DISCUSSIONS

.
The work embodied in the thesis entitled "Liver Function Tests, Thyroid Function Tests and Oral contraceptives", dealing with problem of contraception and oral contraceptives in women of reproductive age group are discussed below :-

Keeping in view the results of clinical considerations of females of reproductive age the author has come across complaints of joint paint, vague irregular body-ache, head-ache, insomnia, precordial pain, mild jaundice, phlebo thrombosis specially of the inferior extremity. The Bio-chemical parameters, as employed in this work are :-

I. LIVER FUNCTION TEST -
   a) Serum Bilirubin
   b) Serum Total Protein
   c) Serum Albumin globulin ratio
   d) Electrophoretic pattern of serum protein
   e) Thymol turbidity and flocculations of serum
   f) Serum Alkaline Phosphatase
   g) Serum glutamic oxaloacetic transaminase
   h) Serum glutamic pyruvic transaminase
   i) Serum Ceruloplasmin
   j) Basma Prothrombin Time
   k) B.S.P. excretion test.

II. BLOOD SUGAR
   a) Fasting
   b) Post Pandriel.
III. THYROID FUNCTION TESTS

a) Serum Protein Bound Iodine
b) Serum Butanol Extractable Iodine
c) Serum Cholesterol

IV. SERUM SODIUM

In order to study the effect of above mentioned biochemical parameters, 60 control subjects and 91 treated subjects are selected. The data collected on the two types of subjects are given in chapter of results and statistics. The decisions on the basis of data are based on the statistical criterion, student's t-test whose theory is outlined in the same chapter. The biochemical tests were done at the end of first month; 3rd month, 6th month, 1 year and 2 years in the same ladies, who could continue the pills. A few of them could not continue due to various complaints or they wanted pregnancy.

The first case discontinued at the end of 1st month due to evidence of clinical jaundice and she was medically advised to give up the pills, and to adopt other methods. Between 4-6 months, 40 cases discontinued, of them 16 wanted a pregnancy and another 24 gave up due to various complaints. 4 had nausea and vomiting in the early morning similar to the morning sickness of the pregnancy. 6 had insomnia and headache, 2 had thrombophlebitis of the deep veins of the legs, and 12 gave up due to various vagina complaints like vague body ache discomfort etc. Between 1-2 years, 20 cases discontinued the pills. Of them 5 ladies complained of over-weight which they
could not resist even with control of diet and exercise. 6 of them had continuous joint pain specially the last 15 days of the cycle, and 9 voluntarily discontinued for having a pregnancy. Of the tests for different biochemical parameters only bilirubin showed marked change at the end of 3rd month, which was statistically significant, all the other tests were almost the same upto 2 years. Even in rest of the times bilirubin also showed the same level.

It is seen that the joint pain occurs in almost 74.5% of cases. It is probably due to physiopathological disturbances in water metabolism as depicted by the electrolyte balance in these cases. When there is a rise of Serum Sodium Level 136.37 meq/L to 147.37 meq/L on an (vide Chapter of Results) average there will be rise in extracellular fluid which accumulates in the joint space initiating joint pain. Moreover estrogen induces some reversible change either on the capillary wall, the lymphatics or the extra-capillary tissues, thereby causing retention of extracellular water (Backman et al, 1936, Zuckerman, 1939). The swellings are associated with discharge of protein and mucoprotein (Aykroyd & Zuckerman, 1938, Ogston et al, 1939). Oestrogen tends to cause sodium and water retention (Shaw's Gynaecology 9th edition, 1971). Progesterone is also responsible for this sodium and water retention in the extracellular spaces (Shaw's 9th Edition, 1971).

The vague body ache is seen in almost all cases excepting a very few. This again may be due to the increase in the extracellular fluid due to elevation of sodium level.
In estrogen progesterone combined oral contraceptives or in only progesterone containing contraceptives have shown to produce elevations in plasma binding of aldosterone (Layna and Meyer, 1965). Aldosterone is the main hormonal factor in maintaining sodium metabolism. It acts on renal tubular epithelial cells to promote reabsorption of sodium ion. This is accompanied by reabsorption of chloride and water in the distal tubules. (Cantarow and Shepartz 4th Edition P 706, 1967), This may be the cause of increase of serum sodium level and water retention in the extracellular space which in turn may be responsible for vague body-ache. (W.H.O. Report series No. 326 - Geneva 1966). 12 ladies discontinued the pill in our series due to vague body ache and discomfort between 4-6 months.

