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Effects of Oral Ovulation Inhibitors on Serum Protein-Bound Iodine and Thyroxine Binding Proteins.* (29495)

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Most oral ovulation inhibitors in clinical use are compounded of a progestational agent and an estrogen. Estrogens, even in low dosage, are known to elevate circulating levels of thyroxine binding globulin (TBG)(1) and the PBI(2,3). However, the clinical effect of ovulation inhibitors depends primarily upon the progestational agent and may range from strongly estrogenic, as with norethynodrel preparations(4) to mildly androgenic, as has been claimed for norethindrone(5). Steroids structurally related to norethindrone's progestagen have been shown to depress both PBI and TBG(6). The effects of oral ovulation inhibitors upon PBI and thyroxine binding proteins thus are not predictable *a priori*. In view of the large number of patients taking these drugs a study of their effect upon commonly used criteria of thyroid function seemed desirable.

Hollander(7) reported recently that norethynodrel caused an elevation of the PBI and a depression of the erythrocyte triiodothyronine uptake test. Medroxyprogesterone acetate did not affect either test in Hollander's study. We have extended his study to cover the effect of 4 commonly used ovulation inhibitors and have measured their effects on PBI and the 2 principal thyroxine binding proteins.

Materials and methods. The drugs studied were: 1. Norethynodrel, 17-α-ethinyl-5(10)-estrenolone, marketed as "Enovid" by G. B. Searle & Co. Tablets contain either 5 mg norethynodrel plus 0.075 mg ethinyl estradiol-3-methyl ether (mestranol) or 2.5 mg norethynodrel plus 0.10 mg mestranol.

2. Medroxyprogesterone acetate, 6-α-methyl-17-α-hydroxyprogesterone acetate, supplied as "Provest" by Upjohn Co., containing 5 mg medroxyprogesterone acetate plus 0.10 mg ethinyl estradiol.

3. Norethindrone, 17-α-ethinyl-19-nortestosterone, marketed as Orthonovum by Ortho Pharmaceutical Co., containing 10 mg norethindrone plus 0.06 mg mestranol.

4. Chlormadinone, 6-chloro-Δ6-dehydro-17-α-acetoxyprogesterone, supplied by Eli Lilly & Co., containing 2 mg chlormadinone plus 0.06 mg mestranol.

Sera from healthy women of child-bearing age who had been on the appropriate drug regimen for at least 3 months were obtained through the kindness of Dr. E. T. Tyler of Los Angeles and Dr. C. Gassner of Long Beach. Sera from 4 of these patients were available before initiation of therapy. In a further case we obtained repeated serum specimens during a 2-year period of norethynodrel therapy followed by 2 months off the drug and during a subsequent pregnancy.

PBI's were measured by a modification of Bodansky's wet ashing technique(8) which has given a normal range of 4.5 to 7.5 μg per 100 ml in our hands in several thousand determinations.

Serum thyroxine binding capacities, both pre-albumin (TBPA) and TBG were determined by Beierwaltes' modification(9) of Robbins' reverse flow technique. The thyroxine concentration in sera enriched with thyroxine-I^{131} (Abbott Laboratories) and stable thyroxine (California Corp. for Biochemical Research) was determined by Posner's butanol-extractable iodine method(10) after suitable dilution of the butanol extracts.

Results. As summarized in Table I, the subjects who had been treated with the 4

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TABLE I. Effect of Oral Ovulation Suppressor Treatment on Serum PBI and Thyroxine Binding Capacity in Normal Women.

	No. of subjects	μg/100 ml PBI	μg/100 ml TBPA	μg/100 ml TBG
Controls	15	5.9 ± .2*	79 ± 9	15.5 ± .7
Norethynodrel, 5 mg	11	7.6 ± .5†	81 ± 5	25.6 ± .9‡
" , 2.5 mg	4	7.6 ± .6	87 ± 27	27.6 ± 3.3†
Medroxyprogesterone acetate	14	7.4 ± .3‡	81 ± 12	26.1 ± 1.4‡
Norethindrone	10	6.6 ± .4	120 ± 10†	24.0 ± 1.4‡
Chlormadinone	5	7.7 ± .2‡	81 ± 10	28.2 ± 1.0‡

* Mean and S.E. † Significant at 0.01 confidence level. ‡ Significant at 0.001 confidence level.

TABLE II. Changes in Thyroxine Binding Capacity and PBI During Norethynodrel Therapy.

