Acta Genet Med Gemellol 34:59-66 (1985)

Received 6 November 1984
Final 27 February 1985

Human Placental Lactogen and dU-Estrogen Levels in Normal Twin Pregnancies

B. Kappel, K. Hansen, J. Moller, J. Faaborg-Andersen
Department of Obstetrics and Gynecology, Kommunehospital, Aarhus, Denmark

Abstract. Reference intervals for human placental lactogen (hPL) and dU-estrogens (dU-E) in uncomplicated twin pregnancies are found to be higher than singleton pregnancies and considerably wider. Significantly more monozygotic than dizygotic pregnancies, and significantly more monoplacental than diplacental pregnancies showed hPL values below the median on the normal range curve. The same was not found for dU-E. Significantly more growth-retarded fetuses were found in monoplacental than diplacental pregnancies. The benefit of measuring the two parameters in order to identify the intrauterine growth retarded fetuses were evaluated in terms of sensitivity and specificity. Both parameters were found less suitable for the purpose.

Key words: Twin pregnancy, Hormone reference intervals, Human placental lactogen, dU-estrogens, Intrauterine growth retardation

INTRODUCTION

Twin pregnancy is a high-risk pregnancy with increased frequency of preterm delivery, intrauterine growth retardation and increased perinatal mortality [11]. Careful supervision throughout pregnancy is therefore essential. For this purpose, measurement of hPL and dU-E has been used. Reference intervals for twin pregnancies have been published [1,3,5,7,9,15]: the problem is that they are very broad. This has made the evaluation of isolated measurements difficult. The twin pregnancies in all the mentioned investigations have been unselected, which means that, eg, twin pregnancies with various complications have been included. All investigations, except one [3], have included growth-retarded fetuses. These were, however, defined on a singleton weight scale.

The purpose of the present investigation has been to prepare a reference interval for hPL and dU-E for twin pregnancies considered to be normal by obstetric criteria, in order

to obtain a more restricted reference interval with a possible greater applicability in disclosing intrauterine growth retardation.

MATERIAL AND METHODS

The investigation is a study of 145 consecutive twin pregnancies, terminated at the Department of Obstetrics and Gynecology, Aarhus University Hospital, from 1 January 1977 till 1 October 1980. The material was divided into the following three groups:

1) A group of 43 normal twin pregnancies used to define the normal reference intervals for dU-E and hPL. This group included pregnancies fulfilling the following conditions: no chronic disease, no complications of pregnancy at all (eg, bleeding, hypertension and preeclampsia), time of gestation at delivery exceeding the 35th week, both children with a birthweight exceeding the 10th percentile on a curve showing the relation between gestational age and birthweight for every twin in the material (Fig. 1), maternal age at conception less than 36 years, and parity less than 5. dU-E was monitored in 35 pregnancies where the above mentioned criteria were fulfilled (a total of 226 24 hr urine measurements, 1-20 measurements (mean 6.5) on each woman). hPL measurements were not performed until the 1st of October, 1977. For this reason, hPL was monitored in 24 pregnancies only (a total of 150 serum assays, 1-14 masurements (mean 6.3) on each woman).

2) A group of twin pregnancies where one or both newborn babies were found light for date (birthweight below the 10th percentile Fig.1)). A total of 34 babies were light for date, and were found in 26 pregnancies. In 18 of these pregnancies, dU-E was measured (a total of 106 24 hr urine measurements, 1-18 measurements (mean 5.9) on each woman), while hPL was measured in 14 of these pregnancies (a total of 62 serum assays, 1-12 measurements (mean 4.4) on each woman).

3) This group comprises the rest of the material, ie, pregnancies not used to define the normal hormone reference interval and pregnancies where none of the babies were found light for date. A total of 76 pregnancies were found in this group. In 40 of these, dU-E was measured (a total of 332 24 hr urine measurements, 1-20 measurements (mean 8.3) on each woman), while in 35 pregnancies, hPL was measured (a total of 253 serum assays, 1-20 measurements (mean 7.2) on each woman).

