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BACHELOT A., PLU-BUREAU G., THIBAUD E., LABORDE K., PINTO G., SAMARA D., NIHOUL-FEKETE C., KUTTENN F., POLAK M., TOURAINE P.

Long-term outcome of patients with congenital adrenal hyperplasia due to 21-hydroxylase deficiency.

Horm. Res., 67 (6), 268-276, 2007 ; (Facteur d'Impact 2006 : **1,385**)

(Services cités : Endocrinologie & Médecine de la Reproduction, Endocrinologie Pédiatrique et Gynécologie, Chirurgie Viscérale Pédiatrique, Explorations Fonctionnelles)

AIMS: Conflicting results exist regarding bone mineral density (BMD), metabolism and reproductive function of adult patients with congenital adrenal hyperplasia (CAH). We evaluated the long-term outcome and the impact of chronic glucocorticoid replacement in these patients. METHODS: Physical characteristics, serum hormone concentrations, BMD and metabolism were studied in 45 consecutive CAH adult patients. RESULTS: Among the 36 women, only 14 (39%) had regular menses. Among the 27 women with classical CAH, the mean number of surgical reconstructions of virilized genitalia was 2.1 +/- 0.2. Twenty of them (74%) were sexually active. Three men presented with testicular adrenal rest tumors. Twenty-five patients (55%) had decreased BMD at the femoral neck and/or at the lumbar spine. BMI was correlated with the BMD T-score at the femoral neck ($p < 0.001$) and at the lumbar spine ($p < 0.01$). Hydrocortisone dose was negatively correlated with the BMD T-score at the femoral neck ($p = 0.04$). Subjects with osteopenia had a significantly lower BMI and received higher hydrocortisone dose than those with normal BMD. Overweight was found in 21 patients (47%). There was a significantly positive correlation between HOMA and BMI ($p < 0.001$), and between HOMA and 17-OHP levels ($p = 0.016$). CONCLUSIONS: Adult patients with CAH treated with long-term glucocorticoids are at risk for decreased BMD, increased BMI, and disturbed reproductive function.

GASTAUD F., BOUVATTIER C., DURANTEAU L., BRAUNER R., THIBAUD E., KUTTEN F., BOUGNERES P.

Impaired sexual and reproductive outcomes in women with classical forms of congenital adrenal hyperplasia.

J. Clin. Endocrinol. Metabol., 92 (4), 1391-1396, 2007 ; (Facteur d'Impact 2006 : **5,799**)

(Services cités : Endocrinologie & Médecine de la Reproduction, Endocrinologie Pédiatrique et Gynécologie)

Objectives: The objectives of the study were 2-fold: 1) a detailed description of sexual and reproductive outcomes in adult women with congenital adrenal hyperplasia (CAH) of different phenotypic severity at birth; and 2) comparisons of these outcomes among CAH subtypes and between CAH women and non-CAH control women. Design: This was a cross-sectional study using a face-to-face interview, a written questionnaire, the Female Sexual Function Index, and a gynecological examination. Patients: Patients included 35 women with CAH, representing Prader stages I-V at birth, aged 18-43 yr, who had been treated from birth to adolescence in the same pediatric endocrine clinics. Sixty-nine non-CAH healthy control women were selected from hospital-staff families. Results: None of the CAH women expressed doubts about their gender

assignment. Twenty percent (seven of 35) had homosexual inclinations; 23% (eight of 35) were married; three reported a complete lack of sexual activity; and 37% (13 of 35) said they never had heterosexual intercourse with vaginal penetration. Sexual functioning as assessed by the Female Sexual Function Index was much lower in CAH women than controls and lowest in CAH women with high Prader stages. Eighty-one percent (18 of 22) experienced pain during vaginal penetration. Only eight women became pregnant, and 17% (six of 35) had children. Conclusions: Despite expert medical and surgical care by physicians dedicated to this rare disease, women with CAH still suffer major limitations in their sexual function and reproductive life.

2006

LAISSUE P., CHRISTIN-MAITRE S., TOURAINE P., KUTTENN F., RITVOS O., AITTOMAKI K., BOURCIGAUX N., JACQUESSON L., BOUCHARD P., FRYDMAN R., DEWAILLY D., REYSS A.C., JEFFERY L., BACHELOT A., MASSIN N., FELLOUS M., VEITIA R.A.

Mutations and sequence variants in GDF9 and BMP15 in patients with premature ovarian failure. *Eur. J. Endocrinol.*, 154 (5), 739-744, 2006

(Services cités : Endocrinologie & Médecine de la Reproduction)

BACKGROUND AND OBJECTIVE: Mutations in bone morphogenic protein 15 (BMP15) and growth/differentiation factor 9 (GDF9) lead to altered fertility in animal models. In the human, a heterozygous point mutation of BMP15 has been associated with premature ovarian failure (POF). **SUBJECT AND METHODS:** We have directly sequenced both genes in a cohort of 203 POF patients presenting with primary or secondary amenorrhea and high FSH levels and in a control population including 54 women with regular menstrual cycles who had at least one child. **RESULTS:** We have identified several heterozygous variants. One alteration in GDF9 (S186Y) and one in BMP15 (L148P) may have pathogenic effects as both positions are conserved in vertebrate species, ranging from the chicken to mammals. These variants were absent in the

control samples. We also found synonymous and neutral substitutions. CONCLUSIONS: We propose that although mutations in BMP15 and GDF9 are not a major cause of ovarian insufficiency, they may be involved in POF.

MASSIN N., CZERNICHOW C., THIBAUD E., KUTTENN F., POLAK M., TOURAINE P.

Idiopathic Premature Ovarian Failure in 63 Young Women.

Hormone Res., 65 (2), 89-95, 2006

(Services cités : Endocrinologie & Médecine de la Reproduction)

Background: Premature ovarian failure (POF) in adolescents is defined as primary or secondary amenorrhea associated with high follicle-stimulating hormone (FSH) levels. In normal 46,XX patients, its etiology is most often unknown. We have evaluated the clinical, hormonal and ovarian phenotypes in patients with a normal karyotype who were diagnosed with POF before the age of 18. Methods: Sixty-three patients were included in this retrospective study. Results: The mean patient age was 20.4 years. The patients presented with three clinical patterns: lack of pubertal development (n = 23), primary amenorrhea with interrupted puberty (n = 18), and secondary amenorrhea with normal puberty (n = 22). Ten patients had a familial history of POF and 6 presented with hypothyroidism. The FSH, estradiol and inhibin B levels were not statistically different in the three clinical groups. Fifty percent of the patients presented small ovaries (length <2 cm) at ultrasonography. The presence of follicles was found at histology in only 7 of the 27 patients who underwent an ovarian biopsy. Conclusion: 46,XX patients presenting with early POF rarely presented a specific, identifiable disorder. We discuss the clinical management and different diagnosis strategies to improve our current knowledge of this syndrome. Copyright (c) 2006 S. Karger AG, Basel.

PLU-BUREAU G., LE M.G., SITRUK-WARE R., THALABARD J.C.

Cyclical mastalgia and breast cancer risk: results of a French cohort study.

Cancer Epidemiol. Biomarkers. Prev., 15 (6), 1229-1231, 2006

(Services cités : Endocrinologie & Médecine de la Reproduction)

Cyclical mastalgia is a common complaint, with a potentially important relationship to breast cancer risk. In the last decade, case-control studies have reported that cyclical mastalgia could be considered as an independent risk factor for breast cancer. The subjectivity of a retrospectively collected symptom questioned the validity of this finding. We have examined the association between cyclical mastalgia and breast cancer risk in the French cohort study of women with benign breast disease diagnosed in two breast clinics between 1976 and 1979 and followed-up until 1997. The present study was restricted to the women free of any hormonal treatment (n = 247). The mean foll mastalgia was 5.31 (95% confidence interval, 1.92-14.72). We show here that the conclusion still holds when the symptom cyclical mastalgia was collected prospectively in a nging additional evidence that cyclical mastalgia may represent an independent marker of increased breast cancer risk. It might be a confounding factor when assessing the effects of hormonal treatments on breast cancer risk such as hormonal replacement therapy or oral contraceptives.

RUANO R., JOUBIN L., AUBRY M.C., THALABARD J.C., DOMMERGUES M.,

DUMEZ Y., BENACHI A.

A nomogram of fetal lung volumes estimated by 3-dimensional ultrasonography using the rotational technique (virtual organ computer-aided analysis).

J. Ultrasound Med., 25 (6), 701-709, 2006

(Services cités : Endocrinologie & Médecine de la Reproduction, Obstétrique)

OBJECTIVE: The purpose of this study was to build a nomogram of normal fetal lung volumes throughout gestational age estimated by 3-dimensional ultrasonography using the rotational technique (Virtual Organ Computer-Aided Analysis [VOCAL]; GE Healthcare, Kretztechnik, Zipf, Austria). **METHODS:** Fetal lung volume was assessed in 146 healthy fetuses by 3-dimensional ultrasonography using the technique of rotation of the multiplanar imaging (VOCAL). Inclusion criteria were healthy women with singleton normal pregnancies, normal fetal morphologic ultrasonographic findings, reliable dating established by dates and by ultrasonographic measurement of the crown-rump length in the first trimester, and gestational age from 20 to 37 weeks. Exclusion criteria were discordance between clinical and ultrasonographic dating, patients lost to follow-up, and birth weight disorders. Each patient was scanned once during pregnancy. **RESULTS:** The right, left, and total mean pulmonary volumes ranged, respectively, from 5.37, 4.66, and 9.95 cm³ at 20 weeks to 46.06, 37.34, and 84.35 cm³ at 37 weeks. The logistic transformation analysis yielded the following formulas: right lung volume = $\exp(4.07/[1 + \exp(21.90 - \text{gestational age}/5.44)])$; left lung volume = $\exp(3.82/(1 + \exp[22.03 - \text{gestational age}/5.17]))$; and, total lung volume = $\exp(4.72/[1 + \exp(20.30 - \text{gestational age}/6.05)])$. **CONCLUSIONS:** A new nomogram of fetal lung (right, left, and total) volumes throughout gestational age using the rotational technique (VOCAL) is described, and reference values have been generated.

2005

BACHELOT A., MEDURI G., MASSIN N., MISRAHI M., KUTTENN F., TOURAINE P.

Ovarian steroidogenesis and serum androgen levels in patients with premature ovarian failure.

J. Clin. Endocrinol. Metabol., 90 (4), 2391-2396, 2005

(Services cités : Endocrinologie & Médecine de la Reproduction)

Women with premature ovarian failure (POF) have been reported to have lower serum androgen levels compared with normal women. We reviewed the androgen profiles of 143 POF patients and found androgen levels above normal for postmenopausal women in 16% of these subjects. To determine the source of androgens in those women, we studied the available ovarian biopsy samples of 15 POF patients with increased androgen levels using immunohistochemistry, with a panel of antibodies directed against the main steroidogenic enzymes. Five of the ovarian biopsies exhibited abnormal follicles characterized by hypertrophied theca interna expressing steroidogenic enzymes involved in androgen synthesis. In five other biopsies, the steroidogenic activity was scarce and confined to a small number of ovarian stromal cells, sometimes situated in the proximity of follicular remnants. Finally, in five patients, we found no histological evidence of present or past follicular development beyond the quiescent follicular stage, and no steroidogenic cells were detected by immunohistochemistry. Our findings suggest that ovarian theca-derived cells are a source of androgens in some women with POF, whereas in others, as in most postmenopausal patients, the adrenals or the ovarian hilus cells may synthesize a significant quantity of androgens under LH stimulation.

BACHELOT A., COURTILOT C., TOURAINE P.

When and how should hyperprolactinemia be treated ?

Presse Médicale, 34 (10), 731-737, 2005

(Services cités : Endocrinologie & Médecine de la Reproduction)

Hyperprolactinemia affects the gonadotropic axis. Its results in women include amenorrhea, menstrual disorders and galactorrhea; in men, the frequency of macroadenomas tends to lead to problems related to sexual performance or tumor volume. Radioimmunoassays make diagnosis easy. Secondary causes of hyperprolactinemia, drug reactions in particular, must be ruled out before MRI exploration to look for a pituitary tumor. First-line treatment of prolactin adenomas is based on the use of dopaminergic agonists, especially cabergoline, because of their excellent efficacy and the risk of relapse following surgery. For patients who wish to become pregnant, the dopaminergic agonist must be continued during pregnancy for those with macroadenoma and withdrawn for women with microadenoma. When hyperprolactinemia is induced by anti-psychotic agents, treatment requires an in-depth assessment,

GOFFIN V., BERNICHTEIN S., TOURAINE P., KELLY P.A.

