

SERUM HISTAMINASE IN PREGNANCY

by

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Distribution in tissues of histaminase, an enzyme concerned with the oxidative deamination of histamine (Tabor, 1951; Tabor *et al.*, 1953), has been studied in a number of animal species (Ahlmarm, 1944; Anrep *et al.*, 1947; Haegar *et al.*, 1952). The blood of human beings has very little histaminase activity; however, during pregnancy, the enzyme activity in the serum rises markedly to about 400 to 1000 times the amount observed in the serum from non-pregnant women (Tabor, 1954). The dramatic rise of histaminase in the serum during pregnancy appears to be restricted almost entirely to human beings (Anrep *et al.*, 1947). Rise of this order of magnitude is not associated with any other known physiological or pathological state, except bronchial carcinoma (Casati *et al.*, 1963). The plasma titre of histaminase has, therefore, been proposed as a diagnostic test for pregnancy (Werle and Effkemann, 1940; Zeller and Birkhauer, 1940; Anrep *et al.*, 1947; Kobayashi, 1963).

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The significance of raised histaminase activity in the serum during pregnancy has remained obscure. The high enzyme levels may be responsible for the protection of the foetus from a possible histamine intoxication (Kapellar-Adler, 1966). Histaminase may also play a more basic role in intracellular metabolism and transport of ions and molecules (Tabor, 1954; Zeller, 1952).

Abnormally low histaminase activity in the serum has been reported in patients of severe pre-eclamptic toxæmia, hyperemesis gravidarum and threatened abortion (Kapellar-Adler, 1966; Anrep *et al.*, 1947).

The present work was undertaken to study histaminase activity in the serum during normal pregnancy, and during pregnancy complicated with pre-eclampsia and/or anaemia. An attempt is also made to evaluate the influence of treatment of anaemia and/or pre-eclampsia on the enzyme activity in the serum.

Material and Methods

Histaminase activity was estimated in the serum samples obtained from female subjects attending the Obstetric and Gynaecological Department of Shree Sayaji General Hospital, Baroda. No attention was given to the

age or parity of the subjects. The method of estimation was as described by Kapeller-Adler (1951). The enzyme activity was expressed in terms of "Permanganate Units"/ml. One Permanganate Unit represents the amount of enzyme which (after incubation for 24 hours with 1 mg histamine dihydrochloride at pH 7.2 and 37°C in an atmosphere of oxygen and aqueous solution of indigo disulphonate) takes up 0.1 ml of 0.002 N potassium permanganate solution. Tests were done in duplicate within 36 hours of collection of the blood samples.

Histaminase activity was measured in the serum obtained (a) from non-pregnant women, (b) from pregnant women with normal pregnancy of different durations, (c) from pregnant women having anaemia, (d) from pregnant women having pre-eclamptic toxæmia, (e) from parturient women immediately after the third stage of labour, and (g) from postpartum women during the first four days of puerperium. Whenever the enzyme activity was estimated in the maternal serum obtained immediately after delivery, the enzyme activity in the corresponding cord blood sample was also estimated. Enzyme activity was also measured in the serum of patients of eclampsia and/or anaemia undergoing suitable treatment.

Normal pregnancy was taken as the one in which the subject had blood pressure not exceeding 125/80 mm/Hg, was free from antepartum hæmorrhage, cardiac or renal disease, and diabetes mellitus and in which there was a reasonable agreement between the gestational period and

the uterine size. The haemoglobin level of these subjects ranged from 9.0 g. per cent to 12.5 g. per cent. The low haemoglobin level might partly have been due to haemodilution which occurs during pregnancy. Krishna Menon (1965) has reported that the course of pregnancy is not adversely affected by anaemia of this order of magnitude. These subjects were, therefore, considered to have 'normal' pregnancy.

The duration of pregnancy was calculated from the first day of the last menstrual cycle in the case of women who had regular menstrual cycles. In other cases, the duration was estimated from the uterine size.

The existence of one or more of the following manifestations during pregnancy of over 24 weeks' duration prompted a diagnosis of pre-eclampsia: a systolic blood pressure of 140 mm/Hg or more, or a diastolic blood pressure of 90 mm/Hg or more, recorded twice at intervals of 6 hours; significant proteinuria at least on two consecutive days and a persistent oedema of hands or face. Pre-eclampsia was considered as severe if systolic blood pressure was 160 mm/Hg or more; if diastolic pressure was 110 mm/Hg or more; if proteinuria was severe (+++ on qualitative testing) or if there were cerebral or visual disturbances. Otherwise it was graded as mild.

