Review of Literature
REVIEW OF LITERATURE

Menopause defined as the permanent cessation of menses that occurs after the cessation of ovarian function, is one event rather than a period of time. It is a natural event – a part of the normal process of aging.

In 1842, Brerre de Beusmart in “Al La Menstruation” discussed many aspects of menstruation, He was the 1st to discuss about menopause and according to him, it is the termination of reproductive phase of life in women.

Menopause is merely one milestone in this highly complex life cycle of women. It can be spontaneous or surgical and marks a new stage in women’s life (Speroff et al 1994). In cases of surgical menopause, young age of these women compared with those undergoing natural menopause and the abrupt onset of associated symptoms create special problems (Wilcox et al 1988).

As per the WHO technical report series ‘670’, the term menopause is defined as the permanent cessation of menstruation resulting from loss of ovarian follicular activity. Women are considered to be menopausal if menses have ceased because of removal of both ovaries along with uterus or for women with intact uterus, menses have ceased at least for a period of one year period after the age of 40 years (Rocenberg et al 1993).
Since 1960’s, a considerable data has been accumulated regarding human climacteric. Climacteric is that phase of aging process of women which marks the transition from the reproductive stage of life to non reproductive stage. It can be regarded as a specific pathologic process or as an endocrinopathy (Utian, 1980).

**Age of Menopause**

In developed countries median Age of menopause is 50 year (Brambrilla DJ et al, 1989; Mc kinlay Sm et al, 1992). Some studies done in India have reported the variable data regarding the age of menopause from 42.55 years to 48.62 years (Wyan et al 1966, Sharma et al 1980, Randhawa et al 1987).

The age of menopause appears to be genetically determined. It occurs earlier in cigarette smokers, in nulliparous women, and those undergoing hysterectomies (Siddle N et al 1987, Brambilla DJ et al 1989).

**Physiology of Menopause**

Menopause is a hormone deficient state where ovarian activity ceases. No corpus luteum is formed, no progesterone secreted by ovary and there is no menstrual cycle. All ovarian activity also diminishes and atrophic ovaries lead to menopause.
Due to cessation of follicular development, there is a drop in estradiol level causing loss of negative feed back on hypothalamic pituitary centre which, in turn, is responsible for the increase in FSH and LH by anterior pituitary (Yen, 1979). With further advancing years, gonadotrophin activity of anterior pituitary ceases. Hence, there occurs a fall in FSH & LH (Thaws, 1995) Thus, menopause is a hypoestrogenic state, estrogen being secreted in very small amounts, the source of which is the adrenal cortex, (Langcope, 1971).

**Symptoms & Physical Changes**

This estrogen deficient state may provoke a variety of metabolic, endocrine, biochemical, structural and symptomatic alterations in postmenopausal women which may be termed as post menopausal syndrome.

According to Lengaten & Kraines (1966), symptoms of menopause may begin in perimenopausal period, maximum of complaints, usually 2-3 years after menopause and then slowly decrease. There may be short term and long term complications. While short term complications should be treated, long term complications must be prevented.

**Early Symptoms**

Include symptoms of Vasomotor instability i.e. Hot flushes, Night sweats, Vertigo, Palpitations, weakness etc.
**Intermediate Symptoms**

*a) Urogenital atrophy* :- Such as atrophy of vagina, bladder and uterus etc. Symptoms produced are vaginal dryness, pruritus, discharge, dyspareunia and urethral syndrome such as stress incontinence, burning, frequency, urgency of micturition etc.

*b) Psychosomatic Changes* :- Anxiety, irritability, depressive symptoms, insomnia, sexual changes, diminished libido.

**Late Symptoms**

a) Osteoporosis, presenting as bone pains and spontaneous fracture.

b) Cardiovascular complications.

**Early Symptoms**

*Hot flushes* - It is the classic symptom associated with estrogen deficiency. It is described as “recurrent, transient periods of flushing, sweating, and a sensation of heat, often accompanied by palpitations, feeling of anxiety and sometimes followed by chills” (Kroenberg F, 1990). It is felt most commonly on the face, the arms and on upper part of body. It may associated with insomnia or sleep disturbances. It is often the earliest and commonest symptom presented in 60-80% of perimenopausal and post menopausal females. The entire episode lasts usually no more than 1-3 minutes, and may
recur as many as 30 times a day, although 5-10 times per day is more common. During the episode, temperature may rise by 5°C. These flushes are accompanied by vasodilatation & elevation of lutenizing hormones. They are experienced more often after surgical menopause. (Weinstein L 1990 ; Kronenberg F 1990)

Physiologically, they correspond to marked, episodic increase in the frequency and intensity of GnRH pulse from the hypothalamus. Instead, the increased pulsatile activity is a marker for some central disturbance of the body temperature regulation center which is responsible for the hot flushes (Ravnikar V. 1990) Symptoms of vascular instability subside by itself within 3-5 years (Kronenberg F, 1990).

Intermediate Symptoms

Urogenital Atrophy

Atrophic Changes occur with greater severity in tissues with preponderance of estrogen receptors i.e. vaginal tissue, uterus, distal urethra & trigone of bladder. Recent studies have found hormone receptors in females in pelvic musculature like levator ani and ligaments like round ligament (Smith et al 1990). Within 4-5 years, approximately one-third of women develop symptomatic atrophy. (Notelevitz M, 1978).

Decreased estrogen levels result in thinning of vaginal epithelial cells, they remain immature, glycogen content [9]

According to Houser et al, 1981, Vulval atrophy is sometimes accompanied by pruritis and burning is there.

Urinary symptoms may include dysuria, urgency, frequency, recurrent urinary tract infections, nocturia etc. (Notelevitz M, 1989). In addition, genuine stress incontinence may be related to estrogen deficiency. As estrogen receptors are found in distal urethra, post menopausal changes lead to atrophy of mucosa, decreased vascularity, diminished tone of urethral musculature which result in recurrent attacks of UTI and stress incontinence (Reckess et al, 1992). Estrogen therapy may improve or cure stress incontinence in more than 50% of treated women (Bhatia NN et al 1989).

**Psychological Symptoms** –

Psychological Symptoms such as anxiety, irritability, depressive symptoms and insomnia are most common, just before the onset of menopause.

