SERUM OXYTOCINASE LEVELS IN NORMAL AND ABNORMAL PREGNANCIES

by

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Summary

A study was carried out to relate levels of serum oxytocinase to fetoplacental function in normal and high-risk pregnancies. Mean levels and the normal range obtained during different weeks of gestation in 140 samples from normal term pregnancies are graphically represented. Values obtained in 269 samples in 157 cases from the high risk pregnancy group were compared to the control group and results were interpreted in relation to the fetal outcome and placental weight.

This study indicates the importance and value of assessing this enzyme during the later months of pregnancy, as it was found to be a satisfactory index of placental function and a valuable guide to the clinician in the management of such patients.

Introduction

Placenta is the major site of production of hormones and enzymes during pregnancy. In this study the functional significance of one of its enzymes Cystine amino peptidase (CAP) or oxytocinase (O) is presented.

A direct relationship between serum oxytocinase levels and placental functions has been referred to by several investigators (Josephides and Turkington, 1967; Malkani *et al*, 1971; Ryden, 1972 and Curzen and Varma, 1973). Since this enzyme diffuses directly into the maternal serum, its assay could prove to be a better index of placental function than the commonly accepted methods of estimating urinary metabolites of hormones. Serum oxytocinase assay being a simple and rapid method and requiring only 0.2 ml of serum sample may provide the clinician with a more practical method of assessing and managing high-risk pregnancies. Estimation of urinary hormones involve lengthy procedures and more expense even though the latter may be equally or more reliable.

Thus, the object of this study was to see how closely the serum oxytocinase levels parallel the placental function in normal and abnormal pregnancies.

Material

This study was carried out in two groups:

Group I: This group comprised of women who had normal term pregnancy. One hundred and fifty-one samples from 60 such women were collected during different weeks of gestation. The mean of values obtained during different weeks are graphically represented (Fig. 1).

Group II: This group consisted of 269 samples from 157 subjects who were considered as high-risk because of toxemia,

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placental insufficiency or premature labour. In some cases serial estimations in consecutive weeks were carried out.

The results in this group were compared with the control and correlated with the fetal weight, its well being, placental changes and obstetric complications, if any.

Methodology

The enzyme Oxytocinase i.e., cystine amino peptidase breaks the cystine-tryosine linkage of oxytocin in vivo. The same principle is used in vitro for this assay. A rapid and modified method of Babuna and Yenen (1966) was used in this study. The final reaction involved conversion of a diazotised compound to an azodye compound, the optical density of which could be read in a calorimeter at 565 mμ. The results were expressed in oxytocinase units. The assay was carried out on morning blood samples. All hemolysed sera were discarded.

Statistical analysis of this control group was done by using standard deviation and standard error. The normal curve was graphically represented by joining the mean values ± 2 S.E. during different weeks of gestation. The results in group II were compared to control group I for corresponding period of gestation. Values less than the mean minus 2 standard error were considered "low" values and less than the mean minus 3 standard error were considered "very low".

Results

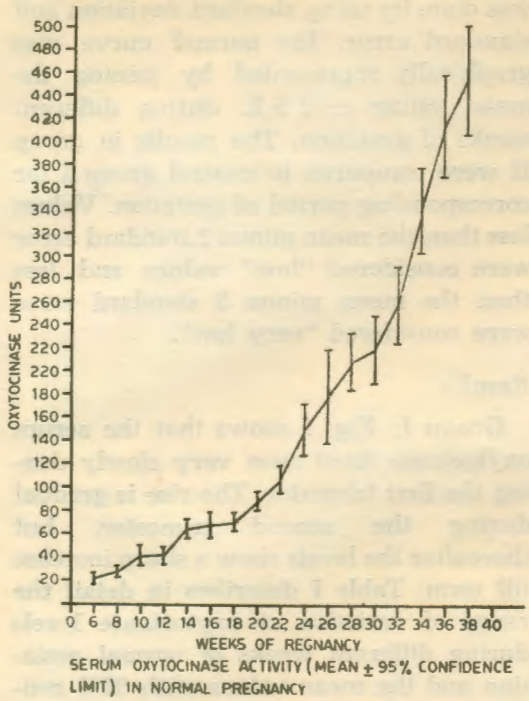
Group I: Fig. 1 shows that the serum oxytocinase level rises very slowly during the first trimester. The rise is gradual during the second trimester, but thereafter the levels show a sharp increase till term. Table I describes in detail the range of variation of oxytocinase levels during different weeks of normal gestation and the mean values with 95% confidence limits (mean ± 2 S.E.).

