**METABOLISM**

**Vitamin D / Vit-D:** It is a fat-soluble, secosteroid, pro-hormone produced photochemically in the skin from 7-dehydrocholesterol (7-DHC). It is an essential vitamin for life because it is primary biological regulators of calcium, magnesium, phosphate homeostasis and multiple other biological effects.

**Structure and forms of Vit-D:**

The term “Vitamin D” refers to a family of compounds that are derived from cholesterol. There are two major forms of Vit-D: vitamin D₂ (ergocalciferol) and vitamin D₃ (cholecalciferol). Vitamin D₃ is produced in human skin following exposure to sunlight, whereas vitamin D₂ originates in certain plants and fungi. Both vitamin D₂ and vitamin D₃ are metabolized into pro-hormones by CYP27A, CYP2R1 and certain other cytochrome P450 enzymes located in the liver. These enzymes metabolize vitamin D₂ and vitamin D₃ into the pro-hormone known as 25 (OH) D₂ and 25 (OH) D₃ (calcidiol). The two pro-hormones are collectively referred to as “25 (OH) D”. Now, these pro-hormones 25 (OH) D₂ and 25 (OH) D₃ are further metabolized by the kidney by a 1α-hydroxylase enzyme known as CYP27B1, located in the proximal kidney tubule into 1, 25 (OH)₂ D₂ (ergocalcitriol) and 1, 25 (OH)₂ D₃ (calcitriol). These hormones, collectively referred to as “1, 25 (OH)₂ D”, is secreted by the kidneys into the bloodstream for systemic delivery (Zhang and Naughton, 2010; DeLuca HF, 2014; King, 2017).

**Sources of Vit-D:** Sunlight exposure is the major source whereas Vit-D can also be obtained from the dietary source like oily fish (mackerel, tuna, salmon, and sardines), cod
liver oil, cheese, egg yolks, and some fortified products like dairy products, cereals, juices, soy milk, contain fewer amounts of Vit-D.

Structure and forms of Vit-D. (Source: King, 2017)

**Synthesis and metabolism of Vit-D:** The production of Vit-D₃ (cholecalciferol) in the skin is not an enzymatic process. It is produced from 7-DHC through a two-step process in which the B ring is broken by UV light (spectrum 280–320 nm UVB) radiation from the sun, forming pre-D₃ that isomerize to D₃ in a thermo-sensitive but a non-catalytic process. Both UVB intensity and skin pigmentation level contribute to the rate of Vit-D₃ formation (Holick et al. 1980; Japelt, and Jakobsen, 2013; Bikle, 2014). Now, this is transported in the blood by the Vit-D binding protein (DBP, a specific binding protein for Vit-D and its metabolites in serum) to the liver. In the liver Vit-D is hydroxylated at C-25 by one or more
cytochrome P450 Vit-D 25 hydroxylases (including CYP2R1, CYP2D11, and CYP2D25), resulting in the formation of 25 (OH) D₃. It has been suggested that CYP2R1 is the key enzyme required for 25-hydroxylation of Vit-D since a homozygous mutation of the CYP2R1 gene was found in a patient with low circulating levels of 25(OH) D₃ and classic symptoms of Vit-D deficiency (Cheng et al. 2004). 25(OH) D₃, the major circulating form of Vit-D, is transported by the DBP to the kidney. In the kidney, magalin, a member of the LDL receptor superfamily, plays an essential role in the endocytic internalization of 25(OH) D₃ (Nykjaer et al. 1999). In the proximal renal tubule 25(OH) D₃ is hydroxylated at the position of carbon 1 of the A ring, resulting in the hormonally active form of vitamin i.e. 1, 25 (OH)₂ D₃ which is responsible for most of the biological actions of Vit-D. The cytochrome P450 monooxygenase 25(OH) D 1α-hydroxylase (CYP27B1) which metabolizes 25(OH) D₃ to 1, 25(OH)₂ D₃ is present predominantly in the kidney. This enzyme is also found in extrarenal sites including placenta, monocytes, and macrophages (Weisman et al. 1979; Gray et al. 1979; Esteban et al. 2004; Stoffels et al. 2007). As with all mitochondrial P450 containing enzymes, during the 1α-hydroxylase reaction electrons are transferred from NADPH to NADPH-ferredoxin reductase through ferredoxin. Inactivating mutations in the 1 α-hydroxylase gene result in Vit-D dependency rickets (VDDR) type 1 in spite of normal intake of Vit-D, indicating the importance of the 1α-hydroxylase enzyme (Kitanaka et al. 1998).