Subject	Before therapy, μg/100 ml PBI	TBPA	TBG	Months of therapy	After therapy, μg/100 ml PBI	TBPA	TBG
GW	5.6	85	18.4	3	7.2	60	23.2
DD	5.4	98	15.6	4	8.8	93	32.2
DM	5.4	63	16.7	5	8.9	66	26.4
FK	5.1	71	15.6	4	10.8	97	28.3
				PBI	TBPA	TBG	
	Mean difference ± S.E.			3.6 ± .8*	1 ± 11	11.0 ± .8†	

* Significant at 0.01 confidence level. † Significant at 0.001 confidence level.

oral ovulation suppressing drugs showed significant elevation of both the PBI and the TBG capacity over the control group. Both dosages of norethynodrel caused very similar effects. TBPA was unaffected except in the group on norethindrone which showed a significant elevation of the TBPA also.

Table II presents the data on the 4 patients who returned for follow-up studies after 3 to 5 months of norethynodrel therapy (5 mg dose). All showed striking elevations of the PBI and TBG, which were significant at the .001 confidence level for TBG and .01 for the PBI, while changes in the TBPA were inconclusive.

Finally, Table III shows PBI and TBG changes in a subject on constant 1 grain per day thyroid replacement therapy (Proloid, Warner Chilcott) over a 2-year period of norethynodrel (5 mg) medication, the reversal of these changes toward normal after cessation of norethynodrel therapy and finally, the normal rise of PBI and TBG in early pregnancy.

Discussion. All 4 of the drugs investigated caused a marked rise in PBI, presumably secondary to the elevation of the serum TBG. This occurred with norethisterone which has clinically a predominant progestational effect (4) as well as with norethynodrel whose effect on the endometrium indicates a strong estrogenic action and with norethindrone, which may be androgenic(5). None of the progestational compounds investigated reverse the effect expected of the estrogen with which they have been compounded in the commercial preparations. The data of Table II show that in the few patients for whom baseline studies were available the effects of norethynodrel were entirely comparable to those in Table I, indicating that our subjects do represent a "normal" sampling and thus can be compared by statistical methods. There appeared to be no progressive effect

TABLE III. Changes of PBI and TBG in a Subject on Constant Thyroid Replacement Therapy During and After Norethynodrel Therapy.

		μg/100 ml PBI	μg/100 ml TBG
Before norethynodrel		4.0	—
On norethynodrel	1 yr	6.3	24.5
On "	2 "	5.8	26.2
Off "	1 mo	5.0	21.7
Off "	2 "	3.9	16.7
Off "	6 " , 4 mo pregnant	6.0	24.4

of the drugs with length of treatment in the cases we studied. In the subject of Table III there was no significant change during 2 years of norethynodrel therapy. Both PBI and TBG appear to rise to a new level which is then maintained for many months. After cessation of therapy, TBG values returned to normal in about 2 months. The PBI levels during treatment, however, may be near the upper end of the accepted normal range and the clinical interpretation of PBI values thus requires a knowledge of the patient's therapeutic history which becomes increasingly important as the use of ovulation suppressing drugs becomes more prevalent.

Our data agree well with Hollander's study (7), except for the results with medroxyprogesterone acetate. The preparation we studied contained ethinyl estradiol not present in the preparation used by Hollander and the differences thus must be ascribed to this estrogen.

One rather surprising result of our study was the finding of a marked elevation of the TBPA level by norethindrone therapy. Since the preparation used contained the same estrogen as 2 other drugs which did not affect the TBPA capacity the effect must be ascribed to the ethinyl-nortestosterone component. Nothing is known about the factors determining serum TBPA capacity other than that it tends to be low in acute illness (11) and in children(12).

The TBPA capacities here reported are somewhat below the 90-160 μg/100 ml normal range given by Ingbar(13). This is largely due to the fact that at high thyroxine "loads" many sera apparently become saturated. We routinely add 400 μg of carrier thyroxine per 100 ml of serum to saturate the TBPA. In many sera thus saturated a portion of the added thyroxine deposited on the glass walls of the container. The actual thyroxine level calculated from the butanol extractable iodide determination and the TBPA capacity calculated from it therefore was considerably lower than the level calculated from the thyroxine added.

It is interesting that the PBI rose whenever the TBG became significantly elevated, but was not further elevated by a 50% increase in the TBPA capacity. This confirms our previous findings(12) and would tend to indicate a lesser metabolic role for TBPA than Oppenheimer's data(11) would assign to it.

Summary. Administration of norethynodrel, medroxyprogesterone acetate, norethindrone and chlormadinone compounds to healthy female subjects elevated both the PBI and the TBG significantly. Norethindrone also elevated the TBPA capacity.

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