Development and potential clinical uses of human prolactin receptor antagonists.

Endocrine Rev., 26 (3), 400-422, 2005

(Services cités : Endocrinologie & Médecine de la Reproduction, U584)

There is a large body of literature showing that prolactin (PRL) exerts growth-promoting activities in breast cancer, and possibly in prostate cancer and prostate hyperplasia. In addition, increasing evidence argues for the involvement of locally produced (autocrine) PRL, perhaps even more than pituitary-secreted (endocrine) PRL, in tumor growth. Because dopamine analogs are unable to inhibit PRL production in extrapituitary sites, alternative strategies need investigation. To that end, several PRL receptor antagonists have been developed by introducing various mutations into its natural ligands. For all but one of these analogs, the mechanism of action involves a competition with endogenous PRL for receptor binding. Such compounds are thus candidates to counteract the undesired actions of PRL, not only in tumors, but also in dopamine-resistant prolactinomas. In this review, we describe the different versions of antagonists that have been developed, with emphasis on the controversies regarding their characterization, and the limits for their potential development as a drug. The most recently developed antagonist, Delta1-9-G129R-hPRL, is the only one that is totally devoid of residual agonistic activity, meaning it acts as pure antagonist. We discuss to what extent this new molecule could be considered as a lead compound for inhibiting the actions of human PRL in the above-mentioned diseases. We also speculate on the multiple questions that could be addressed with respect to the therapeutic use of PRL receptor antagonists in patients.

LACCOURREYE O., CAUCHOIS R., TOURAINE P., GARAY A., BOURLA A.

Preoperative oral information prior to planned thyroid surgery: the surgeon, physician, lawyer and judge's point of view.

Ann. Chir., 130 (8), 458-465, 2005

(Services cités : Endocrinologie & Médecine de la Reproduction)

Objective. - Analysis of the consequence of the preoperative information delivered orally to patients requiring surgery for benign pathology of the thyroid gland from various perspective (head and neck surgeon, physician, judge, lawyer). Methods. - Prospective study conducted in an academic tertiary care referral center. Inception cohort of 123 patients with benign pathology of the thyroid gland consecutively informed orally and by the same surgeon during the years 2003-2004. Analysis of the consequences of the preoperative information, degree of memorization regarding the surgical related risks as well as the patient's opinion regarding this information.

Results. - Due to the information regarding the risks related to surgery 14.6% of patients refused to undergo surgery. None of the patients remembered more than 4 out of the 6 main surgical risks. 68.8% of patients remembered only one or two surgical related risks. 12.2% of patients did not remember a single surgical related risk, 85.5% of patients remembered the risk of a unilateral laryngeal nerve paralysis, 41.1% the risk of death related to the completion of a general anesthesia and 21.1% the risk of a bilateral laryngeal nerve paralysis. Less than 11% of patients remembered the other risks (general risks related to any open field surgical approach, hypocalcemia and loss of breast feeding) Among the patients who had an opinion postoperatively, 87.6% had a positive opinion and 41.9% a negative opinion regarding the preoperative information related to the surgical related risks. Also, 28.4% of patients expressed simultaneously a positive and a negative opinion. Conclusion. - Oral information of the patient regarding the surgical risks resulted in an important stress for the patient and modified the relation patient-surgeon. Due to the information, a non-neglectable group of patients elicited not to follow the advice of the surgeon. Various measures are discussed since the information on the surgical related risks is a must from a legal point of view but is also highly appreciated and requested by most of the patients.

MITCHELL J.C., LI X.F., BREEN L., THALABARD J.C., O'BYRNE K.T.

The Role of the Locus Coeruleus in Corticotropin-Releasing Hormone and Stress-Induced Suppression of Pulsatile Luteinizing Hormone Secretion in the Female Rat.

Endocrinology, 146 (1), 323-331, 2005

(Services cités : Endocrinologie & Médecine de la Reproduction, Biostatistique)

TOURAINÉ P., YOUSSEF N., ALYANAKIAN M.A., LECHAT X., BALLEYGUIER C., DUFLOS C., DIB A., MAY A., CAREL J.C., LABORDE K., SIGAL-ZAFRANI B., GOFFIN V., EYMARD B., BOITARD C., BROUSSE N., KUTTENN F.

Breast inflammatory gigantomastia in a context of immune-mediated diseases.

J. Clin. Endocrinol. Metabol., 90 (9), 5287-5294, 2005

(Services cités : Anatomo-Pathologie, Endocrinologie & Médecine de la Reproduction, Infectiologie, Radiologie Adulte, U584, Explorations Fonctionnelles)

Context: Localized breast lesions have been described in lupic or diabetic patients. However, the description of breast gigantomastia in women presenting with autoimmune diseases has not been reported. Setting: The study took place within the Department of Endocrinology and Reproductive Medicine, Necker Hospital, Paris, France. Patients: We describe eight patients with inflammatory gigantomastia, occurring in a context of immune-mediated diseases: myasthenia, chronic arthritis, or thyroiditis. Main Outcome Measures: Together with hormonal, immunological, and breast magnetic resonance imaging (MRI) evaluation, breast histology enabled us to perform immunocytochemical and indirect immunofluorescence studies. Control sera were obtained from patients with (n = 10) and without (n = 7) antinuclear antibodies. Results: Six of the eight patients developed gigantomastia either at puberty or during pregnancy. Neither a hormonal oversecretion nor a specific immunological pattern was observed. All patients except one presented antinuclear antibodies. Histological study revealed a diffuse, stromal hyperplasia and a severe atrophy of the lobules. A rarefaction of adipocytes was also noted, as previously suggested on MRI. There was a perilobular lymphocytic infiltrate made of CD3+ lymphocytes. Study of sera from five of six cases of gigantomastia showed a nuclear immunofluorescence pattern in normal mammary ductal and lobular glandular epithelium, as well as in kidney and intestine epithelial cells. In control sera, a nuclear signal was observed only

when antinuclear antibodies were present. Conclusions: We suggest that breast tissue may be a target tissue in autoimmune diseases, this process being favored by the hormonal milieu. However, the precise mechanism of such association is not individualized. The fact that stromal hyperplasia is the main histological feature justifies the search for the involvement of growth factors in such a process.

2004

ALGAZI M., PLU-BUREAU G., FLAHAULT A., DONDON M.G., LE M.G.

Could treatments with beta-blockers be associated with a reduction in cancer risk?

Rev. Épidémiol. Santé Publ., 52 (1), 53-65, 2004

(Services cités : Endocrinologie & Médecine de la Reproduction, URC)

BACKGROUND: The relationship between the use of anti-hypertensive drugs and cancer risk remains controversial. The main objective of this study was to assess the potential effect of beta-blocker use on cancer risk. **METHODS:** In a cohort of 839 patients with cardiovascular disease, followed up prospectively for an average period of 10 years, cancer occurrence was recorded according to the exposure to beta-blockers. The relative risk of cancer associated with beta-blocker use was estimated using a Cox model adjusted on gender and age. Ever- vs never-use of beta-blockers and duration of exposure to the drug were analyzed as time-dependent variables. In addition, the standardized incidence ratios (SIR) were calculated using the corresponding age- and gender-adjusted cancer incidences in the French general population. **RESULTS:** A total of 326 beta-blocker users and 513 users of other treatments were included in the cohort. During the follow-up period, representing 8,466 person-years, incident cancer cases were 15 and 59 in beta-blocker ever-users versus never-users, respectively. Using the Cox model, the overall relative risk of cancer was 0.51 (95% confidence interval [95% CI]: 0.29-0.90) in the beta-blocker ever-users versus never-users ($p=0.02$), with a 6% decrease per year of use (95% CI: 1%-12%; $p=0.03$). The corresponding SIR ratio between these two groups was 0.44 (95% CI: 0.24-0.76).

CONCLUSION: In this cohort, the beta-blocker treatments appeared to decrease the cancer risk significantly. However, this result should be considered with caution; further work is needed, as some sources of bias associated with this type of epidemiological study cannot be totally excluded.

BACHELOT A., MEDURI G., BAUDIN E., KUTTENN F., TOURAINE P.

Hyperandrogenism in a postmenopausal woman presenting with a metastatic ileum endocrine tumor.

Fert. Steril., 81 (3), 675-678, 2004

(Services cités : Endocrinologie & Médecine de la Reproduction)

OBJECTIVE: To elucidate the mechanism of the hyperandrogenism found in a postmenopausal woman presenting an ileum endocrine tumor with ovarian metastases. **DESIGN:** Case report. **SETTING:** University hospital. **PATIENT(S):** A postmenopausal woman was referred for hirsutism. Basal plasma testosterone was high, 6.6 nM/L (normal, ≤ 0.7 nM/L). Pelvic magnetic resonance imaging revealed a 6-cm left ovarian mass. **INTERVENTION(S):** Bilateral salpingo-oophorectomy was performed. Pathological examination found a bilateral metastatic endocrine ovarian tumor, associated with a functional stroma. A primary ileum endocrine tumor was discovered and resected. **MAIN OUTCOME MEASURE(S):** Immunohistochemical study of the expression of steroidogenic enzymes and beta and alpha subunits of hCG. **RESULT(S):** Immunohistochemical expression of steroidogenic enzymes was found in the ovarian stromal tissue surrounding the tumor but not in the metastatic tumoral cells. A substantial percentage of

the metastatic tumoral cells was immunopositive for the beta and alpha subunit of hCG but not the ileal cells. CONCLUSION(S): These data suggest an hCG paracrine effect of the ovarian metastases tumor on the adjacent interstitial cells, resulting in the virilization of the patient.

CONARD J., PLU-BUREAU G., BAHI N., HORELLOU M.H., PELISSIER C., THALABARD J.C.

Progestogen-only contraception in women at high risk of venous thromboembolism.

Contraception, 70 (6), 437-441, 2004

(Services cités : Endocrinologie & Médecine de la Reproduction, URC)

The objective of the study was to evaluate the venous impact of a progestogen-only contraception on women at high risk of venous thromboembolism (VTE). In this retrospective cohort study, 204 consecutive women at high risk of VTE were recruited between January 1992 and June 1997 and were prospectively followed. Women using chlormadinone acetate (CMA) at antigonadotropic doses (n=102) were matched by age and date of referral and history of venous thrombosis with women who had no hormonal contraception (n=102). During follow-up (mean of 33 months), nine episodes of VTE were observed: three in women receiving CMA and six in nontreated women. Using the Cox model to adjust for confounding variables such as age, thrombophilia and body mass index, the relative risk of VTE associated with the use of CMA was not significant [relative risk: 0.8 (0.2-3.9)]. These reassuring results need to be confirmed in other prospective studies.

DUFLOS C., PLU-BUREAU G., THIBAUD E., KUTTENN F.

Breast diseases in adolescents.

in: *Pediatric and adolescent gynecology*. (Sultan C. eds.)

Karger (Germany), 2004, pp.183-196.

(Services cités : Endocrinologie & Médecine de la Reproduction)

LETUR-KONIRSCH H., COLLIN G., DEVAUX A., SIFER C., KUTTENN F., MADELENAT P., BRUN-VEZINET F., FELDMANN G., BENIFLA J.L.

Conservation of human embryos in straws: safety in terms of human immunodeficiency virus 1.

Gynécol. Obstét. Fertil., 32 (4), 302-307, 2004

(Services cités : CECOS, Endocrinologie & Médecine de la Reproduction)

OBJECTIVE: The possibility of offering assisted reproductive technologies (ART) to HIV-positive couples has revived questions concerning the safety of the gametes and embryos cryopreservation in liquid nitrogen tanks. PATIENTS AND METHODS: We evaluated the safety of three types of straws - polyvinyl chloride (PVC), polyethylene terephthalate glycol (PETG) and so-called 'high-security' ionomeric resin (IR) - containing HIV-1 under standard conditions of cryopreservation. Potential HIV contamination was assessed by RT-PCR and then nested PCR. RESULTS: Under cryopreservation conditions, the sealed open ends of PVC and PETG straws were not safe. The ultrasound sealing system seems to be the weak link in obtaining total imperviousness of the straws. In contrast, both ends of the IR straws were safe for HIV in the framework of their use for ART. CONCLUSION: Sealing cryopreservation straws ultrasonically could incur the risk of not assuring their impermeability. Under standard cryopreservation conditions thermosealing of IR straws appears to be safe for HIV.