TABLE I

Serum Oxytocinase Activity (In Units) during Different Weeks of Pregnancy

Pregnancy weeks	No. of samples	Oxytocinase values		S.D.	S.E.	95% Confidence limit
		Range	Mean			Mean ± 2 S.E.
Control	6	13-37	25.33	6.536	2.67	30.66- 20.06
6	9	11-32	21.22	6.27	2.33	25.87- 16.57
8	10	16-36	27.2	6.54	2.07	31.33- 23.04
10	7	20-56	41.28	10.88	4.11	49.05- 33.06
12	8	23-62	41.5	12.56	3.53	48.55- 33.45
14	8	42-81	64.0	11.31	4.0	72.0 - 56.0
16	8	40-97	68.0	16.15	5.71	79.42- 57.58
18	7	62-100	70.0	10.35	3.9	77.8 - 62.2
20	7	62-131	88.1	25.6	4.145	96.4 - 79.8
22	8	81-143	107.4	16.34	5.78	118.97- 95.84
24	10	86-215	148.7	34.21	10.81	170.2 -127.0
26	5	102-240	177.2	44.93	20.1	217.4 -137.0
28	9	140-251	207.1	36.62	12.21	231.42-182.58
30	6	181-267	217.6	35.32	14.4	246.4 -188.8
32	12	168-340	257.1	53.28	16.81	290.7 -223.5
34	8	292-456	338.8	49.99	17.67	374.14-303.46
36	11	324-523	412.1	73.77	22.19	456.48-367.72
38-40	12	347-569	452.41	83.46	24.12	500.65-404.17

FIG. 1



SERUM OXYTOCINASE ACTIVITY (MEAN ± 95% CONFIDENCE LIMIT) IN NORMAL PREGNANCY

Group II: Table II shows the number of cases and the number of sample studied in different clinical conditions.

Table III shows the serum oxytocinase levels in relation to the different clinical conditions.

Bad Obstetric History

There were 75 cases of B.O.H.—a sizable group. Only 13% of them showed low levels. the fetal outcome, however, in all cases turned out to be normal, probably due to better antenatal surveillance.

The abortion group consisted of 23 cases, 10 of which were clinically threatening to abort and the rest were either suspected of missed abortion or were cases of habitual abortion during the second trimester. Fig. 2 shows the values obtained in this group and in suspected cases of vesicular mole. All 10 cases of threatened abortion showed very low levels of this enzyme and all of them eventually aborted.

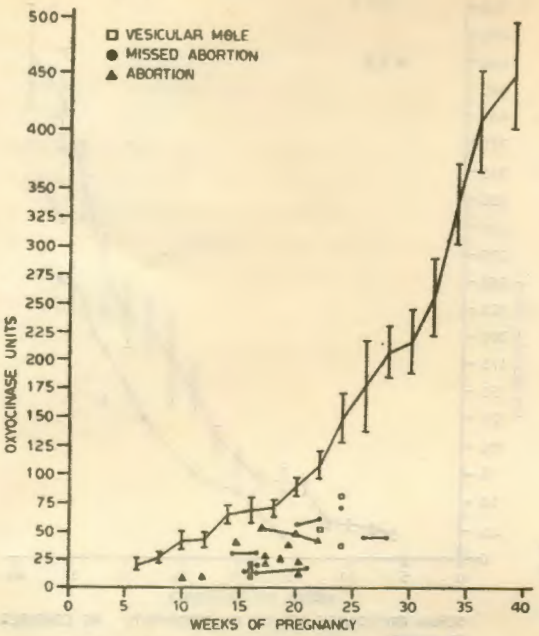
TABLE II
Sample Material for Study

Clinical condition	No. of cases	Serum samples
B.O.H.	75	130
Abortion	23	29
Vesicular mole	6	6
Toxemias	26	54
I.U.G.R. (Placental insufficiency)	16	30
Hydramnios	4	13
Post-date pregnancy	7	7
Total	157	269

