TOTAL AND UNCONJUGATED PLASMA ESTRIOL

A comparison

P. V. GRELLA, L. DI LENARDO,
F. SORMANI-ZODO, G. BARAZZUOL,
B. MOZZANEGA, D. MARCHESONI

Institute of Obstetrics and Gynecology,
University of Padua (Italy)

SUMMARY

Total and unconjugated plasma estriol levels from the 24th to the 40th gestational week in 161 normal and 44 pathological pregnancies were studied. A lack of correlation between the two fractions was observed at the different gestational ages, probably due to the different rates of the several metabolic steps which condition the levels of the hormone fractions also in quite physiological conditions. The onset of pregnancy complications further modifies the ratio between the two fractions, as they differently affect the single metabolic processes which are essential for estriol to be produced, conjugated, recovered and excreted.

E.P.H. gestosis and poor intrauterine fetal growth can be better diagnosed by the assay of total than unconjugated estriol: the total fraction proved to have the higher sensitivity, predictive value and relative risk and should therefore be preferred for pregnancy monitoring, as it better corresponds to the clinical situation.

Plasma estriol radioimmunoassay (R.I.A.) is more and more often used in the evaluation of fetoplacental function, while the contrary happens for the urinary assay. The R.I.A. enables us to determine both the total and unconjugated estriol plasma levels ([1, 2, 3]).

The purpose of our work is to verify whether the ratio between unconjugated and total estriol is constant in normal pregnancies, and whether the two fractions follow similar trends in particular complications of pregnancy. In case the ratio between unconjugated and total estriol is variable as pregnancy progresses, the assay of the two fractions can't be obviously given the same signification.

MATERIAL AND METHODS

To answer the first question we performed 2138 determinations in 161 pregnant women which could be considered normal according to previously established criteria ([3]). Their gestational ages ranged between the 25th and 40th week and had been ascertained by seriate ultrasonic measurements of fetal diameters (crown-rump lenght, biparietal diameter) during the first trimester. Blood samples were drawn every second day at 8.00 a.m.; the R.I.A. was contemporaneously performed of total (Sorin-Biomedica kit) and unconjugated (Immunoanalysis kit) plasma estriol. The amount of conjugated estriol was calculated by deduction.

To answer the second question 44 patients presenting pathological pregnancy were examined: of them, 18 were affected by E.P.H. gestosis, diagnosed according to the indications of Organization Gestosis; 18 presented a poor intrauterine fetal growth, demonstrated through ultrasonic biometric parameters and confirmed by biometry at birth: we considered abnormal the cases presenting values equal to or below the 10th centile calculated on the whole series of our Institute ([3]); 8 patients, at last, were affected by hepatogestosis (cholostatic jaundice of pregnancy).

In all these patients estriol was assayed through the same methods as in the controls.

The values obtained in normal pregnancies were subdivided per week and centiles, as the number of data per week was satisfactory; the centiles were then approximated through a polynomial approximation of second degree, to better fit our curves without any arbitrary interpolation.

The comparative evaluation of the two assays, as to the pregnancy diseases we considered, was

Reprinted from "J. F. Medicine", 2: 8, 1982.

Table 1. — *Absolute variations of total and unconjugated estriol in 161 normal pregnancies (2138 assays), from the 25th to the 40th gestational week.*

Week of gestation	10th centile		50th centile		90th centile	
	Total	Unconjugated	Total	Unconjugated	Total	Unconjugated
25	10.94	1.78	29.83	3.52	58.72	5.64
26	13.81	1.64	35.68	3.47	77.49	6.01
27	16.67	1.57	41.57	3.51	94.79	6.57
28	19.47	1.54	47.51	3.65	110.61	7.01
29	22.27	1.57	53.49	3.87	124.97	7.64
30	25.04	1.65	59.52	4.20	137.84	8.35
31	27.79	1.80	65.59	4.60	149.25	9.15
32	30.52	1.98	71.71	5.10	159.18	10.04
33	33.22	2.22	77.87	5.70	167.64	11.02
34	35.90	2.52	84.07	6.37	174.82	12.08
35	38.56	2.87	90.32	7.15	180.14	13.22
36	41.19	3.27	96.62	8.02	184.17	14.55
37	43.79	3.73	102.96	8.97	186.74	15.77
38	46.38	4.24	109.34	10.02	187.83	17.18
39	48.94	4.81	115.77	11.17	187.45	18.67
40	51.47	5.42	122.24	12.40	185.60	20.40

Table 2. — *Relative variations of the medians of total and unconjugated estriol in normal pregnancy.*