Falling estrogen levels are directly related to mood changes and psychosomatic symptoms. Vasomotor symptoms often lead to sleep deprivation, chronic fatigue and hence related to psychological symptoms such as depressive symptoms, irritability and mood changes. Women often
experience difficulty in concentrating and loss of short term memory which may be attributed to aging alone or may be due to subtle sleep deprivation associated with hot flushes and replacement therapy with estrogen has been shown to improve both short term memory and psychological function in post menopausal women (Dennerstein et al, 1979; Ditkoff EG et al, 1991).

**LONG TERM COMPLICATIONS**

**Cardiovascular Disorder**

Menopause increases the risk of coronary artery disease due to adverse changes in serum lipids and lipoprotein levels and declining estrogen level. The risk of coronary artery disease is at least three times as great for men as for women before menopause and the relative risk for women increases significantly after menopause.

*Lobo RA 1990* has shown that estrogen deficiency significantly increases the risk of cardiovascular disease and this can be reduced by estrogen replacement therapy.

**Osteoporosis**

By definition, osteoporosis is the reduction in quantity of bone. The etiology of osteoporosis is multifactorial. Main factors are age, heredity, estrogen status. Dietary calcium is the most important factor associated with bone loss. Before menopause, the rate of loss is less than 1% of total bone
tissue per year. After menopause, rate of bone loss increases to as high as 5% per year in estrogen deficient women \cite{Riggs BL, 1987} \cite{Peck WA (1990)} Sudden decrease in estrogen levels as seen after oopherectomy or with the onset of amenorrhoea is associated with dramatic changes in remodeling of bone resulting in decrease in trabecular bone lead to an increased predisposition for spontaneous fracture. Symptoms produced are body pain, backache, decrease of height, kyphosis, wrist fracture after minor trauma and spontaneous fracture of long bones.

\textbf{Other Symptoms}

\textit{Skin Changes} – Estrogen receptors are also found in skin & collagen tissue. After menopause, due to decrease in estrogen, skin becomes dry and wrinkled. Due to increased production of melanin, complexion becomes dark.

\textbf{Hormonal Changes following Menopause}

In human ovary, there is continuous and progressive decline in the number of follicles. This loss can not be accounted for by ovulation alone. All types of follicles small, medium, large show decline in number with age continuously through a process of atresia also.

Progressive decline in number of ovarian follicles is responsible for decreased production of ovarian Inhibin \cite{Mc Lachlan et al 1988}. Ovarian Inhibin is a non steroidal,
water soluble protein secreted by granulosa cells of graffian follicle under the influence of estrogen. It suppresses the pituitary follicle stimulating hormones by negative feed back mechanism and ultimately FSH decreases in blood.

In menstruating women, FSH, on day 3 of cycle should be 5-10 IU/L with normally functioning ovaries Elevated FSH levels >40 IU/L are consistent with complete cessation of ovarian functions.

Prior to menopause, LH levels are in the range of 5-10 IU/L. LH levels increase in the menopausal transition in a manner similar to FSH.

First detectable endocrine manifestation is a gradual increase in plasma follicle stimulating hormone. Sometimes, after the rise of FSH, estradiol level decrease slightly and serum LH increases. Eventually as estradiol secretion falls to very low level, both FSH & LH rise in post menopausal period and remain elevated.

**Hill (1967)** showed that physiological symptoms in artificial menopause are not more serious than in normal menopause if the women has already reached the age of this change.

Hot flushes are apparently the result of instability between hypothalamus and autonomic nervous system brought about by a decline in estrogen. They are particularly
disturbing at night, perhaps because the hypothalamus is in relative state of rest.

Hot flushes are the pathognomic symptom of menopause and the one that most frequently prompts the post menopausal women to seek medical care (Maschak et al 1985; Erlik et al 1981).

Young et al (1957) observed that untreated menopausal women may experience dysuria due to senile urethritis caused as a result of atrophy of bladder and urethral mucosa. Rud et al (1980) concluded that the subsequent alteration in urethral and bladder pressure can cause urgency, stress incontinence and frequency. A variety of complaints such as headache, insomnia, myalgia, and change in libido may also be noted. The last but not the least are the health problems secondary to deprivation of estrogen which include the osteoporosis and cardiovascular disease.

Riggs et al (1969) observed that declining level of estrogen after menopause leads to an increased rate of bone resorption and greater urinary excretion of calcium. Women remain asymptomatic for 10 years after menopause but lost 15% of their bone moss, setting the stage for fractures.

The precise understanding of the symptom complex which the patient display is often difficult to achieve. Some patients will experience severe multiple reactions which are disabling while others show no reaction or minimal reactions.
which go unnoticed until careful medical evaluation is done. Symptoms typically appear when plasma estradiol concentration falls below 35pgm/ml and 40% of women develop menopausal symptoms serious enough to seek medical assistance (Carr & Wilson, 1987).

Campbell & Whitehead (1977) have concluded from both long term and short term trials that many symptomatic improvement ascribed to estrogen therapy results from relief of hot flushes, improvement in memory and reduction in anxiety. Schiff et al (1979) showed that estrogen therapy improves the quality of sleep, decrease the time of onset of sleep and increases the rapid eye movement and sleeptime.


Semmens et al (1982) have demonstrated that vaginal factors which influence the enjoyment of sexual intercourse can be maintained by appropriate dose of estrogen.

Brinent et al (1985) observed that the collagen content in the dermis of women on exogenous estrogen therapy was maintained at the premenopausal levels. Also estrogen replacement therapy not only prevents loss of bone mass but also reduces the risk of fractures (Ettiniger et al, 1985 and weiss et al, 1980).
Risk factors for Cardiovascular Disease

Some risk factors for cardiovascular disease include elevated blood pressure, impaired blood glucose tolerance, abnormal blood lipid levels, cigarette smoking, male sex and advanced age. Elevated blood pressure and alteration in lipid and lipoprotein metabolism are major contributors to cardiovascular disease in general and coronary heart disease in particular (Gordon et al 1971, Kannel et al 1986, Show et al 1987).

The gap in prevalence of coronary heart disease between both sexes decrease with advancing age and is not attributable to difference in lifestyle. It seems likely that some feature of reproduction physiology are responsible for this changes.

Gordon et al 1978 in studies at Framingham indicate a prompt loss of resistance to coronary heart disease in women after attaining menopause, as compared to women of the same age who remain premenopausal. Menopause alone, more than double the risk of coronary heart disease.

Colditz et al (1987), Lobo (1990) have shown that incidence of coronary artery disease is significantly increased, specially in those with premature menopause caused by bilateral oopherectomy or premature ovarian failure.

