level of the HEM, where they can increase the t-PA levels and therefore maintain the local hyperfibrinolysis. Adjuvant and prophylactic antihormal treatment may be suggested.

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