Week of gestation	Total	Conjugated	Unconjugated	Conjugated/ Unconjugated ratio	Conjugated %	Unconjugated %
25	29.83	26.31	3.52	7.50	88.20	11.20
26	35.68	32.21	3.47	9.30	90.27	9.72
27	41.57	38.06	3.51	10.84	91.55	8.44
28	47.51	43.86	3.65	12.00	92.31	7.68
29	53.49	49.62	3.87	12.80	92.76	7.23
30	59.52	55.32	4.20	13.30	92.94	7.05
31	65.59	60.99	4.60	13.25	92.98	7.01
32	71.71	66.61	5.10	13.00	92.88	7.11
33	77.87	72.17	5.70	12.60	92.68	7.31
34	84.07	77.70	6.37	12.20	92.42	7.57
35	90.32	83.17	7.15	11.70	92.08	7.91
36	96.62	88.60	8.02	11.00	91.70	8.30
37	102.96	93.93	8.97	10.50	91.20	8.71
38	109.34	99.32	10.02	9.90	90.08	9.16
39	115.77	104.60	11.17	9.36	90.30	9.64
40	122.24	109.84	12.40	8.80	89.80	10.14

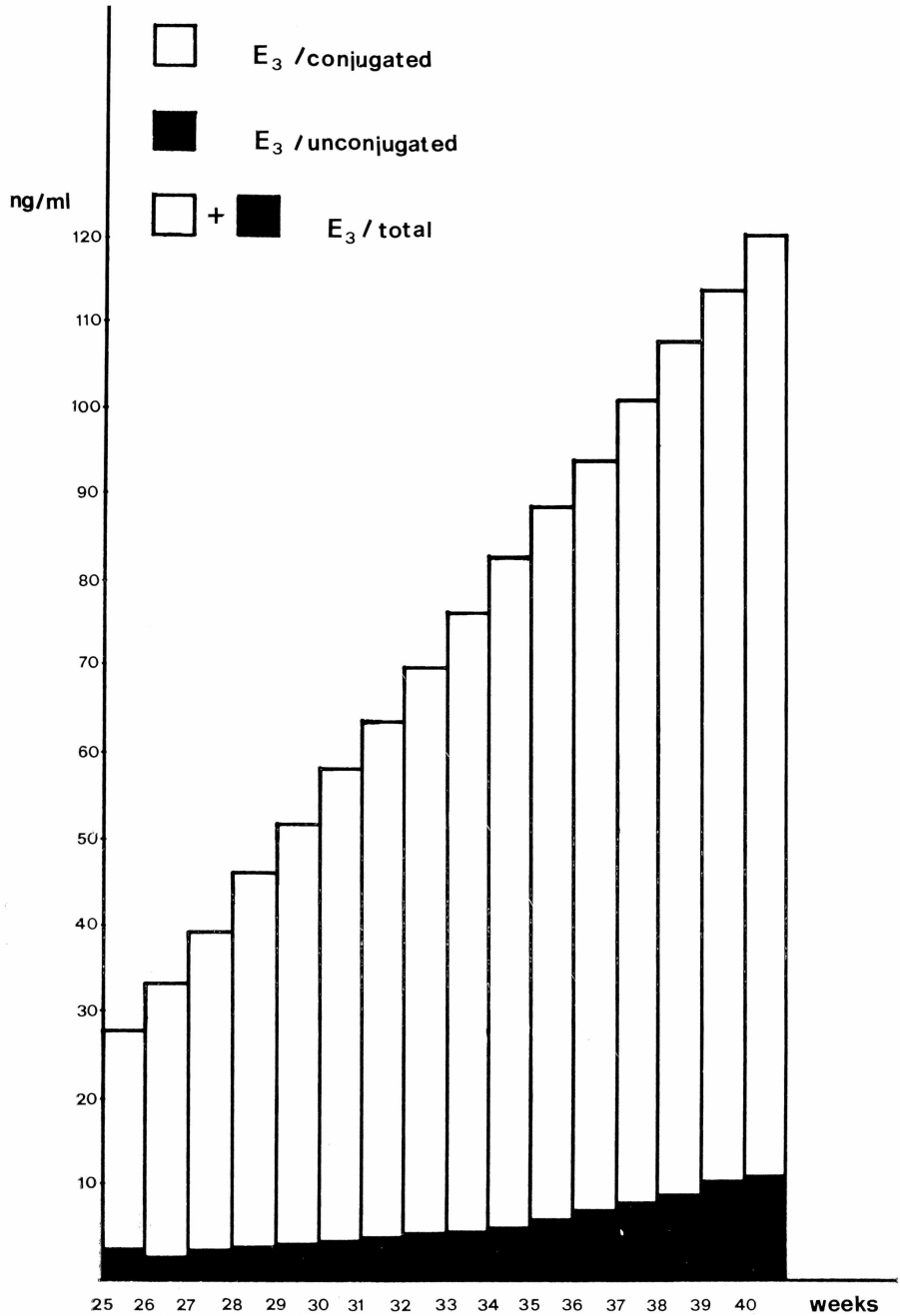

Fig. 1. — Medians of conjugated and unconjugated estriol from the 25th to the 40th gestational week in 161 normal pregnancies.

carried out by comparing the medians and data-distribution in relation to the 10th and 50th centiles through contingency-tables verified, as to their significativity, by the χ^2 method.