It has been shown that post menopausal women have higher plasma levels of cholesterol, triglycerides, very low density lipoprotein as compared to premenopausal females
(Campos et al, 1988 and Halberg et al 1987). Lower levels of high density lipoproteins were found in post menopausal period (Methews, et al 1989).

The term “odd couple” has been given to LDL and HDL cholesterol which together are responsible for 90% of cholesterol in plasma. The concentration of LDL cholesterol is directly related to coronary heart disease and its elevation has been shown as a predictive risk factor (Gordon et al 1981) and Wilson et al, 1980.

The majority of studies state that HDL cholesterol is a stronger predictor of risk of coronary artery disease (Gordon et al, 1977) as compared to LDL cholesterol (Segurdson et al, 1975 ; Janus et al , 1980 ; Talani et al, 1981 ; Thompson et al 1981 ; Rao et al , 1983).

**LIPIDS AND LIPOPROTEINS**

Increasing awareness of the relationship between plasma lipoproteins and risk factors for cardiovascular disease has lead to a renewed interest in the effect of hormone replacement therapy on cardiovascular risk. It requires an understanding of underlying mechanisms involved in the effect of replacement of hormones on serum lipids and lipoprotein.

Once the dietary fat is absorbed , the lipids are synthesized by liver and adipose tissue. The lipids are insoluble in plasma and circulate in blood stream as
macromolecular complexes. The surface layer of lipid particle consist of specific proteins called apoproteins as well as phospholipid and partially polar unesterified cholesterol. The lipid core contains nonpolar triglycerides and esterified triglycerides and cholesterol. The hydrated density of the lipoprotein particles varies with the relative ratio of its different components.

The lipoproteins can be separated by ultra centrifugation. The following system is used for classification of lipoprotein:

Very low density lipoprotein (VLDL)  .96 to 1.006
Low density lipoprotein (LDL) 1.006 to 1.063
High density lipoprotein (HDL) 1.063 to 1.21

\[
\text{HDL fraction} \quad \text{HDL2} \quad \text{HDL3} \quad \text{(Major sub fraction)}
\]

Very low density lipoproteins are secreted in the liver and the endogenously synthesized triglycerides are transported into LDL. The cholesterol is transported from the liver to the peripheral cells via LDL particles. The cellular uptake of cholesterol is regulated by high affinity low density lipoprotein receptor (Brown et al, 1981) which might be influenced by estrogen. Estrogen has been found to increase the number of hepatic LDL receptors in the animal studies. This increase enhances low density lipoprotein degradation. In addition, low density lipoprotein can be broken down in scavenger cells or
macrophages of reticuloendothelial system if LDL concentration is high *Brown et al 1981*. These cells can be overloaded with cholesterol and converted to foam cells.

Estrogen appears to protect against the development of cardiovascular diseases by a number of mechanisms like by beneficial changes in cholesterol levels by decreasing low density lipoprotein (LDL) cholesterol and increasing high density lipoprotein (HDL) cholesterol levels by induction of LDL receptors and destruction of hepatic lipases which degrades HDL cholesterol levels *(Lobo RA, 1990)*. Direct effect are prevention of cholesterol deposition in vascular lesion, prevention of vasoconstrictive action of acetylcholine on artherosclerotic vessels, interference with thromboxane effect on blood vessels, increased production of prostacyclin from blood vessels, stimulation of myocardium causing a positive inotropic effect, increasing cardiac index and heart rate while decreasing systemic blood pressure and systemic vascular resistance *(Schwartz et al, 1995)*.

1. **Serum Cholesterol**

*Oliver and Boyd (1959)* showed that there was a significant rise in serum cholesterol by oopherectomy *Sznajderman and Oliver (1963)* showed a significant rise in serum cholesterol in women with premature menopause compared with premenopausal women of same age group.
Arnold and Ritterband et al (1963) showed that the levels of serum cholesterol was significantly higher in oopherectomized than those of hysterectomized women.

Parcsini et al (1984) showed that there was steady rise in serum cholesterol value in women with bilateral oopherectomy within 6 months of operation.

Farrish et al (1990) showed significant increase in cholesterol level (P<0.05) from a mean of 3.57 mmol/1 to 4.21 mmol/1.

2. Serum Triglycerides

Oliver and Boyd (1959) showed a significant elevation of serum triglycerides in bilateral oopherectomized women

Sznaiderman and Oliver (1963) showed that serum triglycerides were significantly raised in women with premature menopause as compared to healthy women of same age group but there was not significant increase in the level after menopause.

Punnomen and Rawaria (1976) showed that serum triglycerides level rise significantly after 1 month of bilateral oopherectomy.

Notelovitz et al (1983) showed that serum triglycerides were higher in oopherectomized women.

Farrish et al (1990) showed no significant rise in serum triglyceride level in bilateral oopherectomized women.
3. **High Density lipoprotein**

High density lipoprotein particle transport cholesterol from peripheral tissue including vascular endothelium to liver where it is metabolized and excreted through bile. An impairment of High density lipoprotein will accelerate excess storage of cholesterol in endothelium of arterial wall, one of the factors leading to development of atherosclerosis. Lower the level of HDL, higher the risk of atherosclerosis.

HDL has 2 major subfractions – HDL₂ and HDL₃. Low level of HDL₂ are clearly related to high risk of atherosclerosis while total HDL and HDL₃ are not.

**William and Kannel (1976)** showed serum HDL levels are higher in women than is men.

**Punnoven and Rauramo (1980)** observed that HDL levels before and after one month of castration did not differ significantly.

**Notelovitz et al (1981)** showed that HDL levels in oopherectomized women were 27% lower than others of same age.

According to **Pausini et al (1974)** HDL levels showed an initial decrease and later significant increase during later 3 months.

**Farrish et al (1990)** measured HDL subfraction to assess any change in relative amount of cholesterol. No significant change was observed in either fraction.
**Low Density Lipoprotein and Very low density lipoprotein**

LDL and VLDL are directly related to atherogenicity of person. Their elevated levels can be correlated with conditions favouring atherogenesis. The value of LDL are calculated from standard formula. VLDL is 20% of serum triglycerides. VLDL is supposed to carry triglycerides found in liver or possibly in the intestine to body tissue where triglycerides and fattyacids are hydrolyzed by lipoprotein lipase enzyme. Metabolites are used for energy during metabolic process and remnant left behind are taken by liver and converted to LDL. Accumulation of remnant favours atherogenesis and estrogen is reported to enhance removal of remnant.