MASSIN N., GOUGEON A., MEDURI G., THIBAUD E., LABORDE K., MATUCHANSKY C., CONSTANCIS E., VACHER-LAVENU M.C., PANIEL B., ZORN

J.R., MISRAHI M., KUTTENN F., TOURAIN P.

Significance of ovarian histology in the management of patients presenting a premature ovarian failure.

Hum. Reprod., 19 (11), 2555-2560, 2004

(Services cités : Endocrinologie & Médecine de la Reproduction, Endocrinologie et Croissance, Explorations Fonctionnelles, U584)

BACKGROUND: Premature ovarian failure (POF) is a heterogeneous syndrome, possibly due to mutations of genes involved in the normal development of the ovary and/or follicles. Based essentially on animal models, these mutations are associated with various ovarian phenotypes, from a complete absence of follicles to a partial follicular maturation. The aim of the present study was to determine whether ovarian histology, compared to pelvic ultrasonography, would be helpful in identifying which patients display an impaired follicular reserve and/or growth, and in orientating the search for POF aetiology. **METHODS AND RESULTS:** We studied a cohort of 61 patients suffering from POF with a normal karyotype. Their median age (range) at diagnosis was 26 years (15-39). The FSH plasma level was high, 67.0 IU/l (13-155). Estradiol and inhibin B plasma levels were low: 18.5 pmol/l (18.5-555) and 5 pg/ml (5-105) respectively. Both pelvic ultrasonography and ovarian biopsies were performed in each patient. The presence of follicles suggested at ultrasonography was confirmed at histology in 56% of the patients. Ovarian histology led to the distinction of two phenotypes: (i) small-sized ovaries, deprived of follicles; and (ii) normal-sized ovaries with partial follicular maturation. To confirm the value of ovarian biopsies, samples from 20 normal women were studied. These demonstrated that ovarian biopsy at random enables reliable assessment of follicular presence, especially when their size is <2 mm. **CONCLUSION:** Ovarian histology appears to be a reliable tool in evaluating the follicular reserve, and helpful and complementary to clinical and hormonal phenotyping in orienting the search for the various genetic causes of POF syndrome.

PRIMI M.P., SENN A., MONTAG M., VAN DER VEN H., MANDELBAUM J., VEIGA A., BARRI P., GERMOND M.

A European multicentre prospective randomized study to assess the use of assisted hatching with a diode laser and the benefit of an immunosuppressive/antibiotic treatment in different patient populations.

Hum. Reprod., 19 (10), 2325-2333, 2004

(Services cités : Endocrinologie & Médecine de la Reproduction)

BACKGROUND: Assisted hatching (AH) techniques, designed for facilitating the embryo escape out of the zona pellucida (ZP) have been used in IVF centres since 1992. The initial indications for AH were patient's age, ZP thickness, high basal FSH and repeated IVF failures. Several retrospective and prospective studies assessing AH in these indications have given disparate results. Our aims were to evaluate the benefits of AH and immunosuppressive/antibiotic treatment (IA) in patients with either a poor prognosis of success, previous implantation failures or transfers of cryopreserved embryos. **METHODS:** Four IVF centres allocated 426 patients, randomized for AH and IA, into four groups of AH indications between 1997 and 1999. AH was performed with a diode laser. ZP thickness, opening size and embryo score were recorded. Outcome measures were implantation and delivery rates. **RESULTS:** Patients coming for a first or third transfer of cryopreserved embryos and poor prognosis patients admitted for a first trial did not benefit from AH. Even patients with repeated implantation failures of fresh embryos did not gain significantly from AH. **CONCLUSIONS:** Among AH indications, absence of implantation after several transfers of good quality embryos remains the strongest patient

selection criterion. Prescription of an immunosuppressive/antibiotic treatment is essential.

RUANO R., BENACHI A., JOUBIN L., AUBRY M.C., THALABARD J.C., DUMEZ Y., DOMMERGUES M.

Three-dimensional ultrasonographic assessment of fetal lung volume as prognostic factor in isolated congenital diaphragmatic hernia.

Br. J. Obstet. Gynaecol. - BJOG, 111 (5), 423-429, 2004

(Services cités : URC, Maternité, Endocrinologie & Médecine de la Reproduction)

Objective To evaluate the potential of three-dimensional ultrasound to predict outcome in congenital diaphragmatic hernia. **Design** Prospective observational study. **Setting** Tertiary care centre. **Population** Twelve cases of isolated congenital diaphragmatic hernia (11 left-sided, 1 right-sided) and 109 controls. **Methods** Fetal lung volume was assessed by three-dimensional ultrasound using the technique of rotation of the multiplanar imaging. In the control fetuses, a logistic transformation was performed to correlate fetal lung volume with gestational age, and the confidence interval was obtained with a bootstrap resampling. A mathematical equation was then obtained allowing calculation of the expected fetal lung volume as a function of gestational age. In fetuses with congenital diaphragmatic hernia, the observed/expected lung volume ratio was compared with postnatal outcome. **Main outcome measures** Neonatal mortality and pulmonary hypoplasia, which was defined as lung/body weight ratios less than 0.012. **Results** The expected fetal lung volume was derived from the mathematical equation: Fetal lung volume (mL) = $\exp(4.72/(1 + \exp((20.32 - \text{gestational age in weeks})/6.05)))$. The observed/expected fetal lung volume ratio was significantly lower in the congenital diaphragmatic hernia group (median: 0.34, range: 0.16-0.66), than in the control group (median: 1.02, range: 0.62-1.97, $P < 0.0001$). The distribution of this ratio was significantly downshifted in the infants with congenital diaphragmatic hernia who died (median: 0.19, range: 0.18-0.66) compared with survivors (median: 0.44, range: 0.36-0.66, $P = 0.04$). The observed/expected fetal lung volume ratio was also correlated with the postmortem lung/body weight ratio. **Conclusion** In isolated congenital diaphragmatic hernia, fetal lung volume measurement by three-dimensional ultrasound is a potential predictor for pulmonary hypoplasia and postnatal outcome.

SITRUK-WARE R., PLU-BUREAU G.

Exogenous progestagens and the human breast.

Maturitas, 49 (1), 58-66, 2004

(Services cités : Endocrinologie & Médecine de la Reproduction, URC)

The role of progestins (or progestagens) on the breast tissue remains controversial. However, according to the molecule and the duration of application, cell differentiation and apoptosis may predominate over proliferation. Progestins are also used as second-line agents for the treatment of metastatic breast cancer. In young women with benign breast disease, long-term treatment with 19-nortestosterone progestins had a trend to decrease breast cancer risk contrarily to what was observed in postmenopausal women receiving estrogens. Several compounds with progestational activity have been used for HRT. Small differences in the structure of the molecules may lead to pronounced differences in activities, some progestins exerting androgenic effects and some exerting estrogenic or glucocorticoid like activities. While most progestins do not bind to the estrogen receptors, it has been shown that some androgenic progestins stimulate MCF7 cells proliferation while progestins derived from progesterone did not induce cell multiplication in the same cell lines. Therefore, different progestins may induce different effects on the breast cells. Whether the progestins available to date are able to bind specifically to the progesterone

receptors PR-A or PR-B and whether this is of clinical relevance to breast cell proliferation is still unclear. Although the relationship between progestin use and breast cancer risk is still the subject of debate and controversy, the data reported to date suggest that 5 years of treatment carry a low risk but further duration of use increases the risk. Further studies are still needed, randomised long-term prospective studies as well as from the laboratory, especially to determine whether a sequential or continuous regimen would be preferable as far as breast-cell response and apoptosis are concerned, and what are the effects of the various molecules used for HRT.

SKURNIK G., TOURAINE P.

Doit-on continuer le traitement hormonal substitutif de la ménopause chez les femmes diabétiques ?

Journ. Annu. Diabétol. Hotel-Dieu, 151-160, 2004

(Services cités : Endocrinologie & Médecine de la Reproduction)

THALABARD J.C., PLU-BUREAU G.

Hormone replacement therapy after chemo-induced precocious menopause: do the risks exceed the benefits ?

in: *Eurocancer 2004*. (Boiron M., Marty M. eds.)

JOHN LIBBEY EUROTEXT (Montrouge, F), 2004, pp.105-107.

(Services cités : Endocrinologie & Médecine de la Reproduction)

TOURAINE P., GOFFIN V., KELLY P.A., MORANGE I., JACQUET P.

La prolactine : physiologie et pathologie.

in: *Traité de Médecine (4^e édition)*. (Godeau P. eds.)

Flammarion Médecine-Sciences (Paris), 2004, pp.1941-1946.

(Services cités : Endocrinologie & Médecine de la Reproduction, U584)

2003

MASSIN N., PECHEUX C., ELOIT C., BENSIMON J.L., GALEY J., KUTTENN F., HARDELIN J.P., DODE C., TOURAINE P.

X Chromosome-Linked Kallmann Syndrome: Clinical Heterogeneity in Three Siblings Carrying an Intragenic Deletion of the KAL-1 Gene.

J. Clin. Endocrinol. Metabol., 88 (5), 2003-2008, 2003

(Services cités : Endocrinologie & Médecine de la Reproduction)

Kallmann syndrome (KS) is characterized by the association of hypogonadotropic hypogonadism and anosmia. The gene underlying the X chromosome-linked form of the disease, KAL-1, consists of 14 coding exons. It encodes a glycoprotein, anosmin-1, which is involved in the embryonic migration of GnRH-synthesizing neurons and the differentiation of the olfactory bulbs. We describe herein the clinical heterogeneity in three affected brothers who carry a large deletion (exons 3-13) in KAL-1. All three had a history of hypogonadotropic hypogonadism with delayed puberty. Although brain magnetic resonance imaging showed hypoplastic olfactory bulbs in the three siblings, variable degrees of anosmia/hyposmia were shown by olfactometry. In addition, these brothers had different phenotypic anomalies, i.e. unilateral renal aplasia (siblings B and C), high-arched palate (sibling A), brachymetacarpia (sibling A), mirror movements (siblings A and B), and abnormal eye movements (sibling C). Last but not least, sibling A suffered from a severe congenital hearing impairment, a feature that had been reported in KS but had not yet been ascribed unambiguously to the X-linked form of the disease. The variable

phenotype, both qualitatively and quantitatively, in this family further emphasizes the role of putative modifier genes, and/or epigenetic factors, in the expressivity of the X-linked KS.

MEDURI G., TOURAINE P., BEAU I., LAHUNA O., DESROCHES A., VACHER-LAVENU M.C., KUTTENN F., MISRAHI M.

Delayed puberty and primary amenorrhea associated with a novel mutation of the human follicle-stimulating hormone receptor: clinical, histological, and molecular studies.

J. Clin. Endocrinol. Metabol., 88 (8), 3491-3498, 2003

(Services cités : Endocrinologie & Médecine de la Reproduction)

Inactivating mutations of the FSH receptor have been described in rare cases of premature ovarian failure. Only one mutation was associated with a complete phenotype, including delayed puberty, primary amenorrhea, and small ovaries. We describe here a new patient presenting a similar complete phenotype of premature ovarian failure, with high plasma FSH levels associated with very low estrogen and inhibin B levels. No biological response to high doses of recombinant FSH was detected. A novel homozygous Pro(519)Thr mutation was found in this patient. This mutation is located in the second extracellular loop of the FSH receptor, within a motif highly conserved in gonadotropin and TSH receptors. The mutation totally impairs adenylate cyclase stimulation in vitro. FSH binding experiments and confocal microscopy showed that this mutation alters the cell surface targeting of the mutated receptor, which remains trapped intracellularly. Histological studies of the ovaries of the patient showed an increase in the density of small follicles compared with age-matched normal women. A complete block in follicular maturation after the primary stage was also observed. Immunocytochemical studies allowed detection of the expression of c-Kit and proliferation cellular nuclear antigen, whereas no apoptosis was shown by the 3'-end-labeling method. This observation supports the concept that in humans FSH seems mandatory for the initiation of follicular growth only after the primary stage. In our patient complete FSH resistance yields infertility, which is remarkably associated with the persistence of a high number of small follicles.

MESTAYER C., BLANCHERE M., JAUBERT F., DUFOUR B., MOWSZOWICZ I.

Expression of androgen receptor coactivators in normal and cancer prostate tissues and cultured cell lines.