TABLE III
Serum Oxytocinase Levels in Relation to Clinical Condition

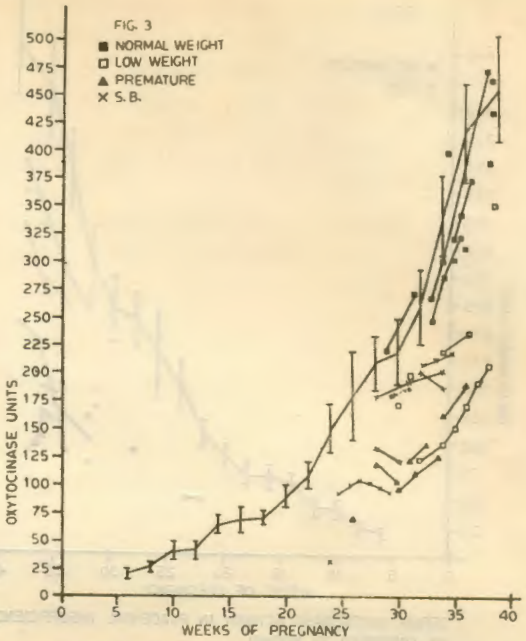
Clinical condition	No. of cases sample	Oxytocinase Values	
		Normal values	Abnormal values
B.O.H.	75/130	65 (87%)	10 (13%)
Abortion	23/29	—	23
Vesicular mole	6/6	—	6
Toxemias	26/54	6 (30%)	20 (70%)
I.U.G.R. (Placental insufficiency)	16/30	—	16
Hydramnios	4/13	—	4
Post-date pregnancy	7/7	7	—