*Arnold and Ritterband et al (1963)* showed that mean serum cholesterol and percentage of betalipoprotein in oopherectomized women under 50 years were higher than hysterectomized women.

*Pausini et al (1984)* showed initial decline, then increase in apoprotein B level – the main carrier of LDL and VLDL fraction – a significant increase upto 12.5% of premenopausal value.

*Farrish et al (1990)* showed a significant rise in LDL cholesterol in the 6 weeks after operation from a mean of 3.57 m-mol/l to 4.21 mmol/l.
BONE MINERAL DENSITY

Osteoporosis is characterized by absolute decrease in bone mass per unit volume of anatomical bone and microarchitectural deterioration of bone tissue with normal mineral to matrix ratio, (both mineral and matrix decreased), leading to enhanced bone fragility and increased fracture risk.

With declining ovarian function and decrease of estrogen levels, the female skeleton undergoes increasing bone remodeling and rate of bone loss increases from the perimenopausal period. The normal bone building takes place till about 30 years of age and after that, as the part of natural aging process, the break down of bone is faster than the formation. The bone mass starts declining at around 40 years and rate of loss varies between 0.25 to 1% /year, the loss being higher in trabecular (1% to 5%) than cortical bone (Usha Krishna, 2000).

Bone density, measured by bone densitometry, is expressed as grams of mineral per Cm².

National osteoporosis foundation recommended B.M.D. Test for

1) Post menopausal female under the age of 65 years who have one or more risk factor beside menopause.

2) All post menopausal female over 65 years regardless of risk factor

3) Post menopausal females in whom fracture has occurred.
4) For females who are on prolonged treatment of H.R.T. or results of testing would affect their decision for or against HRT.

Bone mineral density is the most sensitive and specific test for osteopenia.

It is measured by -

a) **Dual Energy X-Ray absorbiometry (DEXA)** which can be of two type
   
   *Central* - for axial skeleton eg. :- Spine, hip, Whole body
   
   *Peripheral* - for wrist, middle finger, forearm, heel.

b) **Single energy X-ray absorbiometry** - for forearm and heel
c) **Quantitative Computed Tomography** - for trabecular bone only eg. Vertebral body.
d) **Peripheral Quantitative tomography** - for forearm
e) **Ultrasound** - for patella, heel, ankle

f) **Quantitative radiography.**
g) **Others** - Radiogammometry

   Quantitative MRI

   Single and Double Photon absorbiometry

   Radiographic Photodensitometry.

**DUAL ENERGY X-RAY ABSORBIMETRY (DEXA)**

It is the most accurate, gold standard, most widely used method now-a days. It is two dimensional and can not estimate depth or anteroposterior length of bone so cannot give true volumetric density. Results are variable measured form DEXA by different manufacturer so results of B.M.D. are compared to normal value using (T) score or (Z) score.
**T - Score**

Compares individual results to those in young healthy population that is matched for race and sex.

\[ T = \frac{\text{BMD value (measured)} - \text{BMD (mean young)}}{\text{SD Young}} \]

**Z - Score**

Compares individual results to those of an age matched population that is also matched for race and sex.

\[ Z = \frac{\text{BMD value (measured)} - \text{BMD (mean normal)}}{\text{SD normal}} \]

*Z* score is useful for people > 75 years old and in male as it is not advisable to compare them with normal healthy adults because of age related bone loss, measurement artefacts caused by osteophytes, spinal deformity extraskeletal calcification (eg. in aorta).

**Interpretation**

WHO has established general diagnostic categories of bone loss based on the degree of deviation from mean bone mass density (BMD) *(Kanis JA et al, 1994).*

<table>
<thead>
<tr>
<th>Category</th>
<th>Description</th>
</tr>
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<tbody>
<tr>
<td>Normal</td>
<td>Within 1 SD below normal (T Score equal to or above -1)</td>
</tr>
<tr>
<td>Osteopenia</td>
<td>1-2.5 standard deviation below normal adults (T score between -1 and -2.5)</td>
</tr>
<tr>
<td>Osteoporosis</td>
<td>&gt; 2.5 SD below normal, No history of fracture (T score at or below -2.5)</td>
</tr>
<tr>
<td>Severe osteoporosis</td>
<td>&gt; 2.5SD below normal, History of non violent fractures</td>
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</table>
**Advantages**

Minimum radiation exposure, short scanning time, variability of repeat reading is less (1%) and ability to scan entire skeleton

**Disadvantages -**

It is expensive, The changes with disease progression or therapy are small in relation to the variability of measurement. Anteroposterior measurement can not be measured but this last problem can be solved by lateral densitometry.

Bone density studies should be of spine and hip. Peripheral studies (eg. : ankle, forearm, finger) are for screening only. Those patients, whose peripheral study T scores are below -1 SD should have spine and hip bone density studies.

**Quantitative Computed Tomography and Quantitative MRI**

They give true volumetric bone mineral density but are more expensive & less accurate. There is greater exposure to radiation.

**Ultrasound**

It is less accurate than DEXA or SEXA but is relatively less costly. There is no radiation hazard. So it can be used as a screening procedure.

**Radiography**

It is the most important method for detecting fractures in osteoporosis. It shows bone loss only when it exceeds 30% or more.
**Indication**

a) Decrease in height > 2.5 to 3.8 cms to rule out asymptomatic vertebral fracture.

b) Presence of kyphosis or backpain, specially in post menopausal females.

c) To rule out cause of fracture as malignancy.

**Hall marks of Osteoporosis on X-Ray**

a) Progressive decrease in vertebral mineral density (ground glass appearance).

b) Prominence of vertical striations and loss of horizontal trabeculations.

c) Biconcave vertebra or codfish vertebra.

d) Progressive vertebral compression with anterior wedging (reduction in anterior height) and collapse (reduction in anterior and posterior height).

**Single photon absorbmity**

It has low radiation dose but is limited to peripheral skeleton and takes 10-15 minutes scan time. As the isotope has short half life, it requires regular replacement.

At present, DEXA is considered one of the best technique as its precision and accuracy are good. It has high resolution and low radiation and can measure all areas, specially the spine and proximal femur. It can be well documented and co-relates to fracture risk and scan time is less than 5 minutes *(Usha Krishna et al 2000).*

Estrogen has positive effect on bone mass. It produces positive effect on bone mass by following actions -
Estrogen decreases osteoblastic production of interleukin-6 (IL-6), tumor necrosis factor (TNF), thereby interfering with osteoclast activation.

It helps in activation of Vit. D₃ in kidney.