Prostate, 56 (3), 192-200, 2003

(Services cités : Urologie, Endocrinologie & Médecine de la Reproduction)

BACKGROUND: In prostate cancer cell lines, androgen receptor (AR) coactivators modulate the transcriptional activity of AR. However, very little is known about their expression in normal prostate tissue and during progression to cancer. **METHODS:** AR and coactivators ARA54, ARA55, ARA70, and SRC1 RNA were analyzed by RT-PCR in normal and tumoral tissues of the same prostate, in prostate cell lines, and after hormonal treatments of prostate epithelial cells. **RESULTS:** AR-coactivators were expressed in normal and tumoral tissues and in cultured prostate cells; only ARA55 expression was decreased in tumoral relative to normal tissue of all seven prostates analyzed. It was not expressed in LNCaP and DU145 cancer cells and low in PNT2 immortalized cells in which all coactivator's expression were down regulated by DHT and up regulated by E2. In addition, coactivator's expression was increased in hyperplastic relative to normal prostate fibroblasts. **CONCLUSIONS:** ARA55 is both an AR coactivator and a focal adhesion protein (Hic-5). Its role in the progression of prostate carcinoma may therefore involve these two different functions. Its decrease in cancer tissue suggests that it plays a different role than that expected, namely, facilitate cell proliferation and therefore mobility and metastasis.

Prostate 56: 192-200, 2003.

RUANO R., BENACHI A., JOUBIN L., AUBRY M.C., THALABARD J.C., DUMEZ Y., DOMMERGUES M.

P255: Fetal lung volume assessment by three-dimensional ultrasound in isolated congenital diaphragmatic hernia as a prognostic factor.

Ultrasound Obstet. Gynecol., 22 (S1), 139, 2003

(Services cités : Maternité, Endocrinologie & Médecine de la Reproduction)

No Abstract.

SONIGO P., RUANO R., DOMMERGUES M., MAHIEU-CAPUTO D., THALABARD J.C., BENACHI A., SIMON I., DUMEZ Y., BRUNELLE F.

OC169: Prediction of pulmonary hypoplasia with MRI.

Ultrasound Obstet. Gynecol., 22 (S1), 46., 2003

(Services cités : Radiologie Pédiatrique, Maternité, Endocrinologie & Médecine de la Reproduction)

No Abstract.

TOURAINÉ P.

SERMs and the uterus.

Ann. Méd. Intern., 154 (2), 103-108, 2003

(Services cités : Endocrinologie & Médecine de la Reproduction)

The uterus is one of the target organs of sexual steroids synthesized in the ovary. Estrogen is known to stimulate cell proliferation in the endometrium while progesterone has an anti-estrogenic secretory effect on this tissue. Renewed interest in the action of new anti-estrogenic agents on the uterus has arisen over the last decade, but not simply in order to achieve new therapeutic strategies for the prevention or cure of uterine tumors. New compounds were developed for their action on other tissues such as the breast, but it rapidly became clear that they were a source of uterine disease. A clear example is tamoxifen which has a powerful anti-estrogenic effect on breast tissue. It was hoped however that this compound, which behaves either like an antagonist or an agonist, depending on the target tissue, could have an anti-estrogenic effect on the uterus and on the contrary an agonistic estrogenic protective effect on bony and vascular tissue. This approach progressively led to the development of SERMs (Selective Estrogen Receptor Modulators), non-steroidal compounds modulating the action of estrogens. The first member of this new pharmaceutical class was raloxifen, marketed in France under the brand name Evista, which has an estrogenic effect on vertebral bone, warranting its authorization for use in patients with vertebral osteoporosis with or without fracture. Raloxifen thus has a beneficial estrogenic effect, at least on trabecular bone, and an anti-estrogenic effect on the uterus and breast. The goal today is to continue the development of new compounds in the SERM family with well targeted, and well understood, agonistic and/or antagonistic actions on different body tissues.

2002

BLANCHÈRE M., SAUNIER E., MESTAYER C., BROSHUIS M., MOWSZOWICZ I.

Alterations of expression and regulation of transforming growth factor beta in human cancer prostate cell lines.

J. Steroid Biochem. Mol. Biol., 82 (4-5), 297-304, 2002

(Services cités : Endocrinologie & Médecine de la Reproduction)

TGFbeta can promote and/or suppress prostate tumor growth through multiple and opposing actions. Alterations of its expression, secretion, regulation or of the sensitivity of target cells can lead to a favorable environment for tumor development. To gain a better insight in TGFbeta function during cancer progression, we have used different cultured human prostate cells: preneoplastic PNT2 cells, the androgen-dependent LNCaP and the androgen-independent PC3 and DU145 prostate cancer cell lines. We have studied by specific ELISA assays in conditioned media (CM), the secretion of TGFbeta1 and TGFbeta2 in basal conditions and after hormonal treatment (DHT or E2) and the expression of TGFbeta1 mRNA by Northern blot. We have also compared the effect of fibroblast CM on TGFbeta secretion by the different cell types. Compared to PNT2 cells, cancer cell lines secrete lower levels of active TGFbeta which are not increased in the presence of fibroblast CM. LNCaP cells respond to androgen or estrogen treatment by a 10-fold increase of active TGFbeta secretion while PC3 and DU145 are unresponsive. In conclusion, prostate cancer cell lines have lost part of their ability to secrete and activate TGFbeta, and to regulate this secretion through stromal-epithelial interactions. Androgen-sensitive cancer cells may compensate this loss by hormonal regulation.

CLEMENT F., MONNIAUX D., THALABARD J.C., CLAUDE D.

Contribution of a mathematical modelling approach to the understanding of the ovarian function. *C. R. Biol.*, 325 (4), 473-485, 2002

(Services cités : Biostatistique, Endocrinologie & Médecine de la Reproduction, URC)

The biological meaning of folliculogenesis is to free fertilisable oocytes at the time of ovulation. We approached the study of the control of follicular development at the level of follicular granulosa cells, on the experimental as well as mathematical modelling grounds. We built a mathematical model allowing for the processes of proliferation, differentiation and apoptosis. State variables correspond to the numbers of cells undergoing these different processes, while control variables correspond to the cellular transition rates. The model results raised the notion of proliferative resources, which leads to consider the optimal management of these resources and has motivated the settling of an experiment investigating the changes in the growth fraction within the granulosa throughout terminal development. We are now investigating the way gonadotrophins, and especially FSH, operate on granulosa cells, in order to account for the hormonal control of the divergent commitment of granulosa cells towards either proliferation, differentiation or apoptosis. We are thus focusing on the dynamics of cAMP production, which appears to be a keypoint in FSH signal transduction. (C) 2002 Academie des sciences/Editions scientifiques et medicales Elsevier SAS.

DE LIGNIERES B.

Effects of progestogens on the postmenopausal breast.

Climacteric, 5 (3), 229-235, 2002

(Services cités : Endocrinologie & Médecine de la Reproduction)

The potential for an increased risk of breast cancer linked to the use of synthetic progestins combined with oral estrogens is one of the main putative reasons for discouraging postmenopausal women from using any type of hormone replacement therapy (HRT) for more than a few years. Because no definitive proof exists, the available epidemiological results can be interpreted according to what seems biologically plausible to each investigator, including potential differences between various schedules of various steroids in various species and in vitro models. More than 60 years after the discovery of progesterone, the main effects

of this endogenous steroid on the physiopathology of the breast during a normal luteal phase are still controversial. The lack of consensus on such basic knowledge concerning progesterone; one of the most important targets of a natural ovarian hormone discovered in 1934 is amazing. In the most cited studies, nothing has been done to measure progesterone in plasma and to correlate the extremely disparate cytological results with extremely erratic steroid levels at the time of surgical stress. In a recent study, with a better design, the physiological rise of endogenous progesterone during the luteal phase coincided with a drop in proliferation of breast epithelial cells, which appears to be only slightly delayed in comparison with what is described in the endometrium. Differences in doses and schedules of treatments with various synthetic progestins have largely contributed to the inconsistency in clinical recommendations. Based on the analysis of proliferation markers in surgical biopsies from normal human postmenopausal breast tissue, it is plausible that mitogenic activity is not identical during therapy with unopposed estrogens versus estrogens combined with progestogens, and is higher during HRT that combines oral conjugated equine estrogens with medroxyprogesterone acetate than during HRT that combines transdermal estradiol and progesterone. It is misleading to put all progestogens in the same bag irrespective of their chemical structure, and, more important, their effect may vary according to whether it is estrone or estradiol that is mainly accumulated in the breast tissue. The hypothesis of progesterone decreasing the proliferative effect of estradiol in the postmenopausal breast remains highly plausible.

DE LIGNIERES B., de VATHAIRE F., FOURNIER S., URBINELLI R., ALLAERT F., LE M.G., KUTTENN F.

Combined hormone replacement therapy and risk of breast cancer in a French cohort study of 3175 women.

Climacteric, 5 (4), 332-340, 2002

(Services cités : Endocrinologie & Médecine de la Reproduction)

The largest-to-date randomized trial (Women's Health Initiative) comparing the effects of hormone replacement therapy (HRT) and a placebo concluded that the continuous use of an oral combination of conjugated equine estrogens (CEE) and medroxyprogesterone acetate (MPA) increases the risk of breast cancer. This conclusion may not apply to women taking other estrogen and progestin formulations, as suggested by discrepancies in the findings of in vitro studies, epidemiological surveys and, mostly, in vivo studies of human breast epithelial cell proliferation showing opposite effects of HRT combining CEE plus MPA or estradiol plus progesterone. To evaluate the risk of breast cancer associated with the use of the latter combination, commonly prescribed in France, a cohort including 3175 postmenopausal women was followed for a mean of 8.9 years (28 367 woman-years). In total, 1739 (55%) of these women were users of one type of estrogen replacement with systemic effect during at least 12 months, any time after the menopause, and were classified as HRT users. Among them, 83% were receiving exclusively or mostly a combination of a transdermal estradiol gel and a progestin other than MPA. Some 105 cases of breast cancer occurred during the follow-up period, corresponding to a mean of 37 new cases per 10 000 women/year. Using multivariate analysis adjusted for the calendar period of treatment, date of birth and age at menopause, we were unable to detect an increase in the relative risk (RR) of breast cancer (RR 0.98, 95% confidence interval (CI): 0.65-1.5) in the HRT users. The RR of breast cancer per year of use of HRT was 1.005 (95% CI 0.97-1.05). These results do not justify early interruption of such a type of HRT, which is beneficial for quality of life, prevention of bone loss and cardiovascular risk profile, without the activation of coagulation and inflammatory protein synthesis measured in users of oral

estrogens.

DE LIGNIERES B.

L'andropause et sa prise en charge chez l'homme âgé.

Presse Médicale, 31 (37 Pt 1), 1750-1759, 2002

(Services cités : Endocrinologie & Médecine de la Reproduction)

THE "ANDROPAUSE": Also called the "male menopause" or "partial androgen deficiency of the aging male" etc., corresponds to the age at which the progressive decrease in androgen activity reaches a pathogenic threshold. Surveys made in various countries since the seventies conclude that testosterone blood levels start to decrease after the age of 25 and that 20 to more than 50% of the male population no longer benefit from optimal androgen stimulation after the age of 60. THE CONSEQUENCES OF HYPOANDROGENISM: The subsequent progressive hypoandrogenism participates in inducing the commonly-observed clinical symptoms (fatigue, morosity, weight loss, lack of interest in sexual activity); the most specific of which is the disappearance or rarification of "automatic" nocturnal or matinal erections. This appears to influence the prostatic pathology and the frequent cardiovascular risk factors, which, far more than a problem of erection, is a major public health issue. A COMPLEX BIOLOGICAL DIAGNOSIS: Added to the abnormalities in production and transport of testosterone are the abnormalities in its metabolisation by the target tissues. These abnormalities are often undetected in present day blood controls and may explain the elevation in the hepatocyte of SHBG synthesis, the relative inhibition of GnRH pulses and LH secretion in the hypothalamus and the pituitary gland and, in the arterial wall (including penile vascularisation) and the prostate, some of the frequent functional and histological disorders. In current practice today, the best approximation of androgen potential is obtained by the comparison of total testosterone concentrations and SHBG, measurements that require relatively reliable standardised kits. THERAPEUTIC CHOICE: Optimal replacement therapy, for some authors, must mimic the physiology of the young man and above all maintain or reinforce the estrogenic effects of testosterone, related to its aromatisation into estradiol and supposedly beneficial for the cardiovascular system and the bone. For other, the androgenic effects, enhanced by the 5 alpha reduction into dihydrotestosterone (DHT), should be reinforced in older men because the estrogenic effects are ineffective on bone and most of the other targets, and are probably pathogenic for the prostate. This debate is extremely important since the various formulations of androgens authorized by the French Medicines agency (AFSSAPS) induce clearly differing estradiol/DHT plasma ratios.

