Effect of Cyproterone Acetate on Cells of the Pars distalis of the Adenohypophysis in the Beagle Bitch*

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Summary. The effects of oral administration of 100 mg per kg per day cyproterone acetate (CPA) for four weeks on cells of the pars distalis, as revealed by the immunoperoxidase technique and chemical staining, were studied in the ovariectomized beagle bitch. For immunochemical staining antisera to the following hormones were used: canine GH, canine PRL, porcine ACTH, bovine TSHβ, bovine LHβ and human FSHβ1. The most striking effects of the treatment were an overall increase in the relative proportion of GH cells and a marked morphological indication of high secretory activity in these cells. In contrast, PRL cells were not affected significantly. In all ovariectomized control bitches a marked atrophy of the cells stained for FSHβ (FSH cells) and hypertrophy of the cells shown to contain LHβ (LH cells) were observed. FSH cells became enlarged, while LH cells appeared reduced in size by administration of CPA. In some treated bitches ACTH/MSH cells showed atrophy and regressive changes, whereas TSH cells seemed to become enlarged and were more densely arranged. These structural responses indicate that, in addition to its partial anti gonadotropic properties, CPA as a synthetic progesterone derivative may stimulate GH secretion and possibly suppress CRH-ACTH activity in the ovariectomized beagle bitch.

Key words: Cyproterone acetate – Pars distalis – Adenohypophysis – Immunocytochemistry – Dog (Beagle).

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1 Abbreviations of hormones cited in this paper: ACTH Adrenocorticotropic; FSH Follicle Stimulating Hormone; GH Growth Hormone; LH Luteinizing Hormone; MSH Melanocyte Stimulating Hormone; PRL Prolactin; TSH Thyrotropin; CRH Corticotropin Releasing Hormone
Introduction

The recent development of the immunoperoxidase technique for localization of tissue antigens (Nakane and Pierce, 1967; Mason et al., 1969; Sternberger et al., 1970) has resulted in great progress in the identification and characterization of the various cell types of the adenohypophysis (Moriarty, 1973; Sternberger, 1974; Baker, 1974; Nakane, 1975). Relying primarily on such a procedure and in conjunction with chemical staining and stereometric cell analysis, the authors have shown that long-term treatment with high doses of estrogen (17β-estradiol) causes complete regression of gonadotrophs and marked stimulation of PRL cells in the adenohypophysis of the dog (El Etreby et al., 1977). However, the structural response of the hypophysis of this species to administration of progesterone or synthetic progestogens has not as yet been clearly defined (Kraft, 1971; Tucker, 1971; Neumann and Elger, 1971; Capel-Edwards et al., 1973; El Etreby and Günzel, 1973, 1974).

The present investigation was carried out to study the cytological changes in the pars distalis of the adenohypophysis of the ovarioctomized beagle bitch during short-term treatment with high doses of the hydroxyprogesterone type progestogen, cyproterone acetate, using immunocytochemical and histologic techniques.

Material and Methods

The pituitaries of six beagle bitches, aged about 1–2 years and bred by Schering AG, were studied. The animals were ovariohysterectomised 7–8 months prior to the start of the experiment. They were kept in individual kennels with a 12:12 h light: dark cycle and given a standard diet (Altromin H) and tap water ad libitum. Three bitches were treated orally with 100 mg/kg/day CPA (micronized form) in gelatine capsules for 4 weeks. The other three bitches received orally empty gelatine capsules daily for 4 weeks and served as controls. Pituitary glands from seven adult beagle bitches (anestrus phase of the sexual cycle) with intact gonads were also included in this study to investigate the possible effects of ovariecction on cells of the pars distalis (cf. El Etreby and Fath El Bab, 1976).

Upon termination of the experiment all animals were sacrificed by exsanguination under hexobarbital anesthesia. The pituitaries were immediately removed, fixed for 24 h in formol-sublimate (1 part 37% formaldehyde +9 parts of a saturated aqueous mercuric chloride solution), and then washed in running water for at least 24 h.

After embedding in paraffin the pituitaries were sectioned in the paramedian plane. Successive sections, each 3–5 μm thick, were subjected alternately to either the immunoglobulin-enzyme bridge technique (Mason et al., 1969) or the peroxidase-antiperoxidase complex unlabeled antibody method (Sternberger et al., 1970) to localize the different hormones of the pars distalis. Adjacent sections were also stained using the carmoisine L – orange G – aniline blue – acid alizarine blue (El Etreby and Tiishaus, 1973) or performic acid-alcian blue (pH 0.2) – PAS – orange G procedures (El Etreby et al., 1973).

For immunochemical staining rabbit antisera to the following hormones were used: canine GH (anti-cGH), canine PRL (anti-cPRL), porcine ACTH (anti-pACTH), bovine TSHβ subunit (anti-bTSHβ) and bovine LHβ subunit (anti-bLHβ). Specific rat antiserum to human FSHβ subunit (anti-hFSHβ) was also included in this study. The antiserum were kindly provided as follows: antiserum to cPRL by Dr. K.-J. Gräf (Department of Endocrine Pharmacology, Schering AG, Berlin); to bTSHβ and bLHβ by Dr. J.G. Pierce (Department of Biological Chemistry, Los Angeles, California); to hFSHβ by Dr. D.C. Herbert (Department of Anatomy, San Antonio, Texas) and to pACTH by Dr. P.K. Nakane (Department of Pathology, Denver, Colorado). Antiserum to cGH (D 10012 AC, a gift from Dr. A.E. Wilhelmi; Department of Biochemistry, Atlanta, Georgia) was prepared in this laboratory.