The comparison between the two methods, as to their clinical reliability, was carried out in the cases of E.P.H. gestosis and poor fetal growth through the evaluation of their sensitivity, predictive value and relative risk.

RESULTS

Plasma estriol fractions in normal pregnancy

In the table 1 are reported the 10th, 50th and 90th centiles of total and unconjugated plasma estriol levels between the 25th and 40th gestational week.

The comparison between conjugated and unconjugated plasma fractions was performed basing on the medians, as these are non-parametric expression of the normal pregnant population; the results are reported in the table 2.

The variation of conjugated and unconjugated plasma estriol from the 25th to the 40th gestational week is represented in the fig. 1 and 2.

Plasma estriol fractions in pathological pregnancies

Some complications of pregnancy might affect in different degrees the single fractions of plasma estriol by interfering not only with its input into maternal blood by the feto-placental unit, but also with its recovery from the entero-hepatic circulation, with maternal liver conjugating capacity and with excretion of the hormone into maternal bile and urine ([17]).

If this interference is unimportant, in patients presenting a pathological pregnancy no difference would be expected between the distribution of values of total estriol and that of unconjugated estriol in plasma, in relation to normal values. The results are showed in table 3.

The number of data from the cases of E.P.H. gestosis and poor intrauterine fetal growth let us verify the ability of the two estriol fractions to lead to correct diagnoses; to this aim, we considered pathognomonic the values below the 10th centile (table 4).

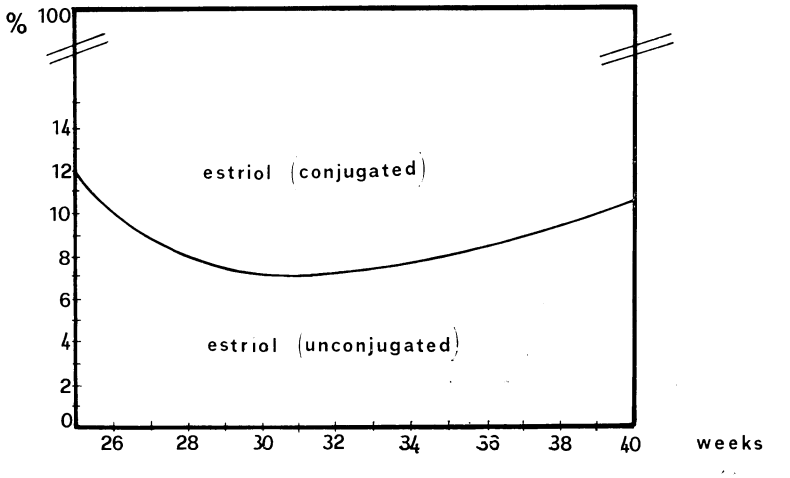

Fig. 2. — Variability of percentages of conjugated and unconjugated estriol in relation to gestational age.

Table 3. — *Significativity of the distribution-differences of total estriol and unconjugated estriol values in relation to the 50th and 10th centiles of normal values. Probability of error of the differences.*

	In relation to			
	the 50th centile		the 10th centile	
	Total	Unconjugated	Total	Unconjugated
E.P.H. gestosis	<0.0005	0.1-0.2	<0.0005	0.7-0.8
Poor fetal growth	<0.0005	<0.0005	<0.0005	<0.0005
Hepatogestosis	0.5-0.6	0.001-0.005	0.05-0.1	0.05-0.1

Table 4. — *Evaluation of the ability of the two estriol fractions to provide correct diagnoses.*

	Sensitivity %		Predictive value %		Relative risk	
	Total	Unconjugated	Total	Unconjugated	Total	Unconjugated
E.P.H. gestosis	28.4	9.3	19.6	9.3	2.46	0.77
Poor fetal growth	59.0	42.3	43.3	33.3	6.46	3.93

DISCUSSION

Estriol level in pregnant women's peripheral blood is conditioned by several factors ([4, 5]): production-rate by the feto-placental unit; conjugation-rate by the liver, which conditions the excretion-rate into bile and urine; recovery-rate from fetal and maternal compartments of the unconjugated-unbound fraction into the blood stream; recovery-rate from entero-hepatic circulation.