KUTTENN F., GERSON M., de LIGNIERES B.

Effets du traitement hormonal substitutif de la ménopause sur le risque cardiovasculaire.

Plaidoyer pour un essai européen.

Presse Médicale, 31 (10), 468-475, 2002

(Services cités : Endocrinologie & Médecine de la Reproduction)

The increased risk of coronary heart disease (CHD) after menopause is currently attributed to estrogen deficiency. Many epidemiological (case-control and prospective) studies have reported a decreased risk (0.5-0.7) of CHD in postmenopausal women receiving hormone replacement therapy (HRT). However, there are some discordant studies, among which the Framingham study. Moreover, the observational studies were subject to several biases that may have falsely elevated the apparent benefit of estrogen: women taking estrogen tend to be wealthier, more educated and healthier than untreated women. Large randomized clinical trials of secondary (HERS, WEST) or primary (WHI) prevention have not confirmed cardioprotection with HRT.

However, these studies used orally administered estrogens, while the non-oral route of administration is frequently used in Europe. From a biological point of view, estrogen has multiple effects that would be expected to be cardioprotective, including favorable changes in lipids, endothelial function, vascular reactivity and blood flow. However, concerning hemostasis factors, a pro-coagulant effect can be induced by the first pass liver effect of estrogen when administered orally, which is not observed with the non oral routes of administration. In addition, the synthetic progestin medroxy-progesterone acetate (MPA) inhibits the beneficial effects of estrogen on the arterial wall, whereas natural progesterone does not. It is therefore urgent that Europeans undertake a European "HERS study" in order to investigate the possible beneficial effect of non-oral estrogens (administered percutaneously or transdermally) associated with natural progesterone.

MAHIEU-CAPUTO D., MULLER F., JOUVET P., THALABARD J.C., JOUANNIC J.M., NIHOUL-FEKETE C., DUMEZ Y., DOMMERGUES M.

Amniotic fluid beta-endorphin: A prognostic marker for gastroschisis ?

J. Pediat. Surg., 37 (11), 1602-1606, 2002

(Services cités : Chirurgie Pédiatrique, Endocrinologie & Médecine de la Reproduction, Maternité, Biostatistique, URC)

PURPOSE: The aim of this work was to study amniotic fluid beta-endorphin as a potential predictor for postnatal morbidity in gastroschisis. **METHODS:** Beta-endorphin was assayed in 43 amniotic fluid samples from 13 pregnant women with fetal gastroschisis undergoing diagnostic amniocentesis or therapeutic amnioinfusion and compared with 33 controls. Within the gastroschisis group, the authors investigated the relationship between postnatal morbidity and the peak value of amniotic fluid beta-endorphin (AFBE). **RESULTS:** Ten AFBE values in 6 cases of gastroschisis were above the upper limit of the 95% confidence interval derived from controls. Postnatal morbidity was significantly higher when peak AFBE exceeded 10 &mgr;g/L (n = 4 pregnancies) compared with below 5 &mgr;g/L (n = 9 pregnancies), as shown by mean duration of mechanical ventilation (15.2 v 3 days; P =.01), of parenteral feeding (77 v. 18.7 days; P =.04), and of hospitalization (84 v 32.2 days; P =.04). There was no statistically significant association between postnatal morbidity markers and prenatal dilation of fetal bowel. **CONCLUSIONS:** The most severe cases of gastroschisis are associated with high levels of AFBE. The authors speculate that this fetal hormonal response could result from stress or pain caused by prenatal bowel damage. *J Pediatr Surg* 37:1602-1606. Copyright 2002, Elsevier Science (USA). All rights reserved.

MALET C., SPRITZER P., CUMINS C., GUILLAUMIN D., MAUVAIS-JARVIS P., KUTTENN F.

Effect of 4-hydroxytamoxifen isomers on growth and ultrastructural aspects of normal human breast epithelial (HBE) cells in culture.

J. Steroid Biochem. Mol. Biol., 82 (4-5), 289-296, 2002

(Services cités : Endocrinologie & Médecine de la Reproduction)

In the search for a breast cancer prevention strategy which would avoid undesirable effects of orally administered tamoxifen, the percutaneous administration of the highly active metabolite 4OHTamoxifen (4OHTam) has been proposed. Percutaneous 4OHTam penetrates the skin to reach breast tissues. It, thus, avoids the hepatic first pass effect, and offers an optimal local/systemic effect. However, trans-4OHTamoxifen can spontaneously isomerize into the cis-isomer, which may have estrogen agonist action. The aim of this study was to examine the effect

of cis-4OHTam on normal human breast epithelial (HBE) cells in culture. Spontaneous isomerization of trans- into cis-4OHTam occurred within 24-48h, but stabilized rapidly at a trans/cis ratio of 70/30, whether in stock solution, culture medium or cultured cells. The cis-4OHTam did not stimulate HBE cell growth according to histometric cell counts and scanning electron microscopy analysis, but inhibited E(2)-induced cell growth, albeit two to three times less than trans-4OHTam. In conclusion, spontaneous isomerization of trans- to cis-4-OHTam is limited and 4OHTam retains a marked antiestrogenic effect. It may prove to be a useful alternative to tamoxifen in breast cancer prevention, especially if administered percutaneously.

MOREL Y., REY R., TEINTURIER C., NICOLINO M., MICHEL-CALEMARD L., MOWSZOWICZ I., JAUBERT F., FELLOUS M., CHAUSSAIN J.L., CHATELAIN P., DAVID M., NIHOUL-FEKETE C., FOREST M.G., JOSSO N.

Aetiological diagnosis of male sex ambiguity: a collaborative study.

Eur. J. Pediat., 161 (1), 49-59, 2002

(Services cités : Anatomo-Pathologie, Chirurgie Pédiatrique, Endocrinologie & Médecine de la Reproduction)

A collaborative study, supported by the Biomed2 Programme of the European Community, was initiated to optimise the aetiological diagnosis in genetic or gonadal males with intersex disorders, a total of 67 patients with external sexual ambiguity, testicular tissue and/or a XY karyotype. In patients with gonadal dysgenesis or true hermaphroditism, the incidence of vaginal development was 100%, a uterus was present in 60%; uni or bilateral cryptorchidism was seen in nearly all cases of testicular dysgenesis (99%) but in only 57% of true hermaphrodites. Mean serum levels of antimüllerian hormone and of serum testosterone response to chorionic gonadotropin stimulation were significantly decreased in both conditions, by comparison with patients with unexplained male pseudohermaphroditism or partial androgen insensitivity (PAIS). Mutations in the androgen receptor, 90% within exons 2-8, were detected in patients with PAIS. Clinically, a vaginal pouch was present in 90%, cryptorchidism in 36%. In 52% of cases, no diagnosis could be reached, despite an exhaustive clinical and laboratory work-up, including routine sequencing of exons 2-8 of the androgen receptor. By comparison with PAIS, unexplained male pseudohermaphroditism was characterised by a lower incidence of vaginal pouch (55%) and cryptorchidism (22%) but a high incidence of prematurity/intrauterine growth retardation (30%) or mild malformations (14%). Conclusion: reaching an aetiological diagnosis in cases of male intersex is difficult because of the variability of individual cases. Hormonal tests may help to discriminate between partial androgen insensitivity and gonadal dysgenesis/true hermaphroditism but are of less use for differentiating from unexplained male pseudohermaphroditism. Sequencing of exons 2-8 of the androgen receptor after study of testosterone precursors following human chorionic gonadotrophin stimulation is recommended when gonadal dysgenesis and true hermaphroditism can be excluded. [References: 27]

MULLER F., THALABARD J.C., NGO S., DOMMERGUES M.

Detection and false-positive rates of maternal serum markers for Down syndrome screening according to maternal age in women over 35 years of age. A study of the agreement of eight dedicated software packages.

Prenat. Diag., 22 (5), 350-353, 2002

(Services cités : Endocrinologie & Médecine de la Reproduction, Maternité, URC)

Maternal serum markers for trisomy 21 screening (MSS) can be assayed in women ≥ 35 years in an attempt to reduce the need for invasive procedures and thereby avoid their side effects. Our

objective was to compare, in women ≥ 35 , eight different software packages dedicated to second trimester MSS, thus providing reliable data for patient counselling. A simulation study was carried out on 189 sera from women with Down syndrome fetuses and 11 962 sera from mothers of unaffected babies. The first step was to estimate the joint distribution of alpha-fetoprotein (AFP) and free beta-human chorionic gonadotrophin (beta-hCG). The second step was to calculate trisomy 21 detection and false-positive rates for each software according to maternal age (35-45 years), using the usual 1:250 risk threshold. Agreement between software packages was measured using 2x2 kappa coefficients. Detection rates and false-positive rates increased with maternal age. Depending on the software, 57-71% detection rates were achieved at 35 years with 12-18% false-positive rates. At 45 years, 61-100% detection rates were achieved with 66-95% false-positive rates. Up to 39 years, all softwares were concordant (kappa coefficients > 0.75). In the range 35-45 years, false-positive and detection rates increased substantially with maternal age and differences between software packages are observed.

PLU-BUREAU G., THALABARD J.C.

Il n'est pas souhaitable de prescrire un traitement hormonal substitutif à une femme porteuse d'une mastopathie proliférante avec atypie..

Gynecol. Obstet. Fertil., 30 (4), 335-339, 2002

(Services cités : Endocrinologie & Médecine de la Reproduction)

SUN A.J., LIN S.Q., YU W., QIN M.W., CHEN F.L., ZHANG Y., de LIGNIERES B.

Percutaneous estrogen in prevention of early postmenopausal bone loss in Chinese women.

Chin. Med. J., 115 (12), 1790-1795, 2002

(Services cités : Endocrinologie & Médecine de la Reproduction)

Objective To identify the optimal dosage of 17beta-estradiol gel + oral progestin for preventing bone loss in postmenopausal Chinese women. Methods A 3-year open label, randomized, prospective clinical trial was conducted. Sixty healthy women who had been postmenopausal for 1 to 5 years were recruited and divided into following 4 groups: group 1, percutaneous gel 17beta-estradiol (E-2) 1.5 mg/d plus micronized progesterone (MP) 100 mg/d; group 2, percutaneous gel 17beta-estradiol (E-2) 1.5 mg/d plus medroxyprogesterone acetate (MPA) 2 mg/d; group 3, percutaneous gel 17beta-estradiol (E-2) 0.75 mg/d plus micronized progesterone (MP) 100 mg/d; and group 4, percutaneous gel 17beta-estradiol (E-2) 0.75 mg/d plus medroxyprogesterone acetate (MPA) 2 mg/d. Estrogen and progestin were given continuously for 25 days per month. Bone mineral density (BMD) was measured using quantitative computed tomography (QCT) for trabecular bone of L2-5 and dual energy X-ray absorptiometry (DEXA) for L2-4 and hip 5 times during the trial at baseline and at the 6-, 12-, 18-, 24- and 36-month visits. Results Fifty-nine patients (98.3%, 59/60) stayed in the study for 1 year, 56 patients (93.3%, 56/60) for 2 years, and 51 (85%, 51/60) for 3 years. On average, menopausal symptoms were relieved by 80% after 6 months of treatment. By the 24th month, the mean increase in BMD ranged from 4.3% to 7.5% in trabecular bone; and by the 36th month, it ranged from 4.2% to 6.2% in L2-4 and 1.61% to 3.77% in the neck. There were significant difference after treatment ($P < 0.05$). Among the four groups, no significant difference ($P > 0.05$) was found in improvement of symptoms, levels of bone markers or BMD. Conclusion A daily dose of estradiol gel, either 0.75 mg or 1.5 mg, is effective in preventing early postmenopausal, bone loss and relieving menopausal symptoms. After 3-year treatment, spinal BMD could increase steadily, so does hip BMD, especially in the first 2 years.

2001

AUDIBERT F., DOMMERGUES M., BENATTAR C., TAIEB J., THALABARD J.C., FRYDMAN R.

Screening for down syndrome using first-trimester ultrasound and second-trimester maternal serum markers in a low-risk population: a prospective longitudinal study.

