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„Pregnancy Zone" Protein in Sera from Patients with Prostatic Cancer Treated with Oestrogens

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Summary. The "pregnancy zone" protein is a human a_2-serum globulin previously found in pregnant women and women taking oral contraceptive drugs. The occurrence of this protein in sera from 22 out of 34 patients with prostatic cancer treated with oestrogen was demonstrated by an immunological technique. Immunological identity was found between this steroid-inducible protein and that in sera from pregnant women and women taking contraceptive drugs. This protein was not detected in 27 patients with prostatic cancer not treated with oestrogens. Oral diethylstilboestrol appeared to induce the "pregnancy zone" protein more often than injections of polyoestradiol phosphate.

Key words: Pregnancy zone protein, pregnancy proteins, oestrogen carrier, prostatic cancer, steroid-inducible proteins.

In 1959 Smithies (12), using starch gel electrophoresis, was the first to describe a new protein band in the a_2-globulins of human serum. The extra protein was found in some pregnant women and was called the "pregnancy zone" protein. Later studies (1, 2, 3, 5, 8, 9) confirmed the occurrence of this protein in pregnant women, and showed that an immunologically identical protein was induced in women taking oral contraceptive drugs (4, 6, 9). The present results suggest that the "pregnancy-zone" protein is truly a steroid-inducible protein.

The a_2-globulin has been purified (11) and a monospecific precipitating rabbit antiserum prepared against it (7).

In a previous study Cooper (8), also using starch gel electrophoresis, detected the "pregnancy zone" (PZ) protein in a man with prostatic cancer, who had been treated with oestrogen. The present experiments were done to confirm this observation using a specific and more sensitive immunological technique for detection of the PZ-protein and also to discover whether its induction was due to the presence of prostatic cancer or to the hormonal treatment.

Material and Methods

Serum samples were collected at the Department of Urology, University Hospital, Umea, from the following two groups of patients.

1. A series of 18 patients with prostatic cancer were examined during treatment with oestrogens. One patient received only polyoestradiol phosphate, and the other 17 a combination of polyoestradiol phosphate and diethylstilboestrol. Bilateral orchidectomy had been performed in one case. Five of the 18 patients had demonstrable metastases.

2. A series of 27 cases of prostatic cancer was examined before treatment with oestrogens; and, from 16 of them a second serum sample was obtained after oestrogen treatment for a period averaging four months. Amongst the 16 patients, five received polyoestradiol phosphate only, and the others polyoestradiol phosphate and diethylstilboestrol. Three of the 16 patients had demonstrable metastases.

In the Department of Urology steroid treatment was usually given according to the following scheme:

A. Injection of 160 mg polyoestradiol

phosphate (Estradurin R) once a month for three months, followed by 80 mg per month.

B. Oral administration of diethylstilboestrol (Stilbol R) 3 x 10 mg per day for ten days, and thereafter 3 x 2 mg per day.

Patients with cardiosclerosis and cardiac insufficiency were not given diethylstilboestrol. The diagnosis of carcinoma of the prostate was verified in all cases by cytological examination of a biopsy of the prostate.

A monospecific rabbit antiserum was used for detection of the "pregnancy zone" protein (7). Double diffusion tests according to Ouchterlony (10) were done on glass slides coated with 2 mm thick layers of 1% agarose (Behring-Werke) in 0.1 M sodium phosphate buffer pH 7.4. Six peripheral and one central well were cut in the agar; the diameter of the wells was 3 mm and the distance between wells was 5 mm. The rabbit immune serum (10 μl) was placed in the central well and the human sera (10 μ) to be tested in the peripheral wells. The glass slides were kept in a moist chamber at 37°C. The precipitates were examined after 1, 2 and 3 days by oblique illumination from below against a dark background.

Results

Of the 18 patients in the first series 14 were found to have the "pregnancy zone" protein. In the second series, before treatment with oestrogen none of the 27 patients with prostatic cancer showed the "pregnancy zone" protein in their sera, but after 16 of them had been treated with oestrogen for a period averaging four months, eight were found to have the "pregnancy zone" protein.

A reaction of complete immunological identity was found between sera from oestrogen-treated patients, purified "pregnancy zone" protein, sera from pregnant women and from women taking oral contraceptive drugs (Fig. 1). The duration of treatment had no significant effect on the development of the "pregnancy zone" protein, but the type of oestrogen administered did seem to be of importance; thus, of 28 patients treated with polyoestradiol phospate and diethylstilboestrol, 22 (79%) developed the "pregnancy zone" protein, whilst of six patients treated with polyoestradiol phosphate alone, none developed it.

Discussion

The present study shows that oestrogen treatment of males may result in the appearance of a serum protein that has been called the "pregnancy zone" protein in previous investigations. In the present study this protein was found in 22 out of 34 patients (65%) with prostatic cancer treated with oestrogen, whereas it was absent from 27 untreated cases. In eight of the latter cases the "pregnancy zone" protein appeared during hormone therapy. These results show that the occurrence of this protein is due to oestrogen treatment and not to the prostatic cancer.

Some patients did not develop the "pregnancy zone" protein, at least not in amounts detectable by the immunodiffusion method. Similarly, a proportion of pregnant women and those treated with oral contraceptives also seem unable to react to the altered steroid levels by producing the "pregnancy zone" protein. In some of the present patients the absence of the "pregnancy zone" protein appeared to depend on the type of oestrogen treatment. None of the six patients treated with monthly injections of polyoestradiol phospate produced this protein, whereas 22

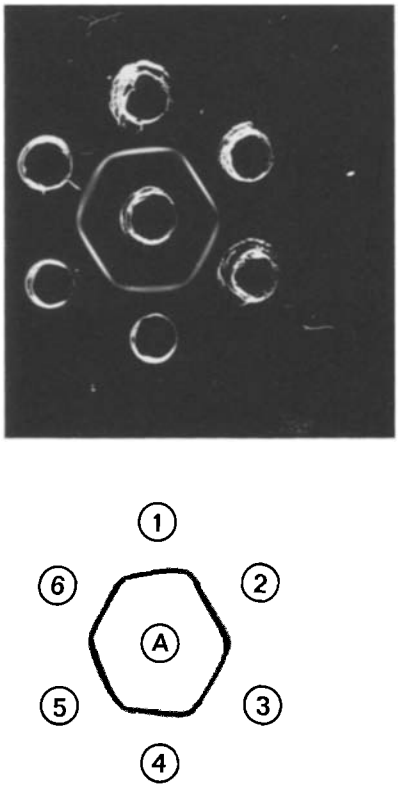

Fig. 1. Precipitation reactions in agar gel between rabbit antiserum (A) in central well and purified "pregnancy zone" protein, and sera in the peripheral wells. Upper part - photograph; lower part - schematic drawing. Peripheral wells: 1 = purified "pregnancy zone" protein; 2, 4, and 6 = sera from men treated with oestrogen; 3 = serum from women taking oral contraceptives and 5 = serum from pregnant woman at term

out of 28 patients treated with the combination of injections of polyoestradiol phosphate and oral diethylstilboestrol did so. In other cases the absence of the "pregnancy zone" protein could not be ascribed to the type, dose, or duration of treatment and the most likely explanation, as in pregnancy and after administration of oral contraceptives, seems to be that there are individual differences with respect to the inducibility of the "pregnancy zone" protein. It has been postulated that the "pregnancy zone" protein is a carrier of oestrogen (1, 2, 6) and it may be asked, therefore, whether variations in the response to oestrogen treatment are in any way related to the cancer suppressing effect of the oestrogen treatment. This question will be investigated in a future study.

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