The relative importance of each single factor could hardly be evaluated; anyway, the conjugated fraction seems to have a not-secondary role in determining the hormone total plasma level.

Our study shows a particular trend of the ratio between conjugated and unconjugated estriol seriately assayed in normal pregnant women from the 25th week until term, with a minimal percentage of the latter around the 31st week (table 2 and fig. 2). This phenomenon might result from the combined action of the first three previously mentioned factors: from the 25th to the 31st week an increase in production (fig. 1) and in conjugation (fig. 2) takes place, unbalanced by a clearance-increase of the same rate. In the last weeks of pregnancy this phenomenon tends to reverse, as if the increasing input were not matched by an equal conjugating capacity.

Consequently, in normal pregnancy unconjugated estriol plasma levels at different gestational ages are not correlated with the total estriol levels, in contrast with the data from other works ([6]).

The onset of pregnancy complications differently affects the total estriol and the unconjugated estriol in plasma: the table 3 points out that *E.P.H. gestosis* significantly modifies total estriol levels ([7, 8]), while unconjugated estriol is often unchanged ([9]); in other words, the hormone excretion would be mainly altered, with a possible reduction of conjugation too.

Poor intrauterine fetal growth significantly modifies both estriol fractions ([3, 7, 10, 11, 12, 13 14, 15]).

Hepatogestosis, at last, significantly alters the unconjugated fraction ([16]), which is commonly found above the 50th centile, while the total level doesn't seem to vary.

Shifting from the theory to the clinical practice, it's important to evaluate which one of plasma estriol fractions can pro-

vide the higher number of correct diagnoses (table 4). Our results point out the higher sensitivity of total estriol assay in the diagnosis of E.P.H. gestosis and poor fetal growth, and its higher predictive value, mainly in E.P.H. gestosis.

The relative risk indicates that the assay of unconjugated estriol is not discriminative in E.P.H. gestosis, while it is useful enough in poor fetal growth, even if, chiefly in this pregnancy complication, total estriol assay still proves much better.

In conclusion we can say that, even if conjugated estriol is the primitive product of the feto-placental unit, the assay of total estriol shows characteristics to be preferred from the view-point of sensitivity, predictive value, relative risk and, then, of correspondence to the clinical situation.

BIBLIOGRAPHY

1) Dubin N. H., Crystle C. D., Grannis G. F., Townsley S. D.: *Am. J. Obst. Gyn., 115*, 835, 1973.
2) Hay D. M., Lorscheider F. L.: *Brit. J. Obst. Gyn., 83*, 118, 1976.
3) Onnis A., Grella P.: *Atti Soc. Ital. Ostet. Ginec., 59*, 17, 1978.
4) Gurpide E.: "Truer Methods in Hormone Research", Springer ed., New York, 1974.
5) Gurpide E., Gandy H. M.: "Dynamics of hormone production and metabolism". In "Endocrinology of Pregnancy", 2nd ed., edited by F. Fuchs and A. Klopper, Hagertown, Harper & Rom, 1, 1974.
6) Katagiri A., Jandial V., Wilson G.: *Am. J. Obst. Gyn., 124*, 272, 1976.
7) Klopper A., Jandial V., Wilson G.: *J. Steroid Biochem., 6*, 651, 1975.
8) Masson G. M.: *J. Obst. Gyn. Brit. Cwlth., 80*, 206, 1973.
9) Shaaban M. M., Hammad W. A., Ghaneimah S. A.: "Simultaneous serial measurements of serum unconjugated estriol and pregnancy-specific beta-1 glycoprotein in E.P.H. gestosis", in "Current status of EPH gestosis", edited by A. Kurjak, E. T. Rippmann and V. Sulovic, Amsterdam, Excerpta Medica, 293, 1981.
10) Masson G. M.: *Brit. J. Obst. Gyn., 80*, 536, 1973.
11) Gorwill R. H., Sarda I. R.: *Am. J. Obst. Gyn., 127*, 17, 1977.
12) Bashore R. A., Westlake J. R.: *Am. J. Obst. Gyn., 128*, 371, 1977.
13) Tulchinsky D., Hobel C. J., Koreman J. G.: *Am. J. Obst. Gyn., 111*, 311, 1971.
14) Edwards R. P., Diver M. J., Davis J. C., Hipkin L. J.: *Brit. J. Obst. Gyn., 83*, 229, 1976.
15) Kunzig H. J., Geiger W., Kuhn H., Schutt A.: "Steroids in the diagnosis of poor intrauterine fetal growth". In "Poor Intrauterine Fetal Growth", edited by B. Salvadori and A. Bacchi Modena, Roma, Minerva Medica, 323, 1977.
16) Levitz M., Slyper A. J., Selinger M.: *J. Clin. Endocrinol. Metab., 38*, 698, 1974.