25 (OH) D₂ differs from 25 (OH) D₃ in having a double bond between C₂₂ and C₂₃ and a methyl group at C₂₄ in the side chain. These differences from 25 (OH) D₃ in the side chain lower its affinity for DBP resulting in faster clearance from the circulation (Hollis, 1984; Horst et al. 1986; Houghton and Vieth, 2006). Therefore, unless given daily, D₂
supplementation does not result in as high a blood level of 25 (OH) D as comparable amounts of D3 (Tripkovic et al. 2012). On the other hand, 1, 25(OH)₂ D₂ and 1,25(OH)₂ D₃ have comparable affinities for the VDR (Hollis, 1984).

Schematic representation of the synthesis, metabolism of Vit-D by regulating calcium, phosphorus and bone metabolism (Source: Holick, 2008)
Metabolism of 25(OH) D to 1, 25(OH)₂ D for non-skeletal functions.
(Source: Holick, 2008; Arabi et al. 2010)

Extra-renal production of Vit-D hormones provides a “local” supply of hormones that often depends on sufficient levels of circulating 25(OH) D. Active form of Vit-D i.e. 1, 25(OH)₂ D₃ generates a biological response by the presence of its cognate receptors in selected target organs and tissues. The genomic responses of 1, 25(OH)₂ D₃ are mediated by the formation of their ligand-receptor complex with their cognate nuclear receptor. 1, 25(OH)₂ D₃ serves as a chemical messenger that transmits signals and rapid responses (RR) (e.g. opening of ion channels). A number of extra-renal sites such as immune cells epithelia of skin, gut, prostate, lung, bone, parathyroid gland, and pancreatic islets may provide 1, 25(OH)₂ D₃
for local use as an intracrine or paracrine factor (Bikle, 2009). Locally produced 1, 25(OH)₂D₃ also inhibits the expression and synthesis of parathyroid hormone. The 1, 25(OH)₂D₃ produced in the kidney down-regulates renin production and stimulates pancreatic β-cell insulin secretion. Control of 1, 25(OH)₂D₃ by non-renal tissues depends on the organ concerned. In macrophages and keratinocytes, CYP27B1 is induced by invading organisms (Schauber et al. 2007). When monocytes and macrophages are stimulated by an infectious agent, the expression of vitamin D receptor and 25-hydroxyvitamin D-1α hydroxylase is up-regulated, so that more 25(OH) D is converted to 1, 25(OH)₂D.

**Serum 25(OH) D levels guidelines recommended by various organizations:**

<table>
<thead>
<tr>
<th></th>
<th>International Endocrine Society</th>
<th>Vitamin-D Council</th>
<th>Food &amp; Nutrition Board</th>
<th>Testing Laboratories</th>
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</thead>
<tbody>
<tr>
<td><strong>Deficient</strong></td>
<td>0-20 ng/mL</td>
<td>0-30 ng/mL</td>
<td>0-11 ng/mL</td>
<td>0-29 ng/mL</td>
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<tr>
<td><strong>Insufficient</strong></td>
<td>21-29 ng/mL</td>
<td>31-39 ng/mL</td>
<td>12-20 ng/mL</td>
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<tr>
<td><strong>Sufficient</strong></td>
<td>30-100 ng/mL</td>
<td>40-80 ng/mL</td>
<td>&gt;20 ng/mL</td>
<td>30-100 ng/mL</td>
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<tr>
<td><strong>Toxic</strong></td>
<td>-</td>
<td>&gt;150 ng/mL</td>
<td>-</td>
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</table>

**Diagnostic importance of serum 25(OH) D is more than serum 1, 25(OH)₂ D:** It is widely accepted that the determination of serum 25(OH) D level is the best indicator of Vit-D status. This is due to the fact that serum 25(OH) D is a measure of the cellular 25(OH) D concentrations which in turn is converted to cellular 1, 25(OH)₂D by loosely regulated extra-renal CYP27B1. Thus, intake of Vit-D by diet or supplementation cause increase in serum 25(OH) D concentration which results in an increase of cellular 1, 25(OH)₂D₃ and increased the action of vitamin D-dependent gene. Along with this 25(OH)
D is the major storage and circulating form with a half-life of 2-3 weeks. While serum 1, 25(OH)₂ D has proven to be a poor biomarker of Vit-D action because it is not an accurate predictor of cellular concentration of 1, 25(OH)₂ D in all target cells and has a short lifespan of few hours (Holick, 2008; Bikle, 2009).