FIG. 2



SERUM OXYTOCINASE ACTIVITY IN ABORTION GROUP AND VESICULAR MOLE AS COMPARED TO NORMAL

FIG. 3

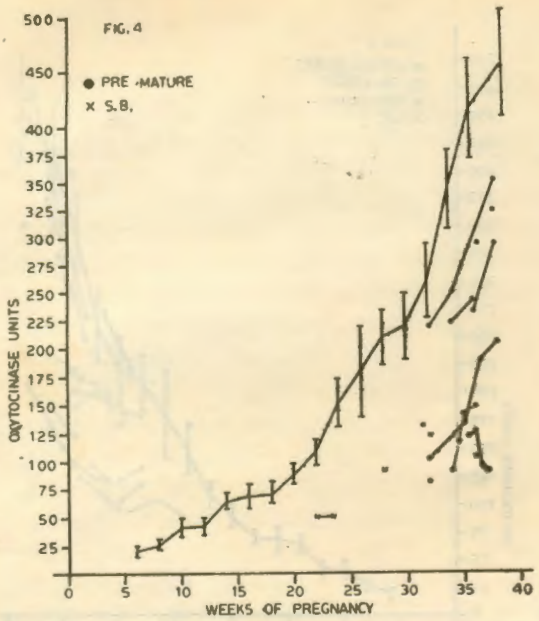


SERUM OXYTOCINASE ACTIVITY IN TOXEMIA AS COMPARED TO NORMAL

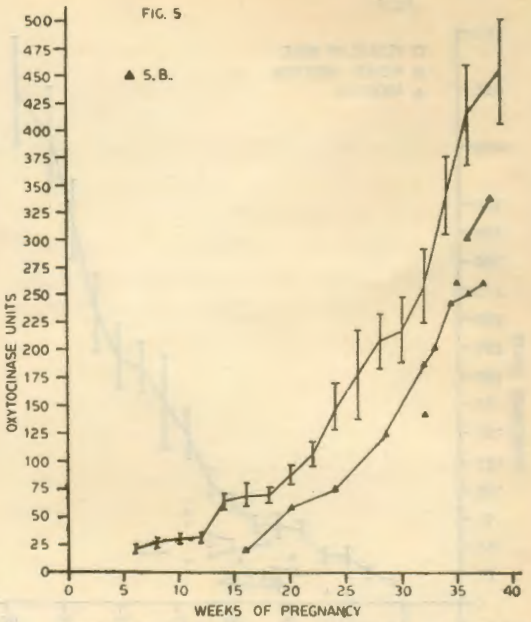
The results also demonstrate that low levels which fail to rise in subsequent samples, indicated a poor clinical prognosis. The cases which were diagnosed as vesicular mole showed "very low" levels of this enzyme.

Toxemia: There were 26 cases presenting with different degrees of toxemic symptoms during the third trimester. 54 serum samples were obtained in this group. Abnormal values were detected in 20 (70%) out of 26 cases. Fig. 3 clearly demonstrates that normal fetal outcome was associated with levels of serum oxytocinase which were rising and within the normal range. Babies with low birth weight or premature showed low or very low values for the period of gestation. Level which tended to rise in subsequent samples was considered to be favourable prognosticating index. Cases which resulted in stillbirth not only showed very low values, but subsequent samples failed to show a significant rise. Most of the stillbirths were associated clinically with severe toxemia or eclampsia.

Placental Insufficiency: (I.U.G.R.): There were 16 cases in this group and 30 samples were assayed. All the samples showed abnormally low values. Fig. 4 graphically indicates the low values obtained in this condition. Stillbirth cases showed very low values and in most cases only single samples were available in those subjects. The cases where premature babies were born showed that serial samples in most of those cases showed a rising curve though the values were well below the normal range (less than—2 S.E.). Whenever the values tended to flatten out in subsequent samples, the clinician was advised to intervene and save the baby by a caesarean section. In 3 such cases, timely advice



SERUM OXYTOCINASE ACTIVITY IN PLACENTAL INSUFFICIENCY AS COMPARED TO NORMAL



SERUM OXYTOCINASE ACTIVITY IN HYDRAMNIOS AS COMPARED TO NORMAL

and intervention resulted in a live fetus.

Hydramnios: There were only 4 cases in this category and 13 samples were obtained. All the sample levels were very much below the normal range. The fetal outcome was poor in all cases. These were associated with anencephaly, hydrocephalus and fetal anomalis. Fig. 5 graphically shows the values in this group in relation to the normal range. It appears that in this group in spite of the rising curve of serum oxytocinase seen in 2 such cases, the fetal prognosis turned out to be poor.

Post-date Pregnancy: There were 7 subjects in this group who had gone over by 10-15 days. The serum oxytocinase was within the normal range in all cases. All the cases had a normal fetal outcome.

Table IV presents a comprehensive assessment of the relation between the serum oxytocinase levels and the pregnancy outcome in all cases of high risk pregnancy.

TABLE IV

Relation of Oxytocinase Values to Pregnancy Outcome

Fetal outcome	No. of cases	Normal value	Low value Mean — 2 S.E.	Very low value Mean — 3 S.E.
F.T.N.D.	73	63 (86.3%)	10 (13.7%)	—
Abortion	8	—	—	8
Vesicular mole	5	—	—	5
Low weight baby	5	—	1	4
Stillbirth	12	—	—	12
Premature fetus	15	—	—	15

It clearly demonstrates that normal fetal outcome was associated with normal oxytocinase levels in 86.3% of cases. In the remaining cases, eventhough the absolute values were lower than normal, the subsequent sampling had demonstrated a rising trend. Abnormally low or very low values were associated with either low weight baby, prematurity, stillbirth, fetal anomalies or premature termination.