The total material was divided according to type of placentation and zygosity. The placentae were studied thoroughly after delivery and the number of chorii and amnii determined. In 16 cases, no information was available on placentation. Zygosity assessment was based on sex and placentation: therefore dizygosity could be determined only in cases of unlike sex and monozygosity only in cases of monochorionic placentae.

dU-E was measured using a Technicon Fluoro Nephelometer [12]. hPL was measured by radio immunoassay using reagents from Amersham plc. For dU-E the total imprecision (interassay) was below 5% (CV), and for hPL below 7% (CV) in the relevant ranges.

The normal range curves for both singleton and twin pregnancies were based on longitudinally obtained curves.

Statistics

Reference intervals for dU-E and hPL for both singleton and twin pregnancies were derived by non-parametric statistical methods [13]. Beside this, χ^2 test was used with a level of significance of 5%.

RESULTS

The reference intervals for normal twin pregnancies as a function of gestational age are shown in Fig. 2. Reference intervals are the 90% confidence intervals. The dotted lines are the corresponding limits for normal singleton pregnancies. The values of the reference intervals for twins are higher and considerably wider. There is a remarkable overlap

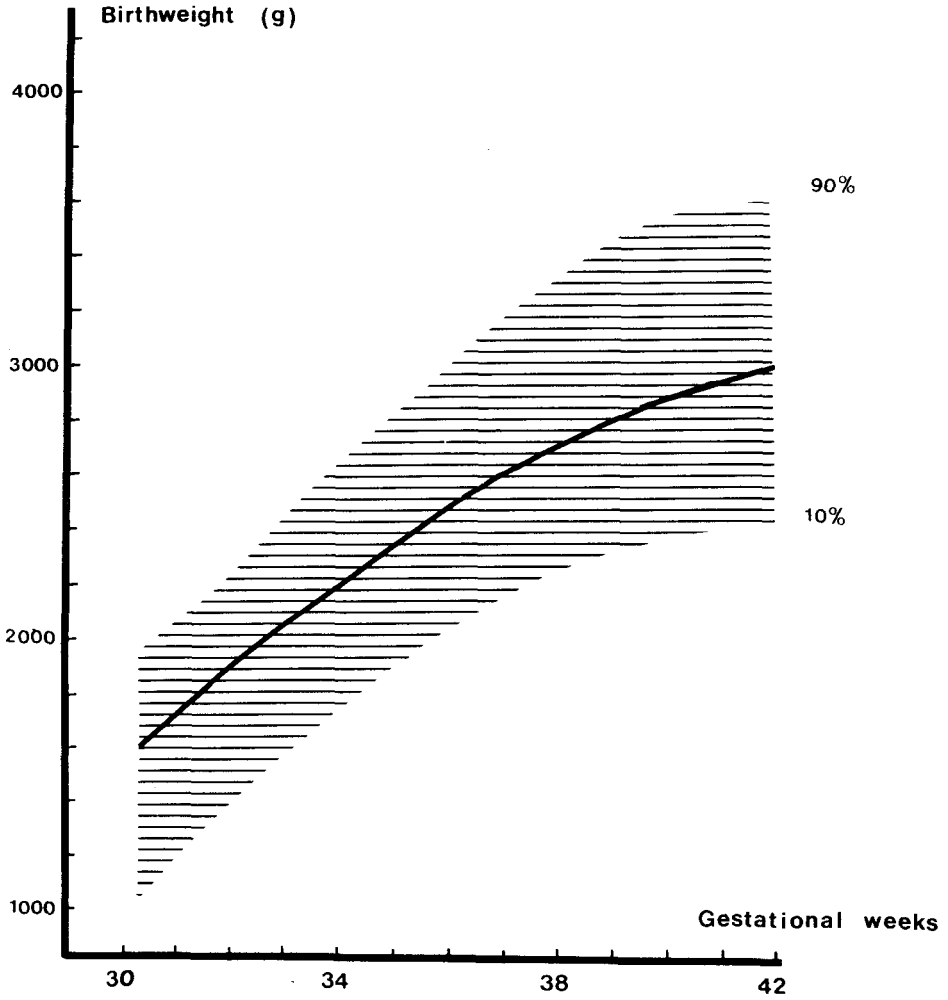

Fig. 1 - The curve shows the 10th and 90th percentile birthweight in grams in relation to gestational week for every twin in the material.