**Prevalence of Vit-D Deficiency and its associated risk factor:**
Hypovitaminosis D is a major health problem in both developed and developing countries across the globe. In India, despite ample sunlight (required for the cutaneous synthesis of Vit-D), Vit-D deficiency has been documented to be in range of 50-90% among all age-groups (Goswami et al. 2000; Arya et al. 2004; Vupputruri et al. 2006; Zargar et al. 2007; Harinarayan et al. 2011; Gupta and Gupta, 2014; Mehlawat et al. 2014; Bachhel et al. 2015). In North India (27°N), 96% of neonates (Sachan et al. 2005), 91% of healthy school girls (Puri et al. 2008), 78% of healthy hospital staff (Arya et al. 2004), and 84% of pregnant women (Sachan et al. 2005) were found to have hypovitaminosis D. In South India (13°N), hypovitaminosis D is equally prevalent among different population groups (Harinarayan, 2005; Harinarayan et al. 2007). Hypovitaminosis is equally prevalent among rural and urban subjects (Balasubramanian et al. 2003; Sachan et al. 2005; Sahu et al. 2009; Goswami et al. 2008; Mithal et al. 2009).

Aging is associated with decreased concentrations of 7-DHC, the precursor of vitamin D₃ in the skin in elderly people. A 70-year-old has approximately 25% of 7-DHC than a young adult does and thus has a 75% reduced capacity to make vitamin D₃ in the skin (Holick, 1989). Because Vit-D is fat soluble, it is readily taken up by fat cells. Obesity is associated with Vit-D deficiency, and it is believed to be due to the sequestration of Vit-D by the large
body fat pool (Wortsman et al. 2000). A study conducted amongst postmenopausal women to evaluate their dietary calcium and Vit-D status documented that, 18% subjects had normal Vit-D levels, 52% had insufficiency and 30% had Vit-D deficiency (Harinarayan, 2005). Marwaha and colleagues in 2011 reported that Vit-D deficiency was present in 91.2% and insufficiency in 6.8% in elderly subjects (Marwaha et al. 2011).

<table>
<thead>
<tr>
<th>Decreased Vit-D synthesis</th>
<th>Skin pigmentation, physical agents blocking UVR exposure, clothing, latitude, season, air, pollution, cloud cover, altitude</th>
</tr>
</thead>
<tbody>
<tr>
<td>Decreased nutritional intake of vitamin</td>
<td>Strict vegan diet</td>
</tr>
<tr>
<td>Age and physiology related</td>
<td>Elderly, obese and institutionalized</td>
</tr>
<tr>
<td>Decreased maternal Vit-D stores</td>
<td>Exclusive breastfeeding</td>
</tr>
<tr>
<td>Malabsorption</td>
<td>Celiac disease, pancreatic insufficiency (cystic fibrosis), biliary obstruction (biliary atresia)</td>
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<tr>
<td>Decreased synthesis</td>
<td>Chronic liver disease</td>
</tr>
<tr>
<td>Increased degradation of 25(OH)D</td>
<td>Drugs such as rifampicin, isoniazid, anticonvulsants, glucocorticoids.</td>
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</tbody>
</table>

**Etiology of Vitamin D deficiency** (Misra et al. 2008; Balasubramanian et al. 2013)

**Risk factors for vitamin D deficiency:** Hypovitaminosis D is highly prevalent across all age-groups in developing countries. Consistent risk factors include-

**Gender:** Female sex and particular age groups (neonates, preschool children, pregnant and postmenopausal women) were at the high risk for Vit-D deficiency.

**Skin pigmentation:** Melanin acts as a natural sunscreen and reduces the production of pre-Vit-D in human skin during sunlight exposure. Lo et al showed that Asian Indians and
Pakistanis have similar capacity as white individuals to produce Vit-D in their skin but need longer exposure to produce a similar response. People with a dark skin tone have natural sun protection but require at least a three- to five-fold longer duration of sun exposure to generate the same amount of Vit-D as a person with light skin tone (Holick, 2004).

