ized in human endometrium biopsy samples (≥ 50 mg), either processed immediately or usually kept in liquid nitrogen. The total concentrations of estradiol and progesterone binding sites (available or occupied with endogenous hormone) were measured in cytosol by Dextran-coated charcoal adsorption and in nuclei by a glass filtre exchange assay. The contribution to total binding of non saturable binding components and of plasma proteins (transcortin or sex steroid binding protein) was taken into account. In 54 patients having completely normal menstrual cycles, total estradiol and progesterone receptors were highest in the late proliferative phase (about 8,000 and 12,000 sites per cell, respectively), and were very significantly lower in the late secretory phase. During the proliferative phase, estradiol receptor was increased only in the nuclear fraction, whereas progesterone receptor increased mainly in the cytoplasm. In the early luteal phase, estradiol and progesterone receptors decreased in the cytosol, whereas they remained high in the nuclei. Both receptors were at their lowest level in cytosol and nuclei in the late secretory phase. They were also measured in menstrual cycle disturbances (anovulation and luteal insufficiency), and in post-menopausal endometrial cancer. In the latter case, estradiol receptor was often high (about 9000 ± 2600 sites/cell), whereas progesterone receptor was generally present in low amounts (about 3000 ± 600 sites/cell). The highest values were found in well differentiated carcinoma.

Estrogen and progestin receptors in normal and malignant human uterine tissue, O. JÄNNE*, A. KAUPPILA†, K. KONTULA‡, P. SYRJÄLÄ‡, P. VIERIKKO* and R. VIHKO‡, Departments of Biochemistry*, Obstetrics and Gynecology† and Clinical Chemistry‡, University of Oulu, SF-90100 Oulu 10, Finland

Estrogen (ER) and progestin (PR) receptors were measured in cytosol fractions from normal human endometrium and myometrium as well as from endometrial carcinoma employing a Dextran-coated charcoal technique and [^{3}H]-estradiol and [^{3}H]-ORG 2058, a synthetic progestin, as the ligands. The cytosol PR levels in the endometrium were similar to those in the myometrium (when expressed per mg cytosol protein), and both were higher during the follicular than luteal phase of the menstrual cycle. One-week treatment of post-menopausal women with estrogens elicited a 15–20-fold increase in the uterine PR level, while one-month treatment with medroxyprogesterone acetate greatly decreased both ER and PR levels in the endometrial cytosol. Both ER and PR were present in 35 out of 45 endometrial adenocarcinoma samples studied. In 8 cases, only ER was present while 2 samples did not contain either receptor. Well differentiated adenocarcinomas contained more ER and PR than poorly differentiated ones. Hyperplastic endometria were characterized by high steroid receptor concentrations, while atrophic samples had minute receptor levels. ER and PR levels correlated significantly with each other in the case of both normal samples ($r = 0.5$, $P < 0.01$) and malignant samples ($r = 0.6$, $P < 0.01$), in marked contrast to the relationship between the same receptors in 165 human breast cancer samples ($r = -0.07$, N.S.). It was of interest to notice that PR/ER ratio was clearly higher in non-malignant (PR/ER = 4–8) than in malignant (PR/ER = 0.5–2) endometrial tissue. PR in the myometrial cytosol was composed of 4*S* and 7*S* receptor forms, the latter one being noted especially after estrogen treatment. Several tritium-labelled synthetic progestins studied preferred association with the 7*S* form of the myometrial PR. In addition to density gradient centrifugation, the PR from myometrial cytosol was resolved into two components during chromatography on DNA–Sepharose. A smaller PR component eluted from the DNA-column with 0.1 M KCl while the major peak required 0.25 M KCl for its elution. Whether these two forms of the PR represent receptor subunits similar to those described for chick oviductal PR, is not known at present.

NEWER TECHNIQUES—I. MASS SPECTROMETRY

Gas-chromatographic–mass spectrometric analysis of metabolic profiles of unconjugated steroids in biological materials, M. AXELSON and J. SJÖVALL, Department of Chemistry, Karolinska Institutet, Stockholm, Sweden

A sensitive method for analysis of metabolic profiles of unconjugated steroids in biological materials has been developed. The final analytical step is based on gas chromatography–mass spectrometry (GC–MS) using repetitive magnetic scanning over the entire or a large part of the mass range *m/e* 10–800. Computer evaluation of data, registered on magnetic tape, permit partial identification and quantitative determination of steroids.

The relatively low sensitivity using repetitive magnetic scanning technique is increased by using a GC–MS system, equipped with an open tubular glass capillary column, connected to the ion source *via* a single stage adjustable jet separator. By deactivation of glass and metal surfaces approximately linear responses for MO-TMS derivatives of steroids can be obtained from about 100 pg.

The limitations in sensitivity are also compensated for by analysis of larger sample sizes. For this purpose a simple and generally applicable method permitting extensive purification of unconjugated steroids in biological materials has been developed. After an extraction step, the sample is filtered through two columns of lipophilic-hydrophobic ion exchanging derivatives of Sephadex, which remove interfering material and separate estrogens from neutral steroids. MO-TMS derivatives of neutral steroids are purified on a column of Lipidex 5000.

Applications of this method to the analysis of steroids in plasma and tissues will be presented.

Use of mass spectrometry for clinical endocrinology, HERMAN ADLERCREUTZ, Department of Clinical Chemistry, University of Helsinki, Meilahti Hospital, 00290 Helsinki 29, Finland

Mass spectrometry (MS) and mass fragmentography (MF) are presently used in many areas of clinical endocrinological research and also to some degree for routine analyses of hormones. The most important applications in this and other laboratories will be reviewed. These are: (1) use in reference or control methods for the investigation of the specificity of radioimmunoassay and other procedures; (2) for routine analyses of catecholamines and their metabolites in plasma, cerebrospinal fluid and tissue samples; (3) to measure endogenous steroids in various biological materials such as plasma, urine and faeces and in tissues, e.g. breast cancer and prostate tissue; (4) in metabolic studies of hormones in man and animals; (5) for identification of new hormones in biological material and structure elucidation by amino acid sequencing; (6) in metabolic studies with stable isotopes, e.g. in *diabetes mellitus*. Thus, because of its high sensitivity and specificity MS can be used to solve a great variety of problems. In this laboratory MS and MF are presently used in studies of estrogen metabolism in the intestinal tract in man and the results obtained suggest that alterations in gut flora and perhaps diet have influence on the estrogen level in the organism, which secondarily may have clinical significance e.g. in breast cancer. In addition, we have recently tentatively identified the unknown steroid called "hypersterone"