Patients of hypochromic microcytic anaemia received iron (orally or parenterally), multivitamin tablets and a high protein diet. Patients of pre-eclampsia received low salt diet (600 mg. sodium chloride daily). A benzothiadiazine diuretic was given if the oedema was severe. If the

blood pressure continued to remain high the patients were given reserpine orally.

Results

Serum histaminase activity in non-pregnant women:

Sera of 3 non-pregnant healthy women and 5 women with gynaecological complaints (leucorrhoea, dysmenorrhoea and uterine prolapse) did not exhibit any enzyme activity when examined repeatedly in different phases of the menstrual cycle.

Serum histaminase activity during normal pregnancy, after delivery and during the puerperium:

Fig. 1 shows the enzyme activity in each of the 86 samples of serum obtained from women with different gestational periods, in each of the 4 samples obtained immediately after normal delivery and in each of the 27 samples obtained during the first 96 hours of the normal puerperium. The mean enzyme activity in the serum for each 4-week period during normal pregnancy is shown in Fig. 2. It is seen that histaminase activity in the

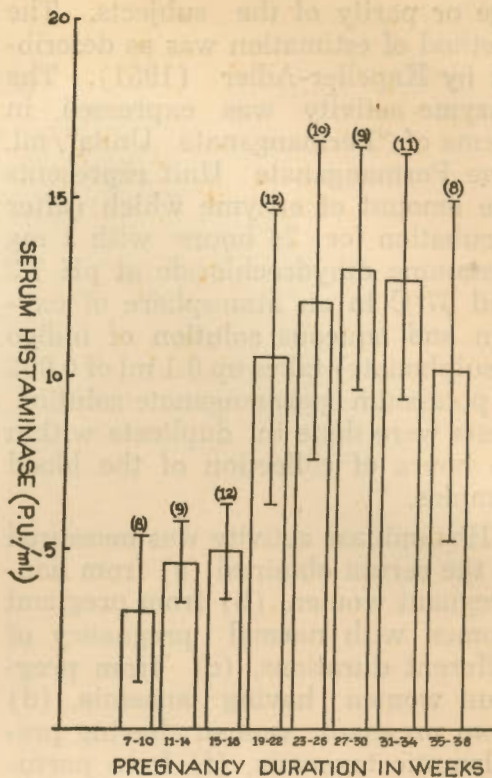


Fig. 2

Histogram showing mean histamine activity in Permanganate Units (P.U.) in the serum during normal pregnancy. Each bar represents the mean activity over a 4 weeks period. Figures in parentheses above each bar indicate the number of observations whose mean the bar represents. Vertical lines indicate the standard deviation.

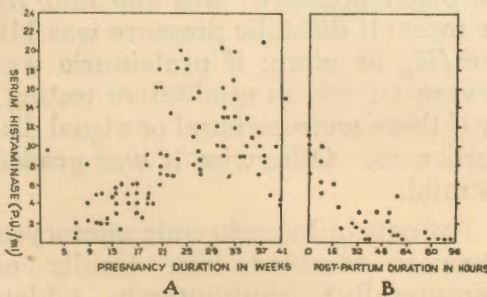


Fig. 1

Scatterdiagram showing histaminase activity in Permanganate Units (P.U.) in serum during normal pregnancy (A) and after delivery (B). Each closed circle represents the enzyme activity in the serum of one patient.

serum starts appearing after 6 to 10 weeks of pregnancy. The advance of pregnancy was initially accompanied by a progressive rise in the enzyme activity in the serum. The rise occurred in two phases. A steady rise occurred upto 18 weeks; thereafter, a steeper rise occurred upto 30 weeks. From 31 week onwards there was a moderate fall in the enzyme activity. Immediately after the third stage of labour the histaminase activity in

the serum was similar to that observed during the last 4 weeks of pregnancy. The enzyme activity was reduced approximately to 35 per cent by 24 hours and to 10 per cent by 48 hours after delivery. The enzyme activity was negligible by 80 hours after delivery.

In 3 of the 5 cord blood samples studied, the enzyme activity was low (1 to 2 P/U/ml); in the remaining 2, no activity could be detected.