**Seasonal and latitude variations:** Lower serum 25-hydroxyvitamin D levels were reported in winter and spring compared to summer and fall in studies from temperate regions in the developing countries. Latitude variations also affect Vit-D synthesis in the skin and therefore, low Vit-D levels are expected at high latitudes in the absence of vitamin D supplementation (Holick et al. 2007).

**Nutritional status and low socioeconomic status:** Individuals with hypovitaminosis D were mostly of low socioeconomic status and were reported to have a low daily intake of calcium. Low calcium intake is probably associated with low Vit-D levels rather than being a causative factor. Malnutrition and protein deficiency causes a decrease in the vitamin D binding protein in blood, which diminishes the ability of the body to conserve 25-hydroxyvitamin D (MacLaughlin and Holick, 1985).

**Clothing style and sunscreen use:** A concealing clothing style was a consistent predictor of low Vit-D in India. Women wearing cultural dress customs, sarees (doing Purdah) that limit the skin exposure was also a risk for hypovitaminosis D. Use of sunscreen with high sun protection factor (SPF) avoid UVB to penetrate into the skin and prevent the synthesis of Vit-D (MacLaughlin and Holick, 1985).

**Atmospheric pollution:** Agarwal and colleagues (2002) showed that the higher atmospheric pollution, the lower amount of UVB light reaching ground level. They also
showed that children living in an area with high levels of atmospheric pollution had lower mean serum total 25-hydroxyvitamin D concentrations (31 nmol/l) than those living in similar types of housing but in a less polluted area of Delhi (68 nmol/l). Thus, atmospheric pollution has been suggested to be a cause of reduced Vit-D synthesis in the skin (Agarwal et al. 2002).

**Obesity:** Person with high BMI and fat mass are at major risk for hypovitaminosis D (Konradsen et al. 2008). Obesity alters the release of Vit-D into the circulation and decreases its bioavailability because of the deposition in adipose tissue compartments.

**Symptoms of Vitamin D Deficiency:**

- General Muscle pain and weakness
- Low energy and fatigue
- Symptoms of depression and mood swings
- Sleep irregularities
- Joint pains and Weak bones/fractures
- Poor concentration/Headaches
- Constipation or diarrhea

**Vitamin D and Menopause:**

Vitamin D, also known as the ‘sunshine-hormone’ that offers numerous benefits for women who are experiencing menopause. For women going through the menopause, the lack of natural Vit-D can bring many issues like increased risk of developing CVD, reduced cognitive function and osteoporosis.
Vitamin D is also a great tool to reduce menopausal symptoms like hot flashes and night sweat, and improve your mood and cognitive performance, helping women to stay strong, healthy, and positive. Calcium balance studies have shown that calcium absorption declines with menopause (Heaney et al. 2003). Since 25(OH) D serves the purpose of calcium absorption and appears to be hormonally sensitive, thus hypothesized that Vit-D also may influence vasomotor symptoms, which are clearly related to hormone levels. Also, some of the symptoms associated with Vit-D deficiency, such as mood disturbance and musculoskeletal complaints (Arvold et al. 2009), are similar to symptoms women may experience during the transition through menopause (Brunner et al. 2010). The possible link between Vit-D and menopause-related symptoms is that it can protect against experimental serotonin depletion (Cass et al. 2006) and a menopausal decline in serotonin, a neurotransmitter with known effects on thermoregulation, may be a contributor to hot flashes (Berendsen, 2000; Pinkerton and Zion, 2006; Rossmanith and Ruebberdt, 2009). In addition, estrogen increases the activity of the enzyme responsible for activating Vit-D (Buchanan et al. 1986). Therefore, declining estrogen levels during the menopausal transition could lead to symptoms of Vit-D deficiency. Indeed, vitamin D supplementation can improve mood in non-menopausal populations (Lansdowne and Provost, 1998; Gloth et al. 1999; Jorde et al. 2008; Khan et al. 2010).

Menopause, also known as the climacteric, is a biological process characterized by cessation of the menstrual flow for a period of 12 months in women lives (Shriver, 2015). Menopause is usually a natural change that occurs between 45 to 55 years of age (Takahashi and Johnson, 2015). At the physiological level, menopause happens
because of a decrease production of the hormones estrogen and progesterone from the ovaries (Shriver, 2015). Menopause may be spontaneous (natural menopause) or iatrogenic (secondary menopause).