Table - Sensitivity and Specificity of the Hormone Analysis in Detecting Intrauterine Growth Retardation

One or more hormone values below the 5th percentile	Test result	Light for date (N)	Not light for date (N)	Sensitivity	Specificity
dU-E	Positive	7	12	0.39	0.70
	Negative	11	28		
hPL	Positive	7	5	0.50	0.86
	Negative	7	30		
Either dU-E or hPL	Positive	8	13	0.57	0.63
	Negative	6	22		
Both dU-E and hPL	Positive	4	2	0.29	0.94
	Negative	10	33		

https://doi.org/10.1017/S000156600000492X Published online by Cambridge University Press

between the reference intervals for normal singleton pregnancies and twin pregnancies for both parameters.

When the whole material was divided according to placentation and hormone measurements, we found significantly more hPL values below the median in monoplacental than diplacental pregnancies ($\chi^2 = 7.59$, $P < 0.01$; one placenta: 12 above, 33 below; 2 placentae: 45 above, 42 below). In the same way, significantly more hPL values were found below the median in MZ than DZ pregnancies ($\chi^2 = 6.91$, $P < 0.01$; MZ: 5 above, 16 below; DZ: 21 above, 14 below). No similar significant difference could be found for dU-E values.

In Fig. 3, hormone values from pregnancies with one or both babies light for date are plotted with the reference curves. A large fraction of the hormone values falls within the normal confidence limits for twins.

The benefits of measuring the two hormone parameters in order to identify intrauterine growth retardation are expressed in terms of specificity and sensitivity. The results can be seen in the Table. A positive test result is defined as at least one hormone value (dU-E or hPL) below the lower limit in the normal reference curves (Fig. 2).

Monoplacentation was significantly more frequent ($\chi^2 = 4.60$, $P < 0.05$) in pregnancies with one or two light for date babies (20 one placenta, 5 two placentae, 1 unknown) than in pregnancies with no light for date baby (45 one placenta, 59 two placentae, 15 unknown). No significant difference could be found according to zygosity, but a tendency was seen towards more MZ pregnancies among those with light for date babies (11 MZ, 6 DZ), compared to those with no light for date babies (29 MZ, 29 DZ).

DISCUSSION

hPL and estrogens are hormones both produced in the syncytiotrophoblast, the latter partly from fetal precursors. The synthesis and secretion of hPL is considered to be autonomous [15], and changes in the hPL values are therefore supposed to express changes in placental function. The relation between placenta and estrogens is more complex, because the serum and urine concentrations of the hormones are the results of fetoplacental interrelationships.

As expected, the hormone values are found to be higher in twin than in singleton pregnancies [1-3,7-10,15]. The twin pregnancies in previous studies were not all normal, but included undefined pregnancies, pregnancies with complications, and pregnancies where one or both fetuses were growth-retarded. In a single study [3], cases with intrauterine growth retardation were isolated, using, however, a singleton weight scale. The reference intervals were wide, perhaps because of the heterogeneous materials used in these studies. However, the reference intervals in the present study are quite similar. This indicates that the placental hormone production shows great variation even in normal twin pregnancies. The fact that it was urine estrogens that were measured and not serum estriol, total or unconjugated, should not modify this conclusion [14].

It is interesting to note that monoplacental pregnancies show lower hPL values than diplacental pregnancies, and the same is true for MZ pregnancies compared to DZ pregnancies. This finding implies that hPL production differs in monoplacental and diplacental twin pregnancies. One would expect the placental weight of monoplacental pregnancies to be less than the combined placental weight of diplacental ones.