Ultrasound Obstet. Gynecol., 18 (1), 26-31, 2001

(Services cités : Biostatistique, Endocrinologie & Médecine de la Reproduction, URC)

Objectives To compare nuchal translucency and second-trimester maternal serum measurements as alternative methods of antenatal screening for Down syndrome in a low-risk population and to evaluate the consequence of combining the results in the estimation of risk. **Design** In a consecutive series of 4130 women aged less than 38 years with a singleton pregnancy, we examined both the detection rate of Down syndrome by nuchal translucency measurement at 10-14 weeks and maternal serum screening by human chorionic gonadotrophin and alpha-fetoprotein at 14-18 weeks. Women with a nuchal translucency measurement of greater than or equal to 3 mm and women with a maternal serum screening-derived risk greater than or equal to 1/250 were recommended to have amniocentesis. A second-trimester detailed ultrasound scan was also performed in all women. The outcome of all pregnancies was recorded prospectively and the detection rate and false-positive rate of different screening strategies were retrospectively analyzed. **Results** Out of the 4130 pregnancies that were followed (mean maternal age, 30.1 years), 12 cases of Down syndrome were observed (0.28%), all detected prenatally. Seven of 12 cases had a nuchal translucency measurement of greater than or equal to 3 mm (58%), and six out of 10 cases with available maternal serum screening had a calculated risk of greater than or equal to 1/250 (60%). Four of the five Down syndrome cases with a nuchal translucency measurement of < 3 mm were detected by subsequent maternal serum screening. At a threshold giving 5% of positive tests, the sensitivity of nuchal translucency, maternal serum screening and combined risk screening were 75%, 60% and 90%, respectively. **Conclusions** In screening for Down syndrome, an approach which combines the results from first-trimester nuchal translucency and second-trimester biochemistry, is effective and increases the detection rate compared to the use of any single test. However, this strategy is likely to raise the false-positive rate and the interpretation of maternal serum screening-derived risk should be combined with the first-trimester nuchal translucency measurement. [References: 23]

BLANCHERE M., MESTAYER C., SAUNIER E., BROSHUIS M., MOWSZOWICZ I.

Transforming growth factor beta in the human prostate: its role in stromal-epithelial interactions in non-cancerous cell culture.

Prostate, 46 (4), 311-318, 2001

(Services cités : Endocrinologie & Médecine de la Reproduction)

BACKGROUND. Stromal-epithelial interactions play a critical role in prostate development, but the precise mechanisms are still unknown. Transforming growth factor-beta (TGF beta) could be a potential mediator of these interactions, but there is as yet no clear demonstration of its role. **METHODS.** Separate cultures and co-cultures of fibroblasts and epithelial human prostate cells were performed. We measured TGF beta1 and TGF beta2 secretion by specific ELISA assay, cell growth by DNA assay, and TGF beta type II receptor expression by RT-PCR. **RESULTS.** Co-culture resulted in a 20% inhibition of epithelial cell growth, similar to that obtained after TGF beta treatment (2 ng/ml for 48 hr), but without affecting fibroblast proliferation. This was accompanied by a five- to six-fold increase in epithelial TGF beta2 secretion. **CONCLUSIONS.** These results demonstrate for the first time that TGF beta2 secretion by prostate epithelial cells is

under the direct control of a diffusible factor secreted by fibroblasts. They emphasize the role of TGF beta in stromal-epithelial interactions. *Prostate* 46:311-318, 2001. (C) 2001 Wiley-Liss, Inc. [References: 39]

DE BAERE E., DIXON M.J., SMALL K.W., JABS E.W., LEROY B.P., DEVRIENDT K., GILLEROT Y., MORTIER G., MEIRE F., VAN MALDERGEM L., COURTENS W., HJALGRIM H., HUANG S., LIEBAERS I., VAN REGEMORTER N., TOURAINE P., PRAPHANPHOJ V., VERLOES A., UDAR N., YELLORE V., CHALUKYA M., YELCHITS S., de PAEPE A., KUTTENN F., FELLOUS M., VEITIA R., MESSIAEN L. Spectrum of *foxl2* gene mutations in blepharophimosis-ptosis-epicanthus inversus (bpes) families demonstrates a genotype-phenotype correlation.

Hum. Mol. Genet., 10 (15), 1591-1600, 2001

(Services cités : Endocrinologie & Médecine de la Reproduction, U344)

Mutations in *FOXL2*, a forkhead transcription factor gene, have recently been shown to cause blepharo-phimosis-ptosis-epicanthus inversus syndrome (BPES) types I and II, a rare genetic disorder. In BPES type I a complex eyelid malformation is associated with premature ovarian failure (POF), whereas in BPES type II the eyelid defect occurs as an isolated entity. In this study, we describe the identification of novel mutations in the *FOXL2* gene in BPES types I and II families, in sporadic BPES patients, and in BPES families where the type could not be established. In 67% of the patients studied, we identified a mutation in the *FOXL2* gene. In total, 21 mutations (17 of which are novel) and one microdeletion were identified. Thirteen of these *FOXL2* mutations are unique. In this study, we demonstrate that there is a genotype-phenotype correlation for either types of BPES by the finding that mutations predicted to result in a truncated protein either lacking or containing the forkhead domain lead to BPES type I. In contrast, duplications within or downstream of the forkhead domain, and a frameshift downstream of them, all predicted to result in an extended protein, cause BPES type II. In addition, in 30 unrelated patients with isolated POF no causal mutations were identified in *FOXL2*. Our study provides further evidence that *FOXL2* haploinsufficiency may cause BPES types I and III by the effect of a null allele and a hypomorphic allele, respectively. Furthermore, we propose that in a fraction of the BPES patients the genetic defect does not reside within the coding region of the *FOXL2* gene and may be caused by a position effect. [References: 36]

DENEUX C., TARDY V., DIB A., MORNET E., BILLAUD L., CHARRON D., MOREL Y., KUTTENN F.

Phenotype-genotype correlation in 56 women with nonclassical congenital adrenal hyperplasia due to 21-hydroxylase deficiency.

J. Clin. Endocrinol. Metabol., 86 (1), 207-213, 2001

(Services cités : Endocrinologie & Médecine de la Reproduction)

Complete analysis of the *CYP21* gene was performed in 56 unrelated French women with symptomatic nonclassical congenital adrenal hyperplasia. The mutational spectrum and the phenotype-genotype correlation were examined. The overall predominant mutation was V281L, which was present on 51% of alleles and in 80% of women. Three novel mutations were found: L317M, R435C, and a 5'-end gene conversion. Sixty-three percent of the women were carrying a severe mutation of the *CYP21* gene, and hence risk giving birth to children with a classical form of the disease. In such cases, screening for heterozygosity in the partner is crucial. Potential genotype/phenotype correlations were examined by classifying the patients into three groups according to the *CYP21* allelic combinations: A (mild/mild), B (mild/severe), and C

(severe/severe). Primary amenorrhea was more frequent, and mean basal and stimulated 17-hydroxyprogesterone levels were higher in compound heterozygotes for mild and severe mutations (group B) compared with women with two mild mutations (group A), but there was a considerable overlap for individual values. Surprisingly, in two women, a severe mutation was found on both alleles (group C). Therefore, the phenotype cannot be accurately predicted from the genotype. Variability in phenotypic expression may be conditioned by mechanisms other than genetic heterogeneity at the CYP21 locus. [References: 38]

KUTTENN F., GERSON M.

Hormone replacement therapy of the menopause, heart and blood vessels.

Arch. Mal. Coeur Vaisseaux, 94 (7), 685-689, 2001

(Services cités : Endocrinologie & Médecine de la Reproduction)

The incidence of coronary heart disease (CHD) is lower in premenopausal women than in men and post-menopausal women of the same age. The higher CHD rate after menopause is currently attributed to estrogen deficiency: many epidemiological (case-control and prospective) studies have reported a reduced risk (0.5-0.63) of CHD in post-menopausal women receiving hormone replacement therapy (HRT). Moreover, estrogens have multiple effects that would be expected to be cardioprotective, including favorable changes in lipids, endothelial function, vascular reactivity and blood flow. However, the observational studies are subject to several biases that could falsely elevate the apparent benefit of estrogens: women taking estrogens tend to be wealthier, more educated and healthier than untreated women. The american HERS (Heart and Estrogen-progestin Replacement Study; 2.763 women) is a large multicenter randomized study of secondary prevention, designed to evaluate the efficacy of HRT. Results are disappointing, since no reduced risk was observed, and the risk of CHD was even higher in women receiving HRT during the first year: 1.52 (CI 95% 1.01-2.29). In HERS study, the treatments consisted of conjugated equine estrogens and the synthetic progestin medroxyprogesterone acetate (MPA) which are rarely used in Europe. Indeed, the effects of HRT are not equivalent depending on the dose, the route of administration, the type of progestogen. It should be emphasized that MPA, contrarily to progesterone, inhibits the beneficial effect of estrogens on lipids and experimental atherosclerosis. The route of administration of estrogens is also involved : estrogens alter hemostasis factors, and when orally administered, they have a first pass liver effect, which favors hypercoagulability. It is therefore urgent that Europeans undertake a European "HERS study" in order to investigate the possible beneficial effect of non oral estrogens (gel or patch) associated with natural progesterone. [References: 37]

MALET C., FIBLEUIL F., MESTAYER C., MOWSZOWICZ I., KUTTENN F.

Estrogen and antiestrogen actions on transforming growth factor beta (tgf beta) in normal human breast epithelial (hbe) cells.

Mol. Cell. Endocrinol., 174 (1-2), 21-30, 2001

(Services cités : Endocrinologie & Médecine de la Reproduction)

We have previously shown that estradiol (E2) increases the growth of normal human breast epithelial (HBE) cells and the antiestrogen 4-hydroxytamoxifen (4-OHT) inhibits estrogen-induced proliferation. These effects of estrogens and antiestrogens on proliferation have also been well documented in breast cancer cells. One mechanism for the antiproliferative effects of antiestrogens is the stimulation of TFG beta in hormone-dependent MCF-7 and T47D cells. The role of this inhibitory growth factor in normal human breast cells has not been well studied. Accordingly, we measured the amounts of total and active TGF beta1 and TGF beta2 by specific

E-max immunoassay (EIA) in culture medium from normal breast cells (epithelial and fibroblasts) and from various ER - and ER + breast cancer cell lines. We established that HBE cells are sensitive to the antiproliferative effect of TGF betas, and studied the effect of E2 and 4-OHT, alone or in combination, on the secretion and activation of TGF betas by HBE cells. HBE cells secrete TGF beta1 and even more TGF beta2, and are sensitive to these factors. However, in contrast to MCF-7 cells, TGF beta secretion in normal breast cells is not regulated by E2 and 4-OHT. (C) 2001 Elsevier Science Ireland Ltd. All rights reserved. [References: 53]

TOURAINÉ F., PLU-BUREAU G., BEJI C., MAUVAIS-JARVIS F., KUTTENN F.

Long-term follow-up of 246 hyperprolactinemic patients.

Acta Obstet. Gynecol. Scand., 80 (2), 162-168, 2001

(Services cités : Endocrinologie & Médecine de la Reproduction, U344, URC)

Background. We wanted to evaluate the very long-term effects of bromocriptine on prolactin (PRL) levels and pituitary tumor size in a large cohort of hyperprolactinemic patients. **Methods.** We conducted a retrospective cohort study in the Department of Endocrinology from Necker Hospital in Paris, France. Two hundred and forty-six patients consulted primarily for menstrual disorders, with diagnosis of hyperprolactinemia. Patients were followed-up for 99.9+/-3.6 months. One hundred and ninety-one were treated with bromocriptine, 32 underwent surgery, and 23 received no treatment. **Results.** The mean initial plasma PRL level was 135.0+/-20.2 ng/ml. Presence of an adenoma was detected in 60% of our patients and comprised a microadenoma in 64% of cases. Compared to oligomenorrheic women, amenorrheic patients had significantly higher levels of PRL and larger pituitary tumor size. In the bromocriptine group, PRL levels decreased from 99.6+/-7.9 to 20.0+/-1.5 ng/ml (p=0.00001). The medical treatment was associated with disappearance of the adenoma in 45% of the women and with stabilization of pituitary tumor size in 40% of patients. Surgery led to disappearance of the adenoma in almost all cases, but failed to definitively cure hyperprolactinemia. **Conclusion.** In this large-scale retrospective study, the medical treatment of mild hyperprolactinemia was shown to be effective and sufficient after 9 years of follow-up. [References: 43]

TOURAINÉ P., PLU-BUREAU G., BERESSI N., DECQ P., THALABARD J.C., KUTTENN F.