**Sign and Symptoms of Menopause:**

Menopausal symptoms can be very distressing and considerably affect a woman’s personal and social life, health-care providers caring for women at all levels of the health-care system must be well prepared to guide patients through this transition and provide advice to improve quality of life.

Symptoms of menopause include central nervous system (CNS)-related disorders, bodily alterations related to cardio-metabolic changes, musculoskeletal alterations, urogenital and skin atrophy and sexual dysfunction. CNS-related symptoms such as hot flashes, mood and sleep disorders, migraine, memory and learning difficulties seem to depend on fluctuations in estradiol levels, which causes secondary neurochemical changes. Apart from developing vasomotor symptoms, women transitioning through menopause might also be vulnerable to developing anxiety, depression, insomnia, and migraine. By contrast, physical symptoms such as urogenital atrophy, aging of the skin and skeletal fractures due to postmenopausal osteoporosis are secondary to the decline in estradiol levels (Monteleone et al. 2018). The accumulation and central distribution of body fat, hair loss, the development of facial hair and sexual dysfunction are seemingly due to transitional increases in androgen bioavailability. An emerging concept is that some menopausal symptoms might be predictive of future health complications. Indeed, severe vasomotor symptomatology and poor quality of sleep are associated with an increased risk of CVD.
and postmenopausal depression. Furthermore, depressive symptoms, vasomotor symptoms, and sleep disorders might increase the susceptibility to developing cognitive dysfunction. Severe hot flashes have also been associated with an increased risk of osteoporosis and bone fracture. Finally, as menopause seems to accelerate the aging process, it is conceivable that, in addition to the loss of ovarian function, the manifestation of menopausal symptoms might be in part due to aging (Monteleone et al. 2018).

**Overview of menopausal symptoms (Monteleone et al. 2018)**
**Stages of Menopause:** There are four stages of menopause including-

A. **Premenopause** is the first stage, starts from the first menstrual period. It is referred to a woman's reproductive or fertile life. It is frequently misused to describe the years immediately before menopause (perimenopause) or to describe premature menopause.

B. **Perimenopause** or “menopause transition” is the second stage, refers to the transitional phase of woman reproductive life that precedes menopause, generally happens around 30 to 40 years of age. It usually occurs 3 to 4 years before menopause. During this time, the ovaries gradually stop releasing eggs and producing a decreased amount of estrogen and other hormones (progesterone, androgen, and testosterone). This menopausal transition was associated with the occurrence of hormonal fluctuations and irregular periods. Women who are in this phase may experience breast tenderness, worsening of premenstrual syndrome (PMS), irregular periods or skipping periods.

C. **Menopause** is the third stage, a point when a woman no longer has menstrual periods for 12 consecutive months and become infertile. At this stage, the ovaries have stopped releasing eggs and producing most of their estrogen. This stage is known as the “change of life” or an irreversible ending of a women menstrual periods and ability to bear children.

D. **Postmenopause** is the fourth stage of menopause, begins after 12 months has passed since the last menstrual cycle. At this juncture, a woman may experience an increase in symptoms associated with reduced estrogen production. During menopausal transition hormonal alteration takes place that may contribute to increased abdominal fat deposition influenced by low estrogen hormone. At this stage, postmenopausal women were at high risk of major health concern like osteoporosis, cardiovascular disease, diabetes and metabolic syndrome.
METABOLIC SYNDROME / INSULIN RESISTANCE SYNDROME / SYNDROME “X”

Metabolic syndrome (MS) is a clinical disorder characterized by clustering of cardiovascular risk factors. They include impaired fasting sugar, insulin resistance, dyslipidemia, hypertension and central obesity. Metabolic syndrome is related to systemic alterations that involve several tissues, such as liver, muscle, adipose tissue, stomach and immune system, varying the production of many biomolecules.