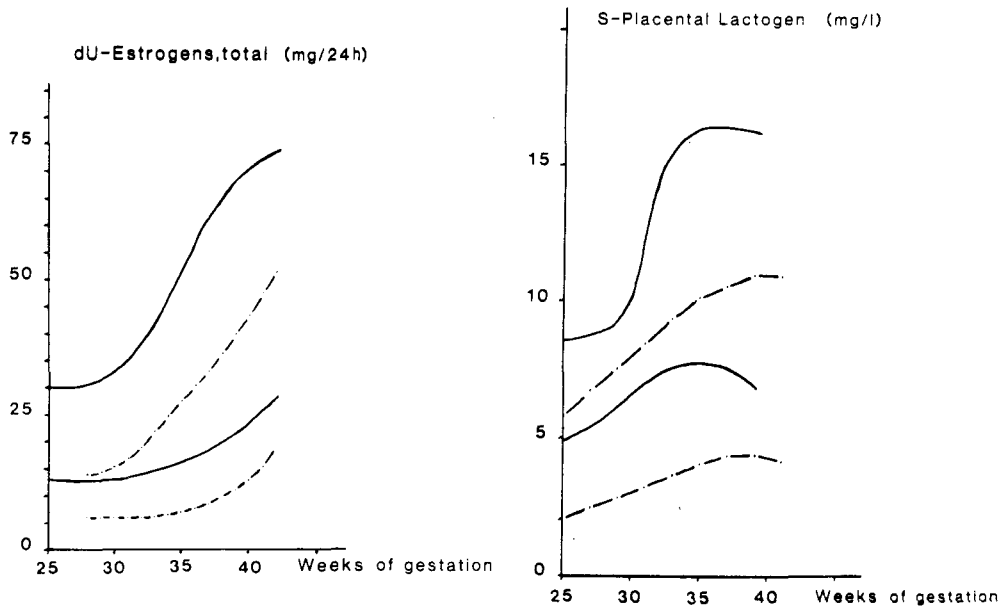

Fig. 2 - The reference intervals for dU-estrogens, total (mg/24 hr) and S-placental Lactogen (mg/l) as a function of gestational age are shown. The reference intervals are the 90% confidence intervals. The dotted lines are the corresponding limits for normal singleton pregnancies.

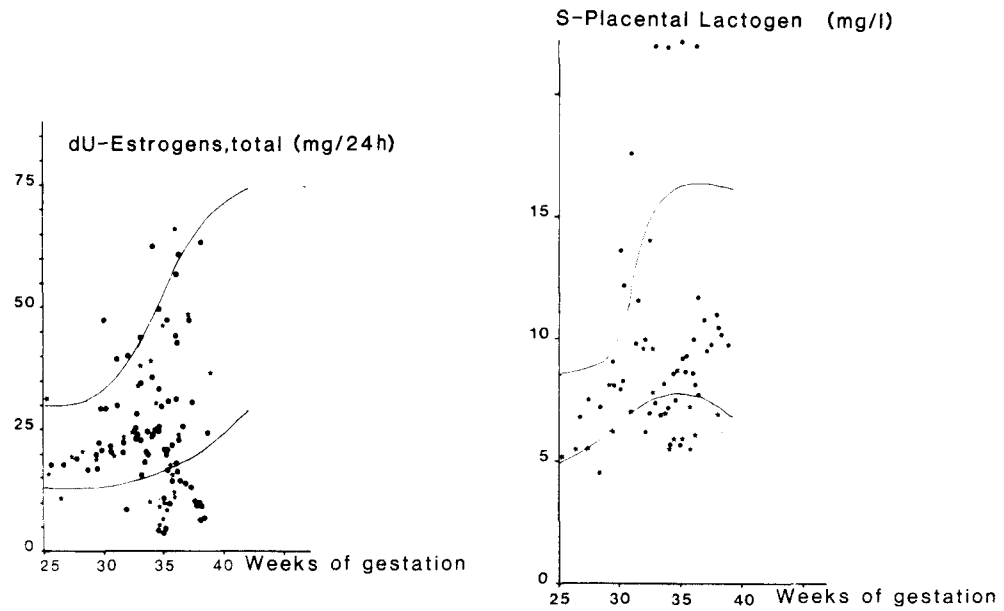

Fig. 3 - Hormone values for those pregnancies where at least one child was born as light for date are plotted on the reference curves.
• One child light for date
* Both children light for date.

https://doi.org/10.1017/S000156600000492X Published online by Cambridge University Press

Similar results did not apply to dU-E values, perhaps because estrogens are the results of fetoplacental interrelationships and not a direct expression of placental function.