Resumption of luteinizing hormone pulsatility and hypogonadotropic hypogonadism after endoscopic ventriculocisternostomy in a hydrocephalic patient.

Fert. Steril., 76 (2), 390-393, 2001

(Services cités : Endocrinologie & Médecine de la Reproduction, U344, URC)

OBJECTIVE: To study gonadotropin pulsatility before and after surgical cure of hydrocephalus. **DESIGN:** Case report. **SETTING:** Department of Endocrinology and Centre d'Investigations Cliniques, Necker Hospital, Paris, France. **PATIENT(S):** A 29-year-old woman who presented with secondary amenorrhea. **INTERVENTION(S):** The patient underwent an endoscopic ventriculocisternostomy that led to restoration of normal menses and resolution of hypogonadism. **MAIN OUTCOME MEASURE(S):** A gonadotropin pulse study was performed before and 2 and 5 months after surgery. **RESULT(S):** No LH pulse was observed before surgery. Emergence of pulsatility was observed 2 months after surgery, and pulses became clearly individualized after 5 months. **CONCLUSION(S):** This observation strongly suggests that amenorrhea, in case of chronic hydrocephalus, is indeed due to a hypothalamic dysfunction of the GnRH pulse generator.

2000

BENIFLA J.L., LETUR-KONIRSCH H., COLLIN G., DEVAUX A., KUTTENN F., MADELENAT P., BRUN-VEZINET F., FELDMANN G.

Safety of cryopreservation straws for human gametes or embryos: a preliminary study with human immunodeficiency virus-1.

Hum. Reprod., 15 (10), 2186-2189, 2000

(Services cités : Endocrinologie & Médecine de la Reproduction)

The aim of this preliminary experimental study was to test the stability of cryopreservation straws to human immunodeficiency virus-1 (HIV-1). Three kinds of straws were tested: four polyvinyl chloride (PVC), four polyethylene terephthalate glycol (PETG) and 20 high-security ionomeric resin (IR). The PVC and PETG straws were sealed ultrasonically, and the IR straw by thermosoldering. Each sealed straw was cut in half to produce two demistraws and then filled with 100 µl of HIV-1-containing supernatant (reverse transcriptase activity: 15 000 c.p.m./50 µl). The unsealed cotton end of PVC and PETG straws and the two halves of the IR straws (cotton and plastic plug ends) were tested. Each demi-straw was two-thirds submerged in RPMI medium at 37 degrees C, and RPMI samples were withdrawn on days 3, 7 and 11. Viral RNA was extracted from the medium and then amplified by reverse transcriptase-polymerase chain reaction (RT-PCR) followed by nested PCR using primers specific to HIV-1 protease. On day 7, no HIV-1 RNA was detected in any of the different samples of medium that had surrounded the unsealed PVC and PETG straws with cotton ends, but three IR specimens were positive. On day 11, PVC and PETG remained negative but HIV-1 RNA was detected in RPMI samples for two more IR demi-straws (n = 5). In conclusion, under these experimental conditions (at 37 degrees C), the unsealed cotton end PVC, PETG and thermosoldered cotton end IR demi-straws appeared to be safe for HIV-1, while IR straws, sealed or unsealed with a plastic plug and with unsealed cotton ends, leaked. [References: 21]

BERNIER M.O., PLU BUREAU G., BOSSARD N., AYZAC L., THALABARD J.C.

Breastfeeding and risk of breast cancer: a metaanalysis of published studies.

Hum. Reprod. Update, 6 (4), 374-386, 2000

(Services cités : Endocrinologie & Médecine de la Reproduction, Biostatistique, URC)

Evidence in favour of an association of breastfeeding with a breast cancer risk reduction remains limited and inconsistent. To evaluate the relation between breastfeeding and breast cancer, a meta-analysis based on a review of the literature was carried out, using as variables ever/never breastfeeding and duration of breastfeeding. Menopausal status at the time of diagnosis of breast cancer was considered to be a potential effect modifier. Only case-control studies could be included in the final analysis. A slight but significant decreased risk of breast cancer was observed in ever breastfeeding, compared with never breastfeeding parous women, using both the fixed and random-effect models. This decrease was more pronounced in non-menopausal women at the time of diagnosis of breast cancer and in long-term breastfeeding women. Hence, breastfeeding appeared to be a protective factor but was of small magnitude compared with other known risk factors for breast cancer. Whether this result should imply a modification in the attitude of both health care providers and women towards breastfeeding, which represents one of the few identified protective factors which is under the control of the mother, and is thus (theoretically) modifiable, remains questionable. [References: 84]

MALET C., SPRITZER P., GUILLAUMIN D., KUTTENN F.

Progesterone effect on cell growth, ultrastructural aspect and estradiol receptors of normal human

breast epithelial (hbe) cells in culture.

J. Steroid Biochem. Mol. Biol., 73 (3-4), 171-181, 2000

(Services cités : Endocrinologie & Médecine de la Reproduction)

The stimulating effect of estradiol (E2) on breast cell growth is well documented. However, the actions of progesterone (P) and its derivatives remain controversial. Additional information is therefore necessary. On a culture system of normal human breast epithelial (HBE) cells, we observed an inhibitory effect on cell growth of a long-term P treatment (7 days) in the presence or absence of E2, using two methods: a daily cell count providing a histometric growth index, and [H-3]-thymidine incorporation during the exponential phase of cell growth. A scanning electron microscopy study confirmed these results. Cells exhibited a proliferative appearance after E2 treatment, and returned to a quiescent appearance when P was added to E2. In both studies, P proved to be as efficient as the synthetic progestin R5020. Moreover, the immunocytochemical study of E2 receptors indicated that E2 increases its own receptor level whereas P and R5020 have the opposite effect, thus limiting the stimulatory effect of E2 on cell growth. In the HBE cell culture system and in long-term treatment, P and R5020 appear predominantly to inhibit cell growth, both in the presence and absence of E2. (C) 2000 Elsevier Science Ltd. All rights reserved. [References: 36]

PLU BUREAU G., BOSSARD N., THALABARD J.C.

Oral contraception and genetic factors in breast cancer: characteristics and limits of case-only studies.

Rev. Épidémiol. Santé Publ., 48 (3), 294-303, 2000

(Services cités : Endocrinologie & Médecine de la Reproduction, URC)

The analysis of the interaction between environmental and genetic factors is a matter of increasing interest in cancerology. More particularly the discovery of the BRCAx family and the high cumulated incidence of familial breast cancers related to mutations of these proteins raised the issue of the differential effect of long term and/or early exposure to oral contraceptives in the presence of these mutations. The classical case-control design assumes the presence of a control group, which can be sometimes difficult to obtain from both the technical and ethical points of view. Case-only or case-case studies, which are based only on series of cases, making them apparently attractive, have been proposed to analyze more specifically the interaction term. The aim of the present paper is to review and discuss the methodological basis and main assumptions of the case-only design, and their applicability to breast cancer studies. The measure of the interaction between an environmental factor and a susceptibility genetic factor differs in an important aspect from the measure of the association between an environmental factor and acquired tumoral genetic factor; this aspect is reminded. [References: 27]

PLU-BUREAU G.

Hormone replacement therapy after breast cancer.

J. Gynecol. Obst. Biol. Reprod., 29 (3), 292-294, 2000

(Services cités : Endocrinologie & Médecine de la Reproduction)

The use of hormone replacement therapy after breast cancer is controversial. The published epidemiological studies shown no association between HRT and recurrence of breast cancer. However, the interpretation of these results is very difficult because of numerous biases in this context of observational studies. Prospective randomised trials that examine the effects of HRT in women with a history of breast cancer are currently underway in United States and in Scandinavia. The markov approach is interesting for determining the global risk-benefit balance

of HRT and for visualising the effects of different risk levels on breast cancer and to compare different therapeutic strategies. Hot flushes and functional symptoms represent the essential unresolved problem of these women. The results of all these will provide valuable data to define efficient and non-deleterious approaches to improve the well being of the women after breast cancer. [References: 10]

SILBERSTEIN S.D., de LIGNIERES B.

Migraine, menopause and hormonal replacement therapy.

Cephalalgia, 20 (3), 214-221, 2000

(Services cités : Endocrinologie & Médecine de la Reproduction)

VIGOUROUX C., MAGRE J., VANTYGHM M.C., BOURUT C., LASCOLS O., SHACKLETON S., LLOYD D.J., GUERCI B., PADOVA G., VALENSI P., GRIMALDI A., PIQUEMAL R., TOURAINE P., TREMBATH R.C., CAPEAU J.

Lamin a/c gene - sex-determined expression of mutations in dunnigan-type familial partial lipodystrophy and absence of coding mutations in congenital and acquired generalized lipoatrophy.

Diabetes, 49 (11), 1958-1962, 2000

(Services cités : Endocrinologie & Médecine de la Reproduction)

Missense mutations of the lamin A/C gene, LMNA, have been recently identified in Dunnigan-type familial partial lipodystrophy (FPLD), which belongs to a heterogeneous group of rare disorders affecting adipose tissue distribution and metabolism. In this study, we sequenced the LMNA coding region from patients presenting with FPLD or other forms of lipodystrophy. We identified two heterozygous mutations in exon 8, R482W and R482Q, in FPLD patients (six families and one individual) with various clinical presentations. In addition, we found a novel heterozygous mutation (R584H) in exon 11, encoding specifically the lamin A isoform, in a patient with typical FPLD. Clinical and biochemical investigations in FPLD patients revealed that the expression and the severity of the phenotype were markedly dependent on sex, with female patients being more markedly affected. In subjects with generalized lipoatrophy, either congenital (13 case subjects) or acquired (14 case subjects), or Barraquer-Simon syndrome (2 case subjects), the entire LMNA coding sequence was normal. Although FPLD mutations are predominantly localized in exon 8 of LMNA, the finding of a novel mutation at codon 584, together with the R582H heterozygous substitution recently described, confirms that the C-terminal region specific to the lamin A isoform is a second susceptibility region for mutations in FPLD. [References: 20]

1999

BAFOUNTA M.L., BEAUCHET A., POISSON-SALOMON A.S., SAIAG P.

Impact of French consensus conference concerning the follow-up of patients surgically treated for stage I melanoma among French dermatologists and oncologists.

Ann. Dermatol. Vénérolog., 126 (11), 795-800, 1999

(Services cités : Endocrinologie Adulte)

Introduction. Our aim was to assess the impact of the 1995 French Consensus Conference (CC) on the follow up of patients surgically treated for WHO stage I (AJCC/UICC stages IA to IIB) melanoma, two years after the recommendations had been provided to French dermatologists and oncologists by direct mailing and literature. Prior to this CC, a study of French dematologists' intentions of practice and dermatologists and oncologists effective practices had revealed a lack of homogeneity and the prescription of many paraclinical tests.

BAFOUNTA M.L., BEAUCHET A., POISSON-SALOMON A.S., SAIAG P.

Implication of French general practitioners in the follow-up of patients surgically treated for a stage I melanoma.

Ann. Dermatol. Vénéréolog., 126 (11), 801-803, 1999

(Services cités : Endocrinologie Adulte)

Introduction, Our aim was to assess the implication of French general practitioners in the follow up of patients surgically treated for stage I melanoma 2 years after the publication of the 1995 French consensus conference recommendations.

BRY-GAULLARD H., TOURAINE P., MAMZER-BRUNEEL M.F., SIMOES-VAZ A., KUTTENN F., LEGENDRE C.

Complete regression of a major hyperprolactinaemia after renal transplantation.

Nephrol. Dialysis Transplant., 14 (2), 466-468, 1999

(Services cités : Endocrinologie Adulte, Transplantation)

DE LIGNIERES B.

Oral micronized progesterone.