Origin of the concept of the metabolic syndrome:

Metabolic Syndrome was born first more as a concept than a diagnosis (Shaw and Chisholm, 2003). In 1923, a Swedish physician firstly established the association of high blood pressure, hyperglycemia, and hyperuricemia (Kylin, 1923). In the late 40’s Vague et al related metabolic dysfunctions found in CVD and T2DM. Twenty years later, Avogaro et al, (1965) described an entity which comprised obesity, hyperglycemia and high blood pressure (BP). More than five decades later in 1988, Gerald Reaven coined the term “Syndrome X” for several risk factors commonly cluster together including glucose intolerance, hypertension, increased level of very-low-density lipoproteins (VLDL-C), increased triglycerides, and decreased high density lipoprotein cholesterol (HDL-C), with insulin resistance being the basic pathophysiological interface. The following year, this entity of obesity, glucose intolerance, hypertriglyceridemia, and hypertension was renamed as “The Deadly Quartet” by Kaplan et al (1989). The greatest contribution of Reaven et al was the inclusion of the concept of insulin resistance. In 1992, a new name proposed and called it as Insulin resistance syndrome (IRS).
Various Organizations Defined Criteria For The Clinical Diagnosis Of The Metabolic Syndrome.

Various organizations have been proposed many definitions in the recent decades. The most important ones are from the World Health Organization (WHO) (Alberti and Zimmet, 1998), the European Group for the study of Insulin Resistance (EGIR) (Balkau and Charles, 1999), the American Association of Clinical Endocrinologists (AACE) (Einhorn et al. 2003) and the International Diabetes Federation (IDF) (Zimmet et al. 2005), the National Cholesterol Education Programme Adult Treatment Panel III (NCEP ATP III) (2005 revised) (Grundy et al. 2005), and NCEP ATP III for Asian Indian (Misra et al. 2009). All of these different classifications are listed in the following table.

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<tr>
<td>IGT, IFG, T2DM, or lowered insulin Sensitivity plus any 2 of the following</td>
<td>Plasma insulin &gt;75th percentile plus any 2 of the following</td>
<td>None, but any 3 of the following 5 features</td>
<td>IGT or IFG plus any of the following based on the clinical judgment</td>
<td>None</td>
<td>None, but any 3 of the following 5 features</td>
<td>None, but any 3 of the following 5 features</td>
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<tr>
<td>Body weight</td>
<td>Men: waist-to-hip ratio &gt;0.90; women: waist-to-hip ratio &gt;0.85 and/or BMI &gt; 30 kg/m</td>
<td>WC ≥94 cm in men or ≥80 cm in women</td>
<td>WC ≥102 cm in men or ≥88 cm in women</td>
<td>BMI ≥ 25 kg/m²</td>
<td>Increased WC (population specific) plus any 2 of the following</td>
<td>WC ≥102 cm in men or ≥88 cm in women</td>
<td>WC ≥102 cm in men or ≥80 cm in women</td>
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<td>Lipids</td>
<td>TG ≥150 mg/dL and/or HDL-C &lt;35 mg/dL in men or &lt;39 mg/dL in women</td>
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<td>TG ≥150 mg/dL and/or HDL-C &lt;40 mg/dL in men or &lt;50 mg/dL in women</td>
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<td>TG ≥150 mg/dL or on TGs Rx. HDL-C &lt;40 mg/dL in men or &lt;50 mg/dL in women or On HDL-C Rx</td>
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<td>Blood pressure</td>
<td>≥140/90 mm Hg or On hypertension Rx</td>
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<tr>
<td>Glucose</td>
<td>IGT, IFG, or T2DM</td>
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<tr>
<td>Glucose</td>
<td>IGT or IFG (but not diabetes)</td>
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<td>Glucose</td>
<td>Fasting glucose 100 mg/dL or Rx</td>
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<td>Other</td>
<td>Microalbuminuria: Urinary excretion rate of &gt;20 mg/min or albumin: creatinine ratio of &gt;30 mg/g.</td>
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<td>Other</td>
<td>Other features of insulin resistance</td>
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<tr>
<td>Other</td>
<td>Fasting glucose 100 mg/dL or Rx</td>
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BMI: body mass index; HDL-C: high-density lipoprotein cholesterol; IFG: impaired fasting glucose; IGT: impaired glucose tolerance; Rx, pharmacologic treatment; TG: triglycerides; T2DM: type 2 diabetes mellitus; WC: waist circumference.
Pathophysiology of Metabolic Syndrome:

Metabolic syndrome is a complex web of metabolic factors that are associated with two-fold risk of CVD and a five-fold risk of diabetes. Individuals with MS have a 30-40% probability of developing diabetes and CVD within 20 years, depending on the number of components present (Mohanan, 2016). The metabolic syndrome has been assigned its own ICD-9 diagnostic code: 277.7. Despite this, there is an ongoing controversy about whether metabolic syndrome is a homogeneous disorder or disease, and whether it merits recognition as a syndrome (Meigs, 2004; Grundy et al. 2005; Kahn et al. 2005; Reaven, 2006; Grundy, 2007). It refers to the co-occurrence of four central known cardiovascular risk factors, including abdominal obesity, insulin resistance, atherogenic dyslipidemia, and hypertension. All these conditions are interrelated and share underlying mediators, mechanism, and pathways. Instead of the four central features described, some other factors including aging, proinflammatory states, and prothrombotic states have been implicated as contributing factors for the pathophysiology of MS (Grundy et al. 2004; Huang, 2009).

A. Abdominal obesity/ visceral adiposity:

Visceral adiposity is the most important factors and is principally driven by an increased consumption of cheap, calorie dense food and reduced physical activity. With obesity and progressive adipocytes enlargement, the blood supply to adipocytes may be reduced with consequent hypoxia (Cinti et al. 2005). Hypoxia has been proposed to be an inciting etiology of necrosis and macrophage infiltration into adipose tissue that leads to an overproduction of biologically active metabolites known as adipokines which includes glycerol, free fatty acids (FFA), proinflammatory mediators- TNF-α and IL-6, plasminogen activator inhibitor-1 (PAI-1), and C-reactive protein (CRP) (Lau et al. 2005). This results
in a localized inflammation in adipose tissue that propagates an overall systemic inflammation associated with the development of obesity-related comorbidities (Trayhurn and Wood, 2004).

**Pathophysiology of MS: Visceral Adiposity (Source: Huang, 2009)**

**B. Hyperglycemia/Insulin Resistance:**

Insulin resistance is defined as a pathophysiological condition occurs when there is a decrease in the responsiveness of peripheral tissues (skeletal muscle, adipose tissue, liver) to the effects of insulin (Kim et al. 2006; Huang, 2009). Insulin signaling occurs following the binding of insulin to the insulin receptor, a ligand-activated tyrosine kinase via phosphorylation of downstream substrates and activation of two parallel pathways: the phosphoinositide 3-kinase (PI3K) pathway and the mitogen-activated protein (MAP) kinase pathway. Tyrosine phosphorylation of insulin receptor substrates (IRS) activates PI3K, leading to activation of the 3-phosphoinositide-dependent protein kinase 1 (PDK1)
kinase and Akt kinase. In vascular endothelial cells, Akt kinase phosphorylates and activates endothelial nitric oxide synthase (eNOS). In skeletal muscle and adipose tissue, Akt kinase stimulates translocation of the insulin-responsive glucose transporter GLUT4 to the cell surface, leading to increased glucose uptake. In insulin resistance, the PI3K-Akt pathway is affected, while, the MAP kinase pathway functions normally during insulin resistance condition. This leads to a change in the balance between these two parallel pathways. Inhibition of the PI3K-Akt pathway leads to a reduction in endothelial NO production, resulting in an endothelial dysfunction, and a reduction in GLUT4 translocation, leading to a decreased skeletal muscle and fat glucose uptake. In these ways, an insulin resistance leads to the vascular abnormalities that predispose to hyperglycemia, advanced glycation products, toxicity from FFA, obesity, dyslipidemia, and other proinflammatory conditions (Kim et al. 2006; Jonk et al. 2007; Huang, 2009).

Pathophysiology of MS: Insulin Resistance (Source: Huang, 2009)
C. Atherogenic Dyslipidemia:

The dyslipidemia is characterized by an elevated serum TG levels, low levels of HDL-C, elevation of lipoproteins containing apolipoprotein-B (apo-B), and increased levels of small particles of LDL-C. Insulin resistance leads to atherogenic dyslipidemia in several ways:

- First, insulin normally suppresses lipolysis in adipocytes, so an impaired insulin signaling increases lipolysis, resulting in increased FFA levels. In the liver, FFA serves as a substrate for the synthesis of TG. FFA also stabilize the production of apo-B, the major lipoprotein of VLDL-C particles, resulting in a more VLDL-C production.

- Second, insulin normally degrades apo-B through PI3K-dependent pathways, so an insulin resistance directly increases VLDL-C production.