The evaluation of hPL and dU-E values in twin pregnancy is complicated by the fact that hormone concentrations result from the combined conditions of the two fetuses. The determination of fetal distress and/or intrauterine growth retardation for one twin is difficult, even when a series of hormone values is available.

The Table 1 shows the specificity and sensitivity for dU-E and hPL in detecting intrauterine growth retardation. It can be seen that none of the combinations – low dU-E alone, low hPL alone, either low dU-E or low hPL, or both low dU-E and low hPL – reveals a sensitivity and specificity sufficient to let a hormone analysis be the only screening procedure for intrauterine growth retardation. No combination could disclose more than 57% of the growth retarded pregnancies. The figures for specificity show that up to 37% (1-0.63) of the not growth-retarded pregnancies would be included, unless the sensitivity is accepted to be as low as 29%.

Intrauterine growth retardation is an important, but difficult diagnosis to make when dealing with twin pregnancies. Prediction of intrauterine growth retardation by biparietal measurements is stated to be of limited value too [6], because of a significant number of false positive and negative results. Ultrasonically measured head/abdominal circumference ratios are found to be of greater specificity for the condition [4], and may represent the method of choice.

REFERENCES

1. Batra S, Sjöberg NO, Aaberg A (1978): Human placental lactogen, estradiol-17β, and progesterone levels in the third trimester and their respective values for detecting twin pregnancy. Am J Obstet Gynecol 131:69-72.
2. Braunstein GD, Karow WG, Gentry WC, Rasor J, Wade ME (1978): Third trimester chorionic gonadotropin measurements as an aid in the diagnosis of early pregnancy disorders. Am J Obstet Gynecol 131:25-32.
3. Briët JW, Horn RKJ (1982): The use of human placental lactogen, oxytocinase and estriol in twin pregnancies with intrauterine growth retardation. Eur J Obstet Gynecol Reprod Biol 13:7-14.
4. Crane JP, Kopta MM (1979): Prediction of intrauterine growth retardation with ultrasonically measured head/abdominal circumference ratios. Obstet Gynecol 54:597,603.
5. Daw E (1977): Human placental lactogen and twin pregnancy. Lancet 2:299-300.
6. Divers WA, Hamsell DL (1979): The use of ultrasound in multiple gestations. Obstet Gynecol 53:500-504.
7. Duff GB, Brown JB (1977): Urinary oestriol excretion in twin pregnancies. J Obstet Gynecol Br Commonw 81:695-700.
8. Gennser G, Grennert L, Kullander S, Persson PH, Wingerup L (1975): Human placental lactogen in screening ofr multiple pregnancies. Lancet 1:274.
9. Gerhard I, Runnebaum B (1980): Hormonbestimmungen bei Mehrlingsschwangerschaften. Geburtsh Frauenheilk 40:101-115.
10. Grennert L, Gennser G, Persson PH, Kullander S (1976): Ultrasound and human-placental lactogen screening for early detection of twin pregnancies. Lancet 1:4-6.
11. Kappel B, Brogaard Hansen K, Møller J, Faaborg-Andersen J (1984): Gemelligraviditet. Obstetriske og neonatale problemer. Ugerskr Laeg 146:3448-3452.
12. Lever H, Powell JC, Peace SM (1973): Improved estriol determination on a continous flow system. Biochem Med 8:188-191.
13. Reed AH, Henry RJ, Mason WB (1971): Influence of statistical method used on the resulting estimate of the normal range. Clin Chem 17:275-284.

14. Svenstrup B (1976): Radioimmunologisk bestemmelse as frit φstriol i serum under graviditet. Ugeskr Laeg 183:1075-1077.
15. Ylikorkala O (1973): Maternal serum hPL levels in normal and complicated pregnancy as an index of placental function. Acta Obstet Gynecol Scand (Suppl) 26:1-52.

Correspondence: Dr. Birgitte Kappel, Cytogenetic Laboratory, Aarhus Psychiatric Hospital, DK-8240 Risskov, Denmark