Clin. Ther., 21 (1), 41-60; di, 1999

(Services cités : Endocrinologie Adulte)

This review sought to examine the rationale for selecting an oral micronized progesterone formulation rather than a synthetic progestin for some of the main indications for progestogens. Unopposed estrogen use is associated with a high risk (relative risk, 2.1 to 5.7) of endometrial hyperplasia and adenocarcinoma, and it has been understood for some time that a progestogen must be added for at least 10 to 14 days per month to prevent these effects. However, the most commonly used synthetic progestins, norethisterone and medroxyprogesterone acetate, have been associated with metabolic and vascular side effects (eg, suppression of the vasodilating effect of estrogens) in both experimental and human controlled studies. All comparative studies to date conclude that the side effects of synthetic progestins can be minimized or eliminated through the use of natural progesterone, which is identical to the steroid produced by the corpus luteum. The inconvenience associated with the use of injectable, rectal, or vaginal formulations of natural progesterone can be circumvented by using orally administered micronized progesterone. The bioavailability of micronized progesterone is similar to that of other natural steroids, and interindividual and intraindividual variability of area under the curve is similar to that seen with synthetic progestins. A clear dose-ranging effect has been demonstrated, and long-term protection of the endometrium has been established. Micronized progesterone has been used widely in Europe since 1980 at dosages ranging from 300 mg/d (taken at bedtime) 10 days a month for women wishing regular monthly bleeding to 200 mg 14 days a month or 100 mg 25 days a month for women willing to remain amenorrheic. This therapy is well tolerated, with the only specific side effect being mild and transient drowsiness, an effect minimized by taking the drug at bedtime. The prospective, comparative Postmenopausal Estrogens/Progestin Intervention trial has recommended oral micronized progesterone as the first choice for opposing estrogen therapy in nonhysterectomized postmenopausal women. [References: 86]

DE LIGNIERES B.

Endometrial hyperplasia. risks, recognition and the search for a safe hormone replacement regimen.

J. Reprod. Med., *44* (2 Suppl), 191-196, 1999

(Services cités : Endocrinologie Adulte)

Endometrial hyperplasia is an acknowledged risk of unopposed estrogen replacement therapy (ERT). Several categories of hyperplasia are now recognized, among which atypical complex hyperplasia is the most likely to progress to carcinoma. Adding a synthetic progestin or natural progesterone to estrogen therapy has been shown to decrease or eliminate the endometrial risk associated with ERT. However, the addition of synthetic progestins has been associated with uncomfortable side effects, reversal of some of the cardiovascular and metabolic benefits of estrogen, and unwanted bleeding. The use of natural micronized progesterone in lieu of synthetic progestins alleviates the former two drawbacks, while careful scheduling of estrogen and progesterone dosing can eliminate the latter. In Europe, where natural progesterone has been in use for some time, a cyclic combined regimen comprising estrogen and 100 mg micronized progesterone administered on days 1-25 has been shown to provide endometrial safety, absence of bleeding and a high rate of tolerability.

DENEUX C., TOURAINE P., KUTTENN F.

Physiopathology of gonadotropic control of gametogenesis.

Ann. Biol. Clin., *57* (3), 318-321, 1999

(Services cités : Endocrinologie Adulte)

EISINGER F., ALBY N., BREMOND A., DAUPLAT J., ESPIE M., JANIAUD P., KUTTENN F., LEBRUN J.P., LEFRANC J.P., PIERRET J., SOBOL H., STOPPA-LYONNET D., THOUVENIN D., TRISTANT H., FEINGOLD J.

Recommendations for medical management of hereditary breast and ovarian cancer: the Inserm-FNCLCC ad hoc committee.

Bull. Cancer, *86* (3), 307-313, 1999

(Services cités : Endocrinologie Adulte)

Background: Almost 10% of breast and ovarian cancer are inherited, and the majority are linked to BRCA1 and BRCA2 germline mutations. Despite the uncertainty, consensus guidelines were defined to assist practitioners and patients' decisions about the health care decisions to be made. Methodology: The ad hoc committee consisted of 14 experts designated by the French National Institute for Health and Medical Research. They all attended eleven workshops at which a systematic analytical review of more than 3,500 articles was carried out. Five additional experts critically analyzed the first version of the report. Process: Two thresholds were defined on a probability scale giving the risk of developing breast or ovarian cancer, to serve as a means of deciding as whether an intervention is worthwhile. The first threshold is that above which an intervention can be envisaged or recommended; the second is that under which an intervention can be ruled out; between the two, the decision has to be made on a case-by-case basis. Screening and preventive strategies analyzed: About breast cancer: 1) hormonal interventions; 2) primary prevention (diet, family planning and chemoprevention); 3) screening (breast self-examination, clinician breast examination, tumor markers, imaging); 4) prophylactic mastectomy. About ovarian cancer: 1) hormonal stimulation; 2) screening (clinical screening, ultrasound and tumor markers); 3) prophylactic oophorectomy. Main conclusions: With each strategy the following points were dealt with: the information to be delivered to the consultand, the procedure and the indications. The committee's opinion about BRCA mutation screening is that population-based or even large scale implementation are not justified. The committee feels that specific management is indispensable and advocates the use of defined and evaluated procedures, and involvement in

clinical trials. [References: 25]

GERSON M., KUTTENN F.

Stakes and perspectives of the treatment for menopause.

M/S - Méd. Sci., 15 (6-7), 873-879, 1999

(Services cités : Endocrinologie Adulte)

Hormone replacement therapy (HRT) represents a major break-through for post-menopausal women : better quality of life and protection against osteoporosis are widely acknowledged benefits, while cardiovascular protection and prevention of dementia are appealing questions to which current large clinical trials should provide answers in the future. The perspectives imply: (1) optimization of HRT, in terms of the drugs, the route of administration, and adjustable dosage forms; (2) alternatives are essential in case of contraindications to HRT, Neither androgens, nor phytoestrogens are acceptable alternatives to HRT. Selective estrogen receptor modulators (SERMs) are the most original and promising agents today. Raloxifen has been the most, widely studied, yet little is known about its potential adverse effects, and it seems more appropriate for osteoporosis prevention than a true HRT alternative. Lastly, the social and economic aspects also need to be dealt with: compliance with HRT must be improved in, the future, in particular by offering constantly up-dated continuous medical education, and by optimizing the cost/efficacy ratio of HRT in order to make it readily available. [References: 49]

PLU-BUREAU G., LE M.G., THALABARD J.C., SITRUK-WARE R., MAUVAIS-JARVIS P.

Percutaneous progesterone use and risk of breast cancer: results from a French cohort study of premenopausal women with benign breast disease.

Cancer Detection Prev., 23 (4), 290-296, 1999

(Services cités : Endocrinologie Adulte)

Percutaneous progesterone topically applied on the breast has been proposed and widely used in the relief of mastalgia and benign breast disease by numerous gynecologists and general practitioners. However, its chronic use has never been evaluated in relation to breast cancer risk. The association between percutaneous progesterone use and the risk of breast cancer was evaluated in a cohort study of 1150 premenopausal French women with benign breast disease diagnosed in two breast clinics between 1976 and 1979. The follow-up accumulated 12,462 person-years. Percutaneous progesterone had been prescribed to 58% of the women. There was no association between breast cancer risk and the use of percutaneous progesterone (RR = 0.8; 95% confidence interval 0.4-1.6). Although the combined treatment of oral progestogens with percutaneous progesterone significantly decreased the risk of breast cancer (RR = 0.5; 95% confidence interval 0.2-0.9) as compared with nonusers, there was no significant difference in the risk of breast cancer in percutaneous progesterone users versus nonusers among oral progestogen users. Taken together, these results suggest at least an absence of deleterious effects caused by percutaneous progesterone use in women with benign breast disease. [References: 24]

PLU-BUREAU G., TOURAINE P., MAUVAIS-JARVIS P.

Interactions between estradiol and progesterone in normal breast - Implications for mammary carcinogenesis.

Contemp. Endocrinol., 11 21-37, 1999

(Services cités : Endocrinologie Adulte)

PLU-BUREAU G.

Cardiovascular risk and hormone replacement therapy in menopause.

Thérapie, 54 (3), 375-380, 1999

(Services cités : Endocrinologie Adulte)

Cardiovascular risk associated with hormone replacement therapy (HRT) has been analysed by large epidemiological studies. This treatment has different effects depending on the type of vessel (venous or arterial) or site (heart or brain). The several meta-analyses which have been published conclude that there is a significant decrease of about 30 to 50 per cent in ischaemic heart disease associated with HRT. In addition, oestrogen replacement therapy is associated with a 25 per cent decrease in cardiovascular mortality. A recent meta-analysis has analysed the effect of HRT on cerebrovascular risk. A significant 20 per cent increase in ischaemic stroke associated with the use of HRT has been shown. However, a protective association of about 30 per cent has been observed in haemorrhagic stroke with HRT use. Recent epidemiological studies have suggested an increased risk of thromboembolic disease associated with HRT. The results of a randomized blind placebo-controlled secondary prevention trial have recently been published. In this clinical trial, women who receive oestrogen (0.625 mg conjugated equine oestrogen daily) plus progestin (2.5 mg medroxyprogesterone acetate daily) therapy did not experience a reduction in overall risk of non-fatal myocardial infarction and cardiovascular heart disease death. This treatment also significantly increases the rate of thromboembolic events. Other randomized trials of HRT for primary prevention are scheduled to yield results by 2000 or 2005. All these studies have been conducted essentially in Anglo-Saxon countries and have analysed the effects of conjugated equine oestrogens alone or combined with medroxyprogesterone acetate. This treatment is not currently used in France. But no randomized trials are under way with the HRT common in France (transdermic oestrogen combined with natural progesterone). The effects of this treatment on cardiovascular disease remain unknown. [References: 23]

REY R.A., BELVILLE C., NIHOUL-FEKETE C., MICHEL-CALEMARD L., FOREST M.G., LAHLOU N., JAUBERT F., MOWSZOWICZ I., DAVID M., SAKA N., BOUVATTIER C., BERTRAND A.M., LECOINTRE C., SOSKIN S., CABROL S., CROSNIER H., LEGER J., LORTAT-JACOB S., NICOLINO M., RABL W., TOLEDO S.P.A., BAS F., GOMPEL A., CZERNICHOW P., CHATELAIN P., ET A.L.

Evaluation of gonadal function in 107 intersex patients by means of serum antimullerian hormone measurement.

J. Clin. Endocrinol. Metabol., 84 (2), 627-631, 1999

(Services cités : Chirurgie Pédiatrique, Anatomopathologie, Biochimie Médicale, Endocrinologie et Croissance, Endocrinologie Adulte)

Fetal male sexual differentiation is driven by two testicular hormones: testosterone (synthesized by interstitial Leydig cells) and antimullerian hormone (AMH; produced by Sertoli cells present in the seminiferous tubules). Intersex states result either from gonadal dysgenesis, in which both Leydig and Sertoli cell populations are affected, or from impaired secretion or action of either testosterone or AMH. Until now, only Leydig cell function has been assessed in children with ambiguous genitalia, by means of testosterone assay.

TOURAINÉ P., BEAU I., GOUGEON A., MEDURI G., DESROCHES A., PICHARD C., DETOEUF M., PANIEL B., PRIEUR M., ZORN J.R., MILGROM E., KUTTENN F., MISRAHI M.

New natural inactivating mutations of the follicle-stimulating hormone receptor: correlations

between receptor function and phenotype.

Mol. Endocrinol., 13 (11), 1844-1854, 1999

(Services cités : Endocrinologie Adulte)

Premature ovarian failure occurs in almost 1% of women under age 40. Molecular alterations of the FSH receptor (FSHR) have recently been described. A first homozygous mutation of the FSHR was identified in Finland. More recently, we described two new mutations of the FSHR in a woman presenting a partial FSH-resistance syndrome (patient 1). We now report new molecular alterations of the FSHR in another woman (patient 2) who presented at the age of 19 with primary amenorrhea contrasting with normal pubertal development. She had high plasma FSH, and numerous ovarian follicles up to 3 mm in size were evidenced by ultrasonography. Histological and immunohistochemical examination of ovarian biopsies revealed the presence of a normal follicular development up to the antral stage and disruption at further stages. DNA sequencing showed two heterozygous mutations: Asp224Val in the extracellular domain and Leu601Val in the third extracellular loop of FSHR. Cells transfected with expression vectors encoding the wild type or the mutated Leu601Val receptors bound hormone with similar affinity, whereas binding was barely detectable with the Asp224Val mutant. Confocal microscopy showed the latter to have an impaired targeting to the cell membrane. This was confirmed by its accumulation as a mannose-rich precursor. Adenylate cyclase stimulation by FSH of the Leu601Val mutant receptor showed a 12 \pm 3% residual activity, whereas in patient 1 a 24 \pm 4% residual activity was detected for the Arg573Cys mutant receptor. These results are in keeping with the fact that estradiol and inhibin B levels were higher in patient 1 and that stimulation with recombinant FSH did not increase follicular size, estradiol, or inhibin B levels in patient 2 in contrast to what was observed for patient 1. Thus, differences in the residual activity of mutated FSHR led to differences in the clinical, biological, and histological phenotypes of the patient.