It promotes positive calcium balance partly by inducing renal hydroxylase enzyme.

**Bone cells express estrogen receptor α**

Incidence of osteoporotic fractures in 2-3 times greater in women than men because peak bone mass is lower and there is accelerated loss after menopause ([Melton et al, 1992](#)).

**Douglas P.Kiel et al, 1987** showed in a large cohort study that post menopausal use of estrogen protects against subsequent hip fracture in women.

**Felson et al (1993)** demonstrated that after 10 years of HRT, bone mineral content was significantly higher in HRT group compared to those who did not receive any treatment.

Daily treatment with Alendronate progressively increases the bone mass in the spine, hip and total body and reduces the incidence of vertebral fractures, progression of vertebral deformities and height loss in post menopausal women with osteoporosis ([URI A. Liberman et al, 1995](#)).

**Post menopausal estrogen / progestin interventions (PEPI) trial, 1996** showed that HRT increases bone density in the spine, hip and produces reduction in bone turnover.

**Schneider et al (1997)** showed that estrogen initiated in early menopausal period and continued into late life is associated with highest bone density.
According to work of Delmas PD 1997, Women receiving Raloxifene had significant increases from baseline values in bone mineral density of lumbar spine, hip, total body where as those receiving placebo had decrease in bone mineral density.

Clemett D et al 2000, John Ston CC et al, 2000 showed that oral raloxifene consistently increased lumbar spine, femoral neck, total hip and total body BMD relative to baseline values in post menopausal women.

Calcitonin is a synthetic hormone that inhibits bone resorption. A five year trial conducted by Chestnut CH (2000) showed that the only 200 µg dose produced significant (36%) reduction of vertebral fractures, with no reduction in hip fractures but no significant change in either bone density or bone turnover.

Black DM et al, 2000 demonstrated that Aledronate, a bisphosphonate reduces the incidence of fractures at the spine, hip and wrist by 50% in patients with osteoporosis.

**ENDOMETRIAL BIOPSY**

In the menopausal period, loss of effect of estrogen on uterus is that uterus becomes smaller and ratio between the body and cervix reverts to the 1:1 ratio. The uterus and cervix show gradual shrinkage due to myometrial atrophy. Estrogen withdrawal causes atrophy of endometrium and it is more marked in the functional layer than in the basal layer. The endometrium is thin i.e., only 1-3 mm in thickness with
atrophic and inactive glands. The atrophic endometrium is susceptible to infection resulting in senile endometritis and post menopausal bleeding. In rare cases, the endometrium becomes hyperplastic under the influence of extra genital estrogen (estrone) produced in the peripheral fat from epiandrostenedione.

According to Smith DC, 1975, unopposed estrogen is associated with an increased risk of endometrial cancer.

Mc Donald et al, 1977 conducted a study which indicated that the risk of developing endometrial cancer increases with duration of ERT use.

Shapiro S et al , 1985 also showed that risk may persist for more than 10 years after discontinuation of just 1 year of ERT.

Result of PEPI trial, 1995 show that women taking unopposed estrogen during the 3 year study had a significantly increased risk for endometrial hyperplasia compared with those taking estrogen plus progestins (34% versus 1%)

Boss SM, 1997 has shown in a study that Raloxifene does not induce endometrial proliferation. Statistically significant estrogenic effects were noted in 77% of estrogen treated women versus 15% of placebo treated women versus 0% of Raloxifene treated women.

Preclinical data from animal models indicates that Raloxifene does not stimulate endometrium nor does it increase uterine thickness (Jordon VC et al 1998).
**Davies GC et al, 1999** have proved that Raloxifene 60 mg/d for upto 30 months is not associated with vaginal bleeding or increased endometrial thickness in post menopausal women.

**Cummings SR et al, 1999 in MORE study** showed that Raloxifene did not increase risk of endometrial cancer. After 40 months of followup, rate of endometrial cancer was slightly lower in these women.

**Fugere P et al, 2000** proved that after a period of 24 months, the women who received ERT showed significant changes in endometrial thickness and uterine volume. In contrast, Raloxifene treated group exhibited no changes in either parameters.

In latest reports from **MORE study, Johnson S. et al 2001** showed that after 48 months of followup, raloxifene continues to maintain a slightly lower rate of endometrial cancer compared with placebo.

**HORMONAL REPLACEMENT THERAPY**

Hormone replacement therapy is available in many formulations and combination and can be given by various routes also.

**Oral Preparation**

HRT using estrogen alone

Cyclic estrogen progesterone preparation.

Continuous estrogen - progesterone preparation

Estrogen - androgen HRT
Non Oral Preparation

Vaginal cream and pessaries
Percutaneous gel
Transdermal patch.

**Oral Preparations available are** -

A. Conjugated Steroidal Estrogens
   a) Estrones
      Conjugated equine estrogen
      Esterified estrogen
   b) Estradiols
      Estradiol Valerate
      micronized 17 beta estradiol
   c) Estriols
      Estriol hemisuccinate

B. Unconjugated steroid analogue
   17 Ethynl estradiol
   17 Ethynl estradiol 3 methyl ether
   17 Ethynl estradiol 3 cyclo pentoether

C. Synthetic Estrogen Analogues
   Bengestrol
   Chlorotrenanene
   Dinestrol
   Diethylstilestrol
   Hexestrol

**Non Oral Preparations are** -
Earliest non oral routes of administration used: -

Vaginal cream
Pessaries
Subcutaneous estradiol implant

More recently

Percutaneous gel
Hormone containing vaginal rings

Most recent

Transdermal therapeutic system in the form of patch
- Only estrogen containing
- Estrogen & Progesterone.

For over three decades, estrogen replacement therapy in the form of oral conjugated equine estrogen or estradiol preparation has been widely used for relief of menopausal symptoms. But this has certain disadvantages as high doses of estrogen must be administered because of rapid metabolism to estrone and pharmacologically inactive metabolism and inactivation of estrogen within the gut wall and liver. The relatively high dosages, which are required to compensate for this, result in high peak plasma concentrations of estrone and estradiol.

Hepatic synthesis of certain protein is enhanced by high concentration of estrogens in the portal circulation, leading to increased plasma level of rennin substrate, Sex hormone binding globulin, Thyroxine binding globulin and cortisol binding globulin which leads to the development of
hypertension, gall bladder disease and thrombosis (Geola et al 1980 & Mandel et al, 1982).