Serum histaminase activity in pregnancy complicated by anaemia:

Of the 21 patients comprising this group, 9 had moderately severe anaemia (haemoglobin from 6.5 g. per cent to 8 g. per cent) and 12 had very severe anaemia (haemoglobin from 1.5 g. per cent to 4.5 g. per cent). Twenty patients had microcytic hypochromic anaemia and 1 had pleomorphic anaemia. All the patients also had variable degrees of hypoproteinaemia. In all but 3 patients the histaminase activity was within or very slightly lower than the range of the values for the corresponding normal pregnancy periods (Fig. 3). Even in these 3 patients the activity was within the range (minus 2 S.D.) observed in subjects with normal pregnancy. It must, therefore, be concluded that anaemia and also hypoproteinaemia does not significantly affect histaminase activity.

Serum histaminase activity in pregnancy complicated by pre-eclampsia:

Of the 5 patients studied, 3 had mild and 2 had severe pre-eclampsia. In 3 patients the histaminase activity in the serum was within the normal range; in 1 patient the activity was

high and in another it was low (Fig. 3). Severity of the disease did not seem to be related to the histaminase activity.

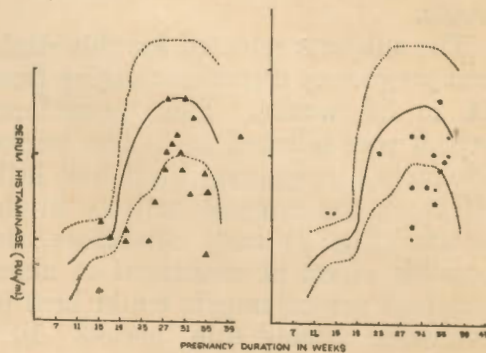


Fig. 3

Histaminase activity in Permanganate Units (P.U.) in the serum of each of 21 pregnant women having anaemia (A), 5 pregnant women having pre-eclampsia (o) and 11 pregnant women having pre-eclampsia and anaemia (*). Middle curves (—) represent mean serum histaminase activity (See Fig. 2) and upper and lower curves (---) were plotted by adding and subtracting value of one S.D. respectively from the corresponding control mean value.

Serum histaminase activity in pregnancy complicated by anaemia and pre-eclampsia:

Of the 11 patients studied, 9 had mild and 2 had severe pre-eclampsia and all had variable degrees of hypoproteinaemia. Anaemia was of microcytic hypochromic type in 10 patients and of macrocytic hyperchromic type in 1 patient. Anaemia was severe (haemoglobin from 1.5 g. to 4.5 g. per cent) in 10 patients and moderately severe in 1 patient.

In 10 patients the histaminase activity in the serum was within the normal range. In 1 patient the enzyme activity was found to be low but within the range of activity (minus

2 S.D.) observed in subjects with normal pregnancy (Fig. 3).

Effect of treatment of anaemia and pre-eclampsia on serum histaminase levels:

The subjects selected for this study had pregnancy duration ranging from 24 to 36 weeks. This gestational period was selected as in this period advancing pregnancy itself had little effect on the enzyme activity in the serum (Fig. 2) and, therefore, the possible effect of treatment of anaemia and pre-eclampsia could best be evaluated during this period. In 8 subjects with normal pregnancy, weekly fluctuations in the enzyme activity were found to be within 20 per cent of the initial value (Fig. 4).

In 6 patients of pregnancy with anaemia, the treatment of anaemia for 3 to 4 weeks induced considerable clinical and haematological improvement. In 2 patients the enzyme activity in the serum increased (more than 20 per cent of the initial level); in 4 patients the enzyme activity was within 20 per cent of the initial value (Fig. 4).

In 8 patients of pregnancy with anaemia and pre-eclampsia, treatment of anaemia and pre-eclampsia resulted in improvement of anaemia and pre-eclampsia. Fluctuations in the enzyme activity were wider during treatment than during normal pregnancy. At the end of treatment, the enzyme activity in serum was found to be reduced in 5 patients (30 to 50 per cent reduction). The fall occurred more or less simultaneously with reduction in proteinuria and oedema. The fall in enzyme activity was not related to the fall in blood pressure.