- Third, insulin regulates the activity of lipoprotein lipase, the rate-limiting and major mediator of VLDL-C clearance.

Thus, hypertriglyceridemia in insulin resistance is the result of both an increase in VLDL-C production and a decrease in VLDL-C clearance. VLDL-C is metabolized to remnant lipoproteins and small dense LDL-C, both of which can promote an atheroma formation. The triglycerides in VLDL-C is transferred to HDL-C by the cholesterol ester transport protein (CETP) in exchange for cholesteryl esters, resulting in the TG-enriched HDL and cholesteryl ester enriched VLDL particles. Further, the TG-enriched HDL is a better substrate for hepatic lipase, so it is cleared rapidly from the circulation, leaving fewer HDL particles to participate in a reverse cholesterol transport from the vasculature (Semenkovich, 2006; Huang, 2009).
Pathophysiology of MS: Atherogenic Dyslipidemia (Source: Huang, 2009)

D. **Hypertension:**

Hypertension is frequently associated with the several metabolic abnormalities, in which obesity, insulin resistance, and dyslipidemia are the most common (Ferrannini and Natali, 1991). Both hyperglycemia and hyperinsulinemia activate the Renin-angiotensin-aldosterone system (RAAS) by increasing the expression of angiotensinogen, angiotensin II, and the AT1 receptor, which, in concert, may contribute to the development of hypertension in patients with insulin resistance (Malhotra et al. 2001). There is also evidence that insulin resistance and hyperinsulinemia lead to sympathetic nervous system (SNS) activation, and, as a result, the kidneys increase sodium reabsorption, the heart increases cardiac output, and arteries respond with vasoconstriction resulting in hypertension (Morse et al. 2005). It has been recently discovered that adipocytes also
produce aldosterone in response to angiotensin II (Briones et al. 2012). In this regard, the adipocyte may be considered a “Miniature RAAS”.

E. Endothelial dysfunction:

It occurs when the endothelium fails to serve its normal physiological and protective mechanisms. There are several mechanisms for endothelial dysfunction (Huang, 2005). The most important ones are a reduction in eNOS phosphorylation at S1177 (Dimmeler et al. 1999; Fulton et al. 1999) and the rapid reaction of NO with superoxide to form peroxynitrite anion (Beckman and Koppenol, 1996). In addition, asymmetric dimethylarginine may compete with arginine to reduce endothelial NO production. eNOS requires enzymatic cofactors, including flavin adenine dinucleotide (FAD$^+$), flavin mononucleotide (FMN), NADPH and tetrahydrobiopterin (BH4). In the absence of BH4, electron transport through eNOS can become ‘uncoupled’, resulting in the
generation of superoxide by eNOS. Superoxide, whether formed by NADPH oxidase or by uncoupled eNOS, reacts with NO in an extremely rapid, diffusion-limited reaction to form peroxynitrite anion, which has its own toxic effects. Insulin resistance and visceral adiposity also cause endothelial dysfunction by decreasing Akt kinase activity and through the effects of resistin, IL-6 and TNFα on eNOS phosphorylation activity (Huang, 2005; Atochin et al. 2007).

Pathophysiology of MS: Endothelial Dysfunction (Source: Huang, 2009)

Other Contributing Factors: Advancing age affects all levels of pathogenesis which explains why the prevalence of MS increases with advancing age (Ford et al. 2002). Recently proinflammatory and prothrombotic states recognized clinically by elevation of CRP and PAI-1 were also associated with the metabolic syndrome as they may be metabolically interconnected.
**Vitamin D deficiency and risk of metabolic syndrome during menopause**

Vitamin D deficiency is becoming a lifestyle problem amongst all age-group of peoples especially in postmenopausal women (Tandon et al. 2014). Predominantly menopause is a phenomenon of aging in women and also age is one of the crucial factor in determining the cutaneous synthesis of vitamin D (Chang et al. 2000). Thus menopausal transition was associated with the gradual reduction in the levels of vitamin D as well as estrogen hormone. Estrogen hormone increases the activity of 1 α-hydroxylase responsible for the conversion of 25(OH) D to the biologically active 1, 25(OH)2 D (Buchanan et al. 1986, Cheema et al. 1989). Vitamin D also plays an important role in anti-inflammatory and immune-