Parenteral administration of estrogen avoids the hepatic first pass metabolism but previously available methods have limitation that high hormone level immediately after administration (with injectable), inadequacy of dosage adjustment (with subcutaneous pellet), inadequate systemic delivery and reduced patient acceptability (Intravaginal cream) (Mandel et al 1983).

Recently, a transdermal delivery method for estradiol administration has been developed to provide a simple approach to deliver estrogen therapy and simultaneously avoid the 1st pass metabolism through liver. It is in the form of thin adhesive patch, with drug reservoir where drug lies between occlusive backing layer and a rate controlling microporous membrane. Thus drug delivery is at constant and predictable rate.

In various clinical studies, it has been observed that beneficial effects of transdermal estradiol on plasma gonadotropin, maturation of vaginal epithelium, metabolic parameter of bone resorption and menopausal symptoms appear to be comparable to those of oral and other parenteral preparation, while the undesirable effect of oral estrogen on hepatic metabolism are avoided. (Pedwick et al 1985), Virgil A place et al 1985 and Chalkowski et al 1986).

Transdermal estradiol results in increase in estradiol concentration rather than estrone concentration and it result
in plasma estradiol; estrone ratio which more closely approximates that found in premenopausal women at early to mid follicular stage (40-100 ng/l) (*Powers et al 1975, Selby et al 1985*).

Non oral routes of administration of estrogen do not appear to have same magnitude of effects on lipid profile (*Chetkowsky, 1986*).

Progesterone, when properly administered, effectively prevent adenomatous hyperplasia and cause regression of pre existing adenomatous hyperplasia in the majority of the patients (*Studd et al, 1980; Gambrell et al 1982; Gal et al 1983*).

Progesterone reduces the concentration of estradiol E2 receptor (*Hsuch et al, 1975*) and increase the activity of dehydrogenase that converts estradiol to estrone, a biologically less active estrogen (*Treng et al 1977*) and cause endometrial cells to differentiate to a secretary state rather than proliferative state (*Novak et al, 1979*).

Estrogen, when combined with progesterone tend to loose some of their beneficial effects (*Knoop et al, 1986; Tikkanan et al, 1986*). However, such effects are dependent on chemical structure, dose of drug and hormonal status of the recipient (*Geola et al, 1980; Jensen et al 1987; Mettsson, 1984; Ottoson 1984 and La-rosa et al 1985, Bush et al, 1987*).

LDL-C is increased and HDL-C specially HDL₂ is increased due to effect of progesterone. These changes can be
correlated with increased risk of coronary heart disease in epidemiological studies (Wallace et al, 1987). However, 19-nortestosterone derivatives eg. Norethindrone, levonorgestral have a less deleterious effect on lipids and lipoproteins than C-21 progesterone derivatives such as megestral acetate, medroxy progesterone acetate.

Various studies have been done to study the effect of HRT on menopause and its effect on lipid profile.

Hallberg and Svanberg 1967 compared cholesterol phospholipid and triglyceride concentration in pre and post menopausal women. It was suggested that post menopausal women have on atherogenic risk profile, having higher plasma level of cholesterol, triglycerides VLDL, LDL than premenopausal counterparts.

No significant change in cholesterol level, significant rise in total triglyceride level, & HDL cholesterol and reduction in LDL cholesterol level was observed in women being treated with conjugated equine estrogen at a dose of 2.5 mg daily for 6 months - shown by Furmen et al 1967

Utian et al 1972 compared the effect of equine estrogen with estradiol valerionate and found that their was no change in cholesterol level in women with equine estrogen but there was decrease in cholesterol level in women with estradiol valerionate.

Bengssto & Lidgnist, 1979 showed that surgically induced menopause by oopherectomy results in profound alteration of lipid balance as compared with age matched
women with intact ovaries. Significantly higher cholesterol, triglyceride and lower relative proportion of HDL were observed in oopherectomized women.

In the lipid research clinic study by Wahl et al 1983, lipid profile was studied in 370 untreated post menopausal patients and 839 post menopausal patients using conjugated equine estrogen. An average decrease of 48% in serum cholesterol and 13.6% reduction in LDL cholesterol level was found in treated group as compared to that of untreated group. Also there was an average increase of 23.7% and 13% in serum triglycerides level and HDL cholesterol level in treated group. An average increase of 12.5% in LDL level was found in treated group.

Padwick et al (1985) studied the efficacy, acceptability and metabolic effect of transdermal estradiol 0.05 mg/d cyclically. The result showed that transdermal estradiol significantly increased plasma level of estradiol, estrone and urinary concentration of estradiol and produced significant improvement in menopausal symptoms and vaginal cytological findings. The drugs were well tolerated and no systemic side effects were noted.

Chetokwoski et al (1986) conducted a dose related response study in post menopausal women to compare the physiologic effects of transdermal estradiol and oral conjugated equine estrogen. The doses were 25,50, 100 and 200 ngm of transdermal estradiol/ 24 hours and .625 mg and 1.25 mg oral equine estrogen. They showed that
transdermal estradiol elicited many of the desirable actions of estrogen while avoided the deleterious pharmacological effects of oral estrogen.

Colditz GA et al (1987) analyzed prospectively a cohort of 1,21,700 U.S. Women, 30-55 years of age who were followed from 1976-1982 and suggested that in contrast to natural menopause, bilateral oopherectomy increased the risk of coronary heart disease which could be prevented by estrogen replacement therapy.

Utian WH (1987) studied that transdermal therapeutic system was both effective and well tolerated and have no effect various liver protein. Moderate bleeding occurred in patients with intact uterus which could be controlled by addition of progesterone. The incidence of endometrial hyperplasia and breast tenderness has been relatively low and minor side effects had been limited. The patients showed a preference for transdermal method of estrogen delivery.

Stanczyk FZ et al (1988) compared the efficacy of transdermal and subdermal routes of estrogen administration 20 post menopausal women were randomized to receive either two 25 mgm estradiol pellets subdermally or 1 mg estradiol transdermal patch twice weekly. Significant rise in HDL cholesterol and decrease in total cholesterol was noted at 12 weeks with pellets but only at 24 weeks with patch. Urinary calcium/creatinine ratio was reduced more consistently with the pellet than with the patch Hot flushes were eliminated in all.
Lobo RA (1990) studied that estrogen appeared to protect against development of cardiovascular disease by decreasing LDL and increasing HDL levels, by induction of LDL receptor and destruction of hepatic lipase which degrades HDL-C.