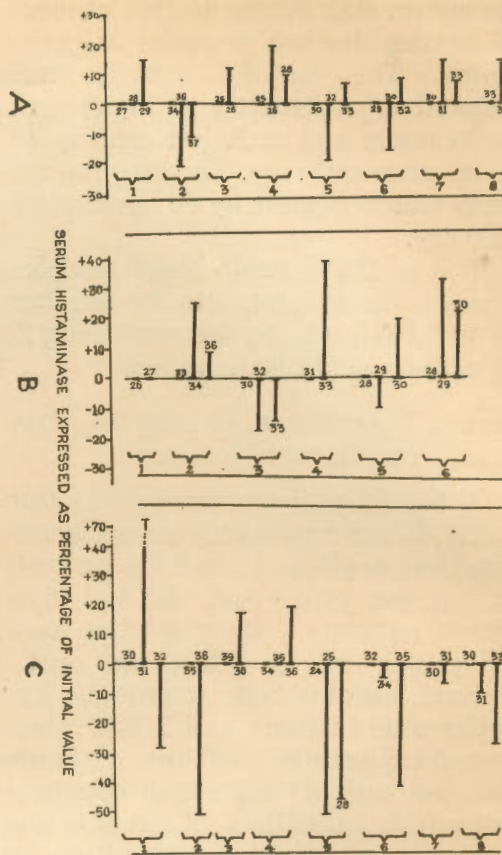


Fig. 4

Effect of treatment of anaemia and pre-eclampsia on histaminase activity on the serum of pregnant women during 24 to 36 weeks of pregnancy, (duration of pregnancy indicated in relation to each bar). A represents normal pregnancy, B represents pregnancy complicated with anaemia and C represents pregnancy complicated with anaemia and pre-eclampsia. Each bar represents per cent change of the enzyme level from the initial value. The figures to the right of each bracket indicate serial number of the patients of each group.

Discussion

Histaminase activity in the serum of non-pregnant and pregnant women has been studied by many workers using different methods of enzyme estimation. The data obtained in the

present study compare reasonably with those obtained by other workers (Table I). rapidly within 2 to 3 hours after birth. A rapid inactivation of the enzyme in the foetus could explain the

TABLE I
Summary of data obtained by different workers on serum histaminase activity in normal pregnancy

Reference	Enzyme activity in non-pregnant women	Gestational period at which activity first detected	Pattern of rise of enzyme activity during pregnancy
Ahlmark, 1944	Some activity	2 months	A steady rise upto 7 months, then a fall, and rise again till term.
Anrep et al., 1947	None	2 months	A steady rise throughout the gestational period reaching peak at term.
Kapeller-Adler, 1952	Negligible	2 to 3 months	A rise upto 6 or 7 months, a slight fall and then steady activity till term.
Dodge, 1952	Some activity	2 months	Activity maximal between 165 to 230 days, a fall and then steady activity till term.
Kobayashi, 1963	Some activity	8 to 14 weeks	—
Present study	None	8 to 10 weeks	Gradual rise upto 18 weeks, then a steeper rise upto 30 weeks (peak 27 to 30 weeks) followed by a moderate fall from 34 week onwards.

Histaminase activity in the serum immediately after labour was not different from that towards the end of the term. This agrees with the findings of Anrep *et al.* (1947) and Kapeller-Adler (1952). It appears, therefore, that there is no relationship between the process of labour and the enzyme activity in the serum.

In comparison with histaminase activity in the maternal serum, the cord blood showed lesser enzyme activity confirming the earlier literature reports (Wicksell, 1949; Kapeller-Adler, 1952; Swanberg, 1948).

Anrep *et al.* (1947) found that the serum of the foetus exhibits about one-third enzyme activity in comparison with the activity in the maternal serum and that the activity declines

transient activity in the blood of the foetus.

A similar rapid maternal inactivation must also be occurring, since 80 hours after the expulsion of placenta the enzyme activity of the maternal blood was found to be negligible. The fate of enzyme in the foetus and in the mother remains largely unknown. The half life of the serum proteins is 10 days. The rapid fall of enzyme activity cannot therefore, be solely attributed to its metabolic degradation.

The placenta is now generally considered to be the site of production of histaminase (Guido, 1942; Denforth and Graham, 1937; Anrep *et al.*, 1947; Ahlmark, 1944; Swanberg, 1948; Kapeller-Adler, 1944). The factors

maintaining a brisk production of the enzyme and its generous distribution to the maternal blood are, however, ill understood. It is therefore, not possible to explain the wide variation in histaminase activity in the serum observed at any given period of normal pregnancy in this and other studies (Ahlmark, 1944; Kapeller-Adler, 1952; Dodge, 1952). However, towards the end of the term the variations may probably be related to the variable senile changes occurring in the placenta.

Unless the normal range of histaminase activity in the serum for different periods of gestation is clearly defined in a large number of subjects, changes in the enzyme activity should not be utilised for diagnostic purposes in abnormal pregnancies. In the present study serum of some cases of anaemia exhibited somewhat lower enzyme activity. The treatment of anaemia did not affect the enzyme activity. It is concluded, therefore, that anaemia is not responsible for lower enzyme activity in the serum.