Gain in weight and oedema of extremities specially inferior extremities has been noticed in about 45% of cases. For this complaint, 5 ladies gave up the pill at the end of 1st year of taking it. Goldzieher et al (1964) noted this weight gain in 14.3% of cases. W.H.O. Report No. 326 (1966) reported it in about 50% cases of women. In our test group 40 women out of 91 suffered from this complaint. None of our cases reported about weight loss. The change of weight gain is very complex and may occur due to improved appetite due to the relief from anxiety of unwanted conception. In control cases also there is evidence of oedema in luteal phase. It has been suggested by Cantarow and Shepartz that premenstrual oedema may be attributed in fact to increased secretion of aldosterone together with the increase in progesterone, rather than a
FIG. III Metabolism of Oestrogen and Progesterone

In the ovary with the help of FSH, LH and LTH, estradiol, estrone, and estradiol are synthesized.

- Absorbed from systemic circulation
- Estradiol glucosiduronide
- Estrone sulfate

Cholesterol

Pregnanediol

Pregnanalone

Pregna-3,20-dione

In adrenal cortex with the help of ACTH:
- Androsterone
- Androsterone
- 17α-hydroxyprogesterone

Excreted through kidney
direct action of the progesterone only. The naturally occurring oestrogen and the aldosterone which are the derivatives of the same source, are denatured in the liver. But as the liver is probably partially less functioning so there may be a rise in the aldosterone level which again disturb physiological homeostasis. Changes in the renin, angiotensin, aldosterone system is well known with oral contraceptives. The 2-3 fold increased in the renin substrate has been reported in the W.H.O. Technical Report No. 473, 1971. Normally in most of the women plasma renin level is low and with a normally functioning feedback mechanism angiotensin and aldosterone levels are normal. But in oral contraceptives as plasma renin levels are higher (W.H.O. Report No. 473, 1971) so it can be well assumed that the angiotensin and aldosterone levels are also raised. The aldosterone and angiotensin level in plasma is significantly raised (Beeckerhoff et al, 1973). The raised level of aldosterone is probably due to stimulation of the adrenal cortex by the renin angiotension system secreting more aldosterone activity causing retention of sodium and water, thereby causing accumulation of fluid in the tissue spaces. Also thrombophlebitis (Inman & Vessey, 1968; Doll & Inman, 1969; Vessey & Doll, 1969; Fegan & Henry, 1969) is very common in these cases, which causes sluggish venous return. Cardiac output depends on 'Vis-a-targo', so as there is sluggish venous return more accumulation of fluid in tissue space, so more oedema. Apart from the rise of serum sodium level produced by the progesterone probably the pars posterior of the pitutary gland which is labelled to secrete an hormone
known as antidiuretic hormone gets an upper hand to secrete more resulting in further accumulation of fluid in the tissue spaces.

About 33% cases complaint of insomina and headache particularly those who used the 'Pill' in the late evening suffered more. Headache in 3.2% cases was also reported by Goldzieher et al (1964). This may be due to alteration of homeostatic mechanism in the cerebral cortex which in turn is probably due to the alteration of hormonic balance. Question of involvement of limbic system can not be ruled out completely although we have no practical experience about that. In our series 60 ladies had to discontinue the pills due to these complaints between 4-6 months.

The precordial pain physiologically can never be due to high rise of estrogen. Estrogen on the other hand produces greater coronary circulation and hence used in the treatment of angina pectoris. But this pain may be due to the increase in B.M.R. which is again due to the progesterone in 'the pill'. Progesterone is believed to be responsible for increase of basal metabolic rate (Cantarrow & Scheparts). This B.M.R. increases the heart rate and enhances the cardiac demand which the poor coronary vessels fail to supply. Saruta et al (1970) has reported that there is a rise in blood pressure both systolic and diastolic and also rise in plasma, renin level in women taking oral contraceptive from a post partum clinic in U.S.A. Lim et al (1970) has shown an increase in systolic pressure after intra-veneous infusion of estrogen. Laragh et al (1967) Kuninet et al
Weir et al (1971) have said that estrogen is responsible for the increase of systolic blood pressure. Weir et al (1974) suggested that both systolic and diastolic pressures are significantly raised with oral contraceptives. So as the periphenal resistance is more the heart has to work more. As a result it needs more blood supply which the coronary vessels cannot manage leading to this pain. Moreover, according to report of W.H.O. scientific group No. 326 Geneva (1966) oral contraceptives produces changes in the blood clotting mechanism. There is a stage of hyper coagulability which is due to changes in the levels of clotting factors, in platelet functions and the physical state of blood vessels. Clotting may be initiated by platelet adhesion to damage endothelium with formation of fibrin thrombus. (W.H.O. No. 473, 1971). There may be intravascular thrombosis in the coronary arteries also in patients taking oral contraceptives which might have partially blocked the vessels leading to less supply of blood to heart muscle causing this pain.