Crook et al (1992) studied the comparative effects of trans dermal and oral estrogen-progesterone therapy on lipid profile at 3 and 6 month in 90 women. The regimen included transdermal estradiol 0.05 mg/d alone or combined with norethindrone acetate .025 mg/d for 14 d and oral conjugated equine estrogen .625 mg/d alone or combined with oral norgestrol 0.15 mg/dl for 12 d. Reduced serum level of total and LDL cholesterol were reported during both phases of transdermal and oral therapy with no significant effect on HDL levels during oral therapy without progesterone, whereas transdermal therapy caused 4% reduction. Oral estrogen increased serum triglycerides by 15% where as there was reduction in triglyceride level by 80% with transdermal estrogen.

Adomi et al 1993 compared the effect on lipoprotein fraction with transdermal estrogen (0.05 mg) and oral conjugated equine estrogen (0.625 mg) in 81 post menopausal women. Both groups were combined with medroxy progesterone acetate for 12 days of the cycle. A third group served as control. The control group experienced no observable significant changes in their lipid profile after 1 year of follow up. Both treated groups experienced significant [39]
decrease in total LDL cholesterol, HDL cholesterol decrease in the transdermal estradiol group, rise slightly in oral estrogen group. Serum triglycerides significantly decrease in the transdermal group only.

_Pong et al (1993)_ studied the effect of transdermal estradiol alone and when given with medroxy progesterone acetate over a 36 week period. No significant changes were observed in HDL and very low density lipoprotein cholesterol and triglycerides. Statistically significant decrease in total cholesterol were not seen until the 72nd week.

_Woodruff et al (1994)_ studied the incidence of endometrial hyperplasia in post menopausal women taking conjugated estrogen with medroxy progesterone acetate or conjugated estrogen alone.

PEPI Trial 1995 studied the effect of estrogen or estrogen / progesterone regimens on heart disease risk factors in 875 healthy post menopausal women.

The patients were divided into following groups :

1) Placebo
2) Conjugated equine estrogen .625 mg/dl.
3) CEE .625 mg/d plus cyclic medroxy progesterone acetate 10 mg/dl for 12 days. They found that estrogen alone or in combination with a progesterone improves lipoprotein and lower fibrinogen levels without detectable effects on post challenge insulin and blood pressure.

Unopposed estrogen is the optimal regimen for elevation of HDL- Cholesterol but the high rate of endometrial
hyperplasia restricts its use in women with an intact uterus. In women with intact uterus, CEE with cyclic MPA has the most favourable effect on HDL-C and no excess risk of endometrial hyperplasia.

**Rosaw et al (1996)** studied that estrogen improves serum lipid profile, carbohydrate metabolism and insulin sensitivity, prevents the formation and development of atherosclerotic plaques, reduces blood pressure and plasma fibrinogen levels and favourably affects overall cardiac features.

Four years randomized study from the university of Texas showed for 1st time an additive effect of intermittent cyclical etidronate and HRT on bone mineral density in vertebral and the hip bone (**Jha, U.P. 1997**).

**RALOXIFENE - SELECTIVE ESTROGEN RECEPTOR MODULATOR**

In order to broaden the treatment options available to post menopausal women, research efforts have been directed at the development of compounds that maintain the vasomotor, skeletal and cardiovascular benefits of estrogen replacement therapy but have little effect to no significant adverse effect on reproductive organs and the clotting process. Raloxifene is a selective estrogen receptor modulator and it belongs to BENZOTHIOPHENES class of compounds.

It's biological actions are largely mediated through binding to estrogen receptors. Estrogen agonist activity is seen
on bone turnover and several cardiovascular intermediate end points such as serum lipids, lipoproteins and fibrinogen. It has no proliferative effects on uterine or mammary tissues and antagonizes estrogen effects on these tissues (Black LJ et al, 1983, Evans G et al, 1993; Sato M et al, 1994).

Both nuclear estrogen receptors (mediating genomic effects of estrogen) and membrane estrogen receptors (mediating non genomic effects of estrogen) exist. Genomic estrogen receptors are found to have two major distinct isoforms - ERα & ERβ. β isoform is expressed more abundantly in some tissues like - bone, prostate, hippocampus where as α isoform is found primarily in breast, liver, uterus, ovary and central nervous system.

Raloxifene binds to ER α with high affinity, Similar to that of 17 β estradiol (Glassbrook L et al, 1993). It also exhibited high affinity binding interactions with ER β subtype (Gize et al, 1997). The unique structure allows it to bind to estrogen ligand binding pocket of estrogen receptors. Both occupy the same site but the basic side chain of Raloxifene extends out of the binding cavity and displaces one of the components of the receptor. This is a region of the ER thought to be important in some of the transcriptional activation functions of the receptors.

There are five structural domains of estrogen receptors. The AF-2 domain is a region of receptor required to activate gene sequences known to modulate estrogen activity in reproductive tissues such as the uterus and breast. The
conformation of ERα bound to estradiol is conducive to interaction with AF-2 where as conformation induced by raloxifene does not favour such an interaction.

In addition to the classic estrogen response element (ERE), multiple genomic sequences appear to exist through which ER : ligand complex may regulate transcription (Yang NN et al 1996). These may include activator protein / promotor, the retinoic acid receptor α-1 promotor, the transforming growth factor β promotor (TGFβ). Raloxifene has been shown to stimulate production of TGFβ3, a cytokine responsible for bone resorption, which substantially inhibits differentiation and resorption activity of osteoclast like cells and may account for antiresorptive properties of raloxifene on bone.

**Pharmacokinetics**

The drug is absorbed rapidly after oral administration. 60% of oral dose is absorbed but presystemic glucuronide conjugation is extensive. Absolute bio availability is 2%.

It can be administered without regards to meals. It is 95% bound to plasma proteins. The drug is primarily excreted in faeces and < .2% excreted unchanged in urine.

**CLINICAL FINDINGS**

(a) **Effect on lipid profile**

The estrogen agonist effects of raloxifene on lipids have been confirmed clinically. The traditional markers of cardiovascular disease risk in post menopausal women include -
Elevated levels of LDL, low levels of HDL, High triglycerides, family history of heart disease, Diabetes, hypertension, obesity, physical inactivity, smoking.