In pregnancy complicated by mild or severe pre-eclampsia, histaminase activity in the serum was within the normal range in most of the cases. This finding is in disagreement with the reports of other workers (Kapeller-Adler, 1951; Ahlmark, 1944), who found persistently low enzyme activity in the serum in this disease. In cases of pre-eclamptic toxæmia treated successfully no rise in histaminase activity in the serum could be demonstrated immediately after the treatment. On the other hand, there was a reduction in the enzyme activity in 5 of the 8 cases studied.

This reduction was seen simultaneously with the reduction in proteinuria and oedema. This observation needs to be verified in a larger number of patients.

Increased cortical function with raised production of glucocorticoids and mineralocorticoids (Venning, 1946; Heard *et al.*, 1946; Tobian, 1949; Gemzell, 1953; Bayliss *et al.*, 1955) including aldosterone, (Martin and Mills, 1956) occurs during normal pregnancy. Pre-eclamptic toxæmia is known to be characterised by a further accentuation of the cortical function (Tobian, 1949; Lloyd *et al.*, 1952; Cort *et al.*, 1951), lower urinary excretion of aldosterone (Rinslor and Rigby, 1957; Kumar *et al.*, 1959) than in normal pregnancy, elevated production of gonadotrophins and reduced level of oestrogens (Swyer, 1952) and progesterone (Landau and Lugibihl, 1958). It is suggested (Kapeller-Adler, 1952) that histaminase activity in the serum during pregnancy is controlled by an optimal balance of natural hormones. At least in animal experiments the controlling effect of adrenal cortex (Haeger *et al.*, 1952, 1953), gonadotrophins and progesterone (Ahlmark and Swanberg, 1953) on histaminase in the tissues has been shown. Low histaminase activity in the serum during pre-eclampsia (Kapeller-Adler, 1951) and fall in the enzyme activity in the serum after treatment of pre-eclampsia observed in the present study may be explained as due to altered hormonal balance. It would be of interest to see if after termination of treatment the histaminase activity rises to initial (or higher) levels in subsequent weeks.

Summary

- (1) The present study was undertaken to study histaminase activity in the serum during normal pregnancy, and during pregnancy complicated with anaemia and/or pre-eclampsia. An attempt is also made to evaluate the influence of treatment of anaemia and/or eclampsia on the enzyme activity in the serum.
- (2) Sera of 3 non-pregnant healthy women and 5 women with gynaecological complaints (leucorrhoea, dysmenorrhoea and uterine prolapse) did not exhibit any enzyme activity when examined repeatedly in different phases of the menstrual cycle.
- (3) In women with normal pregnancy histaminase activity started appearing in the serum after 6 to 10 weeks of pregnancy. The advance of pregnancy was initially accompanied by a progressive rise in the enzyme activity in the serum. The rise occurred in two phases. A steady rise occurred upto 18 weeks; thereafter, a steeper rise occurred upto 30 weeks. From 31 weeks onwards there was a moderate fall in the enzyme activity.
- (4) Immediately after the third stage of labour the histaminase activity in the serum was similar to that observed during the last 4 weeks of pregnancy. The enzyme activity was reduced approximately to 35 per cent by 24 hours and to 10 per cent by 48 hours after delivery. The enzyme activity was negligible 80 hours after delivery.
- (5) In 3 of the 5 umbilical cord blood samples studied, the enzyme activity was low; in the remaining 2 no activity could be detected.
- (6) Histaminase activity in the serum of 21 cases of pregnancy complicated by anaemia was similar to the activity observed in subjects with normal pregnancy.
- (7) In 3 of the 5 patients of pregnancy complicated by pre-eclampsia the histaminase activity in the serum was in the normal range; in 1 patient the activity was high and in another it was low.
- (8) In all the 11 patients of pregnancy complicated by anaemia and pre-eclamptic toxæmia, the histaminase activity in the serum was in the normal range.
- (9) In 6 patients of pregnancy complicated by anaemia, the treatment of anaemia for 3 to 4 weeks led to considerable clinical and haematological improvement. In 2 patients the enzyme activity in the serum increased (more than 20 per cent of the initial level); in 4 patients the enzyme activity was within 20 per cent of the initial value.
- (10) In 8 patients of pregnancy complicated by anaemia and pre-eclampsia, treatment of anaemia and pre-eclampsia resulted in improvement of anaemia and pre-eclampsia. At the end of treatment, the enzyme activity in the serum was found to be reduced in 5 patients (30 to 50 per cent reduction) but unchanged in 3 patients.

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