Next comes the thrombophlebitis which has been seen to occur in a good number of cases. Fegan and Henry (1969) found incidence of thrombophlebitis was 5 times greater in oral contraceptive users. Between 4-6 months of using the pill 2 ladies had to give up the pills due to thrombophlebitis of the deep veins of the legs. The complex system of blood clotting and fibrinolysis are in equilibrium and these are complementary in maintaining the integrity and patency of the vascular system (Stafford, 1964).
FIG. IV MECHANISM OF BLOOD COAGULATION

FIBRINOGEN

VITAMIN-K

PROTHROMBIN

Thromboplastin

Ca++

Ac-globulin

Proconvertin

Thrombin

FIBRIN

Platelets

Christmas factor

AHF

Ca++

Ac-globulin

Thromboplastin
Estrogen alone or combined estrogen progesterone contraceptives increase plasma antiplasmin and decrease serum antithrombin activity. The fall in antithrombin activity during treatment with oral contraceptives was noted by Vonkaulla and Vonkaulla (1970) and Paterson et al (1970). Howie et al (1970) suggested that estrogen component is responsible for the fall in antithrombin activity of serum. Moreover, Miller et al (1965), Wemstaker and Mink (1969), Philips and Turksoy (1961) reported a rise in fibrinogen in oestrogen and progesterone therapy. Egeberg and Owren (1963) found a lowering of prothrombin time and partial thromboplastin time (Poller et al 1963) and an increase in fibrinogen (Nilsson and Kullander, 1967) and Platelet adhesiveness (Hidden et al, 1967) was also found. Possibly the changes in the levels inhibitors of coagulation and fibrinolysis produced by oral contraceptives may contribute to the increase tendency to thrombosis (Imman and Vassey, 1963). It has also been shown that alpha-2 macroglobulin diminish the fibrinolytic activity of plasma in vitro (Williams R.H.- Text Book of Endocrinology p.475, 1968 and Ganret P.O., 1967). It is now assumed that the complex system of blood clotting and fibrinolytic activity are in equilibrium and these are in fact complementary in the maintenance of the integrity and the patency of the vascular system (Strafford, 1964) so an increase in the anti plasmin activity may alter the pre-existing equilibrium unless this tendency is neutralized by a corresponding change of some other factor affecting clotting or affecting fibrinolysis. The raised serum level of
alpha-2-macroglobulins which is found in these cases could thus conceivably contribute to the tendency of thrombosis (Horne et al 1970). Moreover estrogen and combined estrogen progesterone oral contraceptives increases plasma antiplasmin and decreases serum antithrombin activity. The fall in serum antithrombin activity during treatment with combined estrogen progesterone oral contraceptives accords with the findings of Vonkaulla and Vonkaulla (1970) and Paterson et al (1970). Observations of Howie et al (1970) also strongly suggests that estrogen is responsible for this fall of antithrombin activity of serum. The fall in antithrombin activity during combined estrogen progesterone thereby may explain the venous thrombosis (Howie et al 1970). Philips et al (1961); Miller et al (1965); Ambrus et al (1969) reported a rise in fibrinogen in estrogen progesterone therapy. So the venous thrombosis in patients taking oral contraceptives may possible be explained by the changes in the level of inhibitors of coagulation and the fibrinolysis. This view is also supported by Inman and Vessey (1968). Moreover, alteration in blood circulation, the state of the vessel wall and the platelet adhesions may also contribute a major role in intravascular thrombosis (W.H.O. Technical Report series 326, 1956).

Another condition which is complaint by many women is jaundice. The blood level of Bilirubin is elevated in most of the cases, during the initial phase. This is a very mild type of jaundice. This occurrence of cholestatic jaundice
may be expected in users of the pill since its ingredients 17-alkyl substituted steroids are known to produce cholestasis. (Sherlock, 1963). Following the administration large doses of oral contraceptives to rats and a few human beings, an alteration in liver structures are seen under the light and electron microscope. The changes found are cholestasis altered cell membrane staining reactions, changes in lysosomes and dilation of canaliculi with blunting of microvilli (World Health Organisation Technical Report Series No. 336, 1966) manifest jaundice of cholestatic type often occurring within first month of using the pill has been reported by many workers (Adlercum et. al and Ikona, 1964; Boake, Ockner and Davidon, 1967; Schmid, 1969; Ylostalo, 1970).

Serum bilirubin. The average increase of serum bilirubin is from .32 mgm in control cases to 1.39 mgm per 100 ml. of serum in treated cases which is not showing evidence of clinical jaundice. But in all the test cases the serum bilirubin level is raised. Out of 91 test cases, in 90 cases the serum bilirubin value is from 0.9 mgm to 1.9 mgm/100 ml of serum, that is the rise of serum bilirubin value is above the normal level and there is latent jaundice in these cases. The rest one case out of these 91 cases, there is evidence of clinical jaundice, the bilirubin level is 2.1 mgm/100 ml of serum. This clinical jaundice appeared in the 3rd month of the course of the medicine and disappeared after the stoppage of the pill. Hyde and Draisay (1974) noted clinical jaundice after introduction of the oral contraceptives in 1 in 10,000 women. Physiologically it is known that
The accumulation of estrogen from the external sources probably hampers the liver function in the first round as the liver cannot cope up with the incoming of excess estrogen. But later on this cholestatic stage probably passes away and so there is disappearance of jaundice and lowering of bilirubin level is seen. The incidence of hyper-bilirubinaemia in this series is in almost all the cases. This rise lasted for 3 months with gradual lowering of serum bilirubin level.

The serum bilirubin estimation done at the end of 3rd month. By the 2nd week of the 4th cycle of pill taking showed serum bilirubin level at a high normal value, mean being 0.8 mgm/100 ml of serum. Although statistically it is significant. This increase in the serum bilirubin level may be due to alkylated group in the C\textsubscript{17} position of steroid configuration and which is causing cholestasis (Sherlock, 1963). There may be a synergic toxicity when both estrogen and progesterone are together (Borglim 1965, Kislau et al, 1965). The absence of hyper bilirubinaemia during estrogen administration does not mean that capacity of liver to tackle pigment remain intact (Kottra & Kappas, 1967). All the other liver functions test were same in the 4th cycle reading, so we are also agreeing with the opinion of Kottra and Kappas.

There is a decrease of serum protein value from 6.6 gm/100 ml in control cases to 5.1 gm/100 ml of serum in test cases. The albumin value is also decreased from
4.3 gm to 2.6 gm/100 ml. There is an alteration in albumin globulin ratio and rise of globulin from 2.30 gm to 2.6 gm. Although according to Stole (1966) there is no alteration of plasma protein but we have found a definite decrease of plasma protein which is statistically significant.

Electrophoretically we have found significant change, alpha 1, 2 globulin and 3 globulins fractions are definitely increased. Beckman et al (1973) has found an increase in the 2 serum globulin in pregnancy and in cases of users of oral contraceptives. Liappis (1973) has found a significant fall in serum albumin and a rise of 3 globulin which corroborates with our findings but he has also found a raised 1 globulin which is not true in our cases, rather in our series we have found a lowering of 1 globulin which in control cases are in average 1.27 gm/100 ml to 0.78/100 ml in test cases. That is there is a lowering of level of 1 globulin which is statistically quite significant.

Firek, A., et al (1973) found after administering an oral contraceptive femigen for 6 months to some ladies that total globulin 1 and 2 globulins were increased and 1 globulins were decreased which also corroborates our findings but he did not found any change in total protein, albumin and 3 globulin, which is not true in our case.

It is well known that liver is the chief site of formation of albumin (Cantarrow & Schepartz p.561). We know almost all the plasma proteins are originated in the liver excepting 1 globulin which originate from plasma cell and
lymphoid tissue (Herper, 1971). In chronic liver diseases like cirrhosis of liver there is lowering of plasma protein and albumin (Herper, 1971). In case of oral contraceptives we know that there is an impairment of liver function. As we know that hydroxylation of steroids takes place in the liver. Both the estrogen and progesterone used in the pill might be altering microhistology of the liver (W.H.O. Report No. 366, 1966), 473 (1971). So this damage in the liver cells may be the cause of lowering of this total protein and albumin γ globulins. It is known that in synthesis of plasma proteins, any case of deprivation on the liver will have its most marked effect on plasma albumin and γ globulin. So due to the liver damage there is alteration in the protein pattern.

There is a marked increase in thymol turbidity and flocculation tests. In control subject mean is 1.40 units in test subjects 5.50 units. It is well known that thymol turbidity depends upon partial precipitation of certain globulins specially Beta and Gamma globulins in the plasma (Wooton 5th Edition, 1974; P.166), Maclagon & Bunn, 1947. Kleslie oldershaw (1975) observed the increase of beta globulin synthesis after administration of oral contraceptives. We have also observed increase of beta globulin fraction in the electrophoretic pattern of the proteins. The precipitation reaction of thymol turbidity test is markedly enhanced by lowered level of albumin (Wooton, 1974), the level of albumin in our test cases also is lowered considerably which has as a result given a much higher value of thymol turbidity. This increase of the thymol turbidity
value has also been observed, in oral contraceptive users, by the Medical School Mozambique in 1970.

The serum alkaline phosphatase level is higher than in control groups in our test subjects. Zuckerman (1955) and Talay in 1957 observed an increase of alkaline phosphatase level in the endometrium after administration of estrogen. The rise of serum alkaline phosphatase level after administration of estrogen has also been seen by Eisalo et al (1964), Palva and Mustaba (1964), Eisalo et al (1965), Borglin (1965), and Larsson and Cohn (1965), Kottra & Kappas 1967, Jain et al (1973). Although it does not tally with findings of Swaab (1964), Riceway (1964), and Tylor (1964). The probable explanation of this increase may be either retention of the enzyme in the blood which is originally excreted in the bile and which is originating from the intestine or the enzymes accumulations in the blood originating from the liver but due to impaired biliary secretion is stagnant in the blood (Cantarrow and Schepartz, 1967). We know that the estrogen fraction of oral contraceptives causes hepatocellular damage like alteration in cell membrane, staining reactions, changes in lysosomes, dilation of bile canaliculi and blunting of microvilli (No. 326, W.H.O. 1966). So this hepatocellular damage is probably the main cause of this increase in this enzyme level, which is probably due to the estrogen fraction of the oral contraceptives.

Serum glutamic oxaloacetic transaminase and serum glutamic pyruvic transaminase values have been raised
considerably. On an average the S.G.O.T. from 14.55 to 43.29 units and S.G.P.T. from 15.67 to 43.22 units. Benjamin Eckstein & Miriam Lev (1964) found considerable increase of S.G.P.T. activity in uterus, ovary and liver, by sex hormone stimulation. This elevation of these enzymes in serum after in take of oral contraceptives were also noted by Eisalo et al (1964), Palva and Mustaba (1964), Eisalo et al (1965) Borglin (1965), Linthorst (1964), Rice-wway (1964), Seyer and Little (1965) and Baake (1965); Scheels' et al (1972) found no such changes with oral contraceptives and Hague & Cade in both pregnancy and oral contraceptives. But the raised value which we have found in the patients taking oral contraceptives is statistically quite significant. We know that liver cells contain a very rich quantity of transamnase enzymes. It is also seen in W.H.O. Report No. 326 (1966), 366 (1968) and 471 (1971) that there is hepatocellular damage in case of these oral contraceptive users. The estrogen present in these pills causes this damage causing raised enzymd levels (Eisalo et al 1964). But we know that progesterone is also denatured in the liver and so it may also cause damage of the hepatic cells. According to Stoll (1966) progesterone is more responsible for the damage than estrogen. But the damage may be the synergistic effect (Eisalo et al, 1965; Borglin, 1965) of both estrogen and progesterone. Moreover we know transaminases are excreted through the bile and bile only. No transaminases is present in urine. (Rudolph Abderhalden Clinical Enzymology, 1961; P.36). With oral contraceptives there is
impaired biliary secretion of the liver (Eisalo et al. 1964; Palva and Mustaba 1964). This may be another reason of increase in the level of these enzymes in the serum.

The enzyme ceruloplasmin is a copper binding protein of the serum in plasma. It has been found to be increased (from 27.70 mg/100 ml in control subjects to 48.90 mgm/100 ml of serum in treated cases) in this work. The same elevated level of serum and plasma ceruloplasmin after taking combined oral contraceptive was observed by Carruthers et al. (1966), Derrick Torey and Lathe (1968) Clemston (1968), Schenkee et al. (1971), Dautten and Elstein (1973), Olaturoboso et al. (1973).

The serum ceruloplasmin level also rises in cases of pregnancy (O'Reilly and Loucin 1967). Increase of serum ceruloplasmin level with administration of only estrogen was observed by Turfin et al. (1956), Voustudziu and Berezin (1958). It is known that although copper is widely distributed in many tissues of the body but the main site of storage of copper is in the liver (Cantarrow and Schepartz).

Oral contraceptives containing estrogen and progestrone or only estrogen is causing the hepatocellular damage which is no longer capable to store the copper. There is also a rise of the alpha 2 fraction (Beckman et al. 1973; Firek et al 1973). So it may happen that this copper gets attached with the α2 globulin causing a rise of the serum ceruloplasmin level.

There is a marked reduction in prothrombin time in the test cases in this series. Prothrombin is a globulin circulating in the plasma. Prothrombin is measured in a system
which relates the amount of time required for clotting of plasma to the prothrombin levels of the plasma. A reduction in prothrombin time has also been noted by Egeberg and Owren (1963). W.H.O. No.473 (1971). In users of oral contraceptives there is slight hepatocellular damage due to estrogen. We know prothrombin is synthesized in the liver. But acquired hypoprothrombinemia will result only then when the liver damage is very severe to cause interference with the prothrombin synthesis (Harper, 13th Edition P.190). Here in these cases liver damage is not so. According to W.H.O. Report No. 473, 1971, this decrease in prothrombin time is due to progesterone part of oral contraceptives.

There is definite rise of fasting blood sugar level (from 70.5 mgm in control group to 110.00 mg/100 ml. of whole blood in treated group. The post pandrial blood sugar from control 66.7 mgm/100 ml to 109.00 mgm/100 ml of whole blood. The rise in sugar level is quite significant statistically. This is also seen by Greshberg et al (1968), Wynn and Doar (1969), Goeretzleahner (1973), Angeli and Vertes (1973), Spellacy (1973). This abnormality of carbohydrate metabolism may be due to increased resistance of one or more peripheral tissues to the action of insulin. Wynn and Doar (1966) has shown that there is a similarity between carbohydrate metabolism in subjects receiving oral contraceptives and subjects undergoing cortisosteroid therapy, (Wynn, V. & Doar, J.W.H.-Conference on metabolic effects of Gonadal Hormone & Contraceptive Steroids - Edited by H.A. Salhanick, DM Kapnia, and R.L. Vande Weile). Oral contraceptives have
shown to produce the elevations of plasma binding Cortisol (Metcalf and Beaven, 1963; Lloyd & Weisz, 1966). It is suggested that elevated protein bound plasma cortisone levels may produce metabolic changes in certain organs with protein permeable vascular beds such as the liver (Kellar et al, 1969). It has also been shown non-protein bound plasma hydrocortisone levels are increased during estrogen therapy. The major action of cortisol is to produce neoglucogenesis in the liver and thus it produces a rise in blood sugar level. The adverse effect of oral contraceptives on glucose level is probably due to the estrogen fraction and it is secondary to raised plasma hydrocortisone level (Doar and Wynn, 1969).

Bromosulphthalein test - It is the test to know the ability of liver to excrete the dye within a known time. Here the B.S.P. excretion rate has been much reduced in test cases than in control cases. In control cases it is 3.8% in average after 45 minutes when as in test subjects it is 22.5% after 45 minutes of injection of the dye. It was also found high by Eisalo et al (1964), Kottra and Kappas (1967), Allan and Tyler (1967) also found raised level of this dye after intake of oral contraceptives. This test is taken to be most sensitive of all liver function tests (Varley 4th Edition, 1975). This test is the measure of hepatic ability to concentrate the dye and to excrete it in the bile. In oral contraceptive users it has been reported by many authors e.g. Eisalo et al (1964), (1965), Palva and Mustala (1964), Brain et al (1964), Stoll et al (1966), Harben and Gade (1973), Kottra and Kappas (1967),
that there is impairment of hepatic function. In W.H.O. report No. 3 (1966) and 473 (1971) it is reported that in electron microscopy there is change in hepatic cell structure and dilatation of canaliculi, and these changes are mainly due to the estrogen part of the oral contraceptives. Now in these oral contraceptives users due to partial damage of the liver cells there is delay in concentration of the dyes and due to dilatation of the canaliculi (W.H.O. 473, 1971) may be delayed in excretion also.

It is known that pregnalone is the pre-cursor of progesterone and cortisol in the adrenal cortex. As the progesterone is substituted in the pill so it may happen that more formation of cortisol takes place resulting in increasing the output of the glucose by the liver and diminishing peripheral uptake and utilization of glucose and increasing meoglucogenesis may be contributing factors to this abnormality of glucose metabolism.

There is an increase of serum sodium level from 134.50 meq/L in control subjects to 147.37 meq/L in treated cases. There is a rise in plasma aldosterone in the users of the oral contraceptives, Layne & Meyer, (1965), Lloyd & Weisz (1967), (W.H.O. 473, 1971). This rise is due to raised level of plasma renin which causes increase of angiotensin and aldosterone level. Again progesterone and aldosterone are derived from the same sources, so when the progesterone is substituted from outside then the excess one may be transformed into aldosterone in the body. This aldosterone causes reabsorption of sodium by the tubular epithelium of the kidneys, causing a retention of sodium, and as a result there is an increase
of sodium in the extra cellular space.

In this present work there is elevation (from the protein bound iodine from 6.066 mgm/100 ml in control group to 6.154 mgm/100 ml in treated group) and Butanol Extractable Iodine (from 5.300 mgm/100 ml in control group to 5.280 mgm/100 ml in treated group) are found. The rise in P.B.I. and B.E.I. is probably due to the changed metabolic pathway and ‘Feed-back mechanism’. Oral contraceptives are mostly combination of estrogen and progesterone, and their mode of action is via 'hypothalmo - hypophyseal system'. Estrogen component acts on the anterior pituitary through the hypothalmo-hypophyseal system reducing the Follicular stimulating Hormone of anterior pituitory (Diczfalusy, E. 1968). Estrogen component is possibly responsible for increase of P.B.I. and B.E.I. (The effect may be through the effect on pituitary function which are probably mediated by the hypothalamas). Goolden et al (1970) has also shown that increase in P.B.I. is due to estrogen component and according to him this increase is secondary to thyroxine binding globulin. Moreover estrogen increases the concentration of Thyroxine binding globulin (Hollander et al, 1963; Lloyd et al 1966). The administration of small doses of estrogen leads to an acute decrease in serum Thyroid stimulating hormone concentration. From which an escape occurs by the second or third day of continued administration of the drugs. Estrogen increases concentration of thyroxine binding globulin, elevates serum T₃, T₄ concentration.
The effect of estrogens exerted through an inhibition of endogenous Thyroxine releasing hormone release (Gross and Apploman, 1971). Serum T₃ concentration is increased in pregnancy and in oral contraceptive users. However, increase of T₃ is less marked than that of T₄ (Nicoloff and Low 1972).

The amino acid tyrosine is not only the precursor of thyroxine but is also the precursor of melanin and epinephrine and norepinephrine. So it may happen that tyrosine, in its course of metabolism, due to allosteric inhibition in the quinone system passes down to thyroxine. The rise in the value of P.B.I. and B.E.I. may be also due to this.

Value of cholesterol is higher in test groups than in control groups. This finding is corroborating with the findings of Doen et al (1968), W.H.O. Report No. 473 (1971), Older's shaw (1975). Cholesterol in the precursor of the steroids, and basically all the oral contraceptives are steroids. The conversion of cholesterol to estrone by human ovarian slices (Ryan and Smith 1961) and to progesterone by ovine ovary (Tamaoka and Pincus, 1961) had been shown. It can be expected that a part of cholesterol both exogenous and endogenous are converted into these two hormones. But in the cases of oral contraceptive users as these hormones are supplied from outside, so it may be possible that the cholesterol synthesized in the body is not metabolized into these steroids, causing a rise of serum cholesterol level.