In recent years, several new markers for predicting the risk of CVD have emerged:

(i) **C-Reactive Protein**

Levels are higher in people with heart disease. In a 3 years study, women with the highest level of CRP had a five fold increase in the risk of developing CVD and a seven fold increase in the risk of having a heart attack or stroke (*Ridkar PM et al, 2000.*)

(ii) **Homocysteine -**

Elevated levels of homocysteine can be correlated with artery damage, blood clotting, myocardial infarction, stroke and other manifestations of CVD. In women approaching menopause its levels are increased and it is thought to play a role in increased incidence of vascular disease, Cancer, osteoporosis in post menopausal women (*Bores /GH et al, 1983, Anker G, 1995*).

(iii) **Fibrinogen**

High levels of fibrinogen can restrict blood flow and lead to hardening of arteries and accumulation of plaques. Its increased levels are associated with CVD. (*Stec JJ, 2000*).

(iv) **Lipoprotein (a)**

An elevated blood concentration of lipoprotein (a) is associated with an increased risk of atherosclerosis and coronary artery disease (*Zenner G, 1986*).
Love RR et al (1991) ; Draper MW, (1996) have conducted studies showing that raloxifene is capable of reducing LDL levels without affecting HDL or triglycerides levels significantly in post menopausal women.

Sheoman DA et al (1994) have shown that raloxifene and tamoxifen decrease the levels of lipoprotein a.

According to Grey AB et al, 1995. SERMS specially raloxifene have a greater ability to lower fibrinogen levels compared with HRT.


Mjatovic V et al (1999) demonstrated that long term raloxifene treatment significantly lowers serum lipoprotein (a) levels in post menopausal women.


According to Vonholst T (2000) De leo V (2001), Raloxifene at a dose of 60 mg/d reduces serum concentration of LDL cholesterol and total cholesterol as well as cause reduction in fasting homocysteine levels.

(b) Effect on Osteoporosis

Osteoporosis is now defined as a skeletal disorder characterized by compromised bone strength, which predisposes the patient to an increased risk of fracture. The element of bone strength are bone density and bone quality, former being measured by bone densitometry. Bone quality
includes bone architecture, turnover damage accumulation (eg:- microfractures), mineralization.

*Delma PD et al (1997)* demonstrated that women receiving each dose of raloxifene had significant increases from baseline values in bone mineral density of lumbar spine, hip and total body where as those receiving placebo had decreases in bone mineral density.

*Burckhardt P., Khovidhunkit W, Agnusdei D (1999)* demonstrated that Raloxifene can be used in women free of climacteric symptoms for the prevention and treatment of post menopausal osteoporosis, alone or in combination with calcium , vitamin D., bisphosphonates and calcitonin.

Raloxifene prevents post menopausal bone loss (lumbar vertebrae, hip, radius and reduces the risk of osteoporotic fracture (*Fontana A et al, 1999*).

*Bruce Ettinger et al (1999)* demonstrated that compared with placebo group, bone mineral density increased after 36 months by 2.1% and 2.6% at the femoral neck and spine in the 60 mg raloxifene group & by 2.4% 2.7% at the femoral neck and spine in the 120 mg raloxifene group.

According to Von Holst T. (2000), there is evidence that bone mineral density is growing with treatment. In a three year study (MORE), a statistically significant decrease of lumber spine fractures was demonstrated.

(c) **Effect on Endometrium**

One particular advantage of Raloxifene over HRT is its lack of proliferative effect on endometrial tissue.
Susan M. Boss, BS et al, 1997 performed double blind placebo controlled 8 week study which showed that raloxifene did not induce histopathologic evidence of endometrial stimulation in healthy post menopausal women.

Delma PD et al (1997) showed that endometrial thickness was similar in the raloxifene and placebo groups at all times during the study.

According to Burckhardt P et al, Khovidhunkit W et al (1999), SERMS can exert the known estrogen like effects on bone and lipids without exerting any action on the endometrium and the breast, a potentially ideal profile for post menopausal hormone replacement treatment. Endometrial thickness was unchanged during raloxifene therapy and no patients developed proliferative endometrium. In estrogen deficient state, raloxifene acts as an estrogen antagonist. A high estrogen milieu blunts the antagonistic effects of raloxifene on the uterus.

Endometrial neutrality of raloxifene was also demonstrated by Cano A, 2000.

Fugere P et al, 2000 compared the uterine effects of raloxifene with continuous combined HRT in post menopausal women. Assessment was done by endometrial biopsy and transvaginal ultrasonography. In the raloxifene group at the end point of study, 94.4% showed normal benign post menopausal endometrium where as 78.7% of patients treated with continuous combined HRT showed normal benign post menopausal endometrium. Mean endometrial thickness was
unchanged from baseline with raloxifene and was increased by 5 mm with continuous combined HRT.

(d) **Protective Effect on breast Cancer**

Raloxifene has antiproliferative effect on human breast cancer cells and inhibits mammary carcinogenesis in animal models of breast cancer.

*Freedman M et al, 2001* assessed changes in mammographic density in healthy post menopausal women in osteoporosis prevention trial. The study showed that the women who received placebo showed a significant decrease in breast density as did the women who received raloxifene. The women, who received estrogen on the other hand, experienced a non significant increase in breast density.

*MORE trial, 2001* in post menopausal women with osteoporosis showed that raloxifene reduced the risk of newly diagnosed invasive breast cancer by 72% in these women. The risk of estrogen receptor positive invasive breast cancer was reduced by 84%.

**Adverse Effects**

Raloxifene at doses upto 150 mg/day has been well tolerated in clinical trials in post menopausal women. No clinically important changes in hematological, renal or hepatic laboratory variables were observed.

The only adverse effect possibly related to Raloxifene was vasodilatation (hot flushes), most common in the raloxifene Hcl 600 mg group, according to *Draper MW, et la 1996.*
According to **Delmas PD et al, 1997** the proportion of women receiving raloxifene who reported hot flushes or vaginal bleeding was not different from that of women receiving placebo.

**Burckhardt P., 1999** has shown that as in hormone replacement therapy, thromboembolism and leg cramps occur more frequently.

**Agnusdei D. et al, Davis GC et al, 1999** have shown that hot flushes were the most frequently observed undesirable effects at frequency slightly higher in the raloxifene group (25%) than in the placebo group (18%). This undesirable effect was of the low intensity and generally occurred during 1st 6 months of treatment. It did not cause a higher drop out rate.

According to **Weerapan Khovidhunkit, et al, 1999**, incidence of breast tenderness and vaginal bleeding with raloxifene was similar to that seen with placebo but significantly less than that produced by estrogen therapy. The most serious side effect associated is a three fold increase in the risk for venous thromboembolism, an increase similar to that seen with estrogen therapy.