Table V indicates the relation between serum oxytocinase levels and placental weight. Normal weight and appearance of placenta was associated with normal oxytocinase values in 96.3% cases. In cases where the placenta was found to be really small at parturition, the levels of oxytocinase were found to be very low as in 77.4% cases. Infarcted placenta was associated with very low oxytocinase levels in 100% of the cases.

TABLE V

Relation of Oxytocinase Values to Placental Weight

Type of Placenta	No. of cases	Normal value	Low value Mean — 2 S.E.	Very low value Mean — 3 S.E.
Normal placenta > 400 gm	80	77 (96.3%)	7 (3.7%)	—
Small placenta < 400 gm (Mean 227 gm)	31	—	7 (22.6%)	24 (77.4%)
Infarcted placenta	4	—	—	4 (100%)

Discussion

A normally functioning placenta is a very basic requirement for a normal pregnancy and fetal outcome. The increased concentration of CAP in the blood during pregnancy probably reflects increased protein metabolism. Since this enzyme is synthesized in the placenta, in could be used as an indicator of placental growth and function. Its exact physiological role is not yet clear since the true physiological substrates for this enzyme are not known. In general, since the enzyme has a slow half life (Klimek, 1968), its levels may not be consistent with functional activity. There are conflicting reports regarding its correlation to placental weight, infant weight and toxemia of pregnancy (Spellacy *et al*, 1977). However, the ease and simplicity of the testing system continues to encourage the investigator to assess its usefulness in high risk pregnancy.

The present study in normal pregnancy has achieved the following objectives:

(1) The norms for Indian women are established.

(3) The control values obtained during different stages of gestation compare favourably with those published from developed countries (Hurry *et al*, 1972. Carter *et al*, 1974; Spellacy *et al*, 1977).

(3) The slope of the rise in enzyme level was marked after 16 weeks of gestation and showed a rapid decline after parturition.

Thus its usefulness may be limited to later months of pregnancy.

Its scope in the high-risk pregnancy group seems fairly well-substantiated by

the results of this study. There appears to be significant correlation between the placental weight, its degree of infarction and serum oxytocinase levels. Spellacy *et al* (1977) have also shown good correlation between HPL and O with placental weight and toxemia of pregnancy.

In our study, 70% of cases with toxemia of pregnancy showed significant lowering of CAP levels. Among those with very low serum oxytocinase levels and especially where subsequent sampling failed to show a rise the perinatal outcome in 14 out of 20 such cases was poor (Fig. 3). The placenta in those cases were found to be infarcted or very small. One may point out at this stage that one cannot prognosticate on the basis of a single test of oxytocinase, even when found to be very low. In 5 cases from the toxemic group, normal placenta and term babies were born even though single sampling had shown low serum oxytocinase levels.

In cases of I.U.G.R. or second trimester abortions and vesicular mole where the placental growth and function are affected, the oxytocinase levels were low in all cases. The placenta in all such cases were found either infarcted or very small (Hensleigh and Krantz, 1970). There were no full term babies in this group. Premature babies were born in cases where subsequent sampling had shown a rising curve even though the absolute values were much below the normal range.

All 4 cases of hydramnios showed very low values even though in 2 cases subsequent sampling had shown a rising curve. The fetal outcome in all cases was poor probably because of associated anomalies. The placenta in all 4 cases were small and infarcted resulting in low oxytocinase levels.

It must be admitted, however, that oxytocinase levels alone cannot prognosticate on fetal anomalies or stillbirth. It can give warning signals with regard to placental function, but because of its slow half life a single test may not be able to indicate an impending danger to fetal life.

However, repeat serum sampling during high-risk pregnancy along with clinical parameters can be effectively used by the clinician to monitor feto-placental function and the decision for timely intervention.

The data in this study enlarges the scope and the need for developing simple and rapid enzyme tests in high risk pregnancies. These tests could probably be further simplified like a glucose strip test in urine which could be used to screen all high risk cases at frequent intervals. The more suspicious amoung those could then be subjected to more sophisticated testing for evaluating the feto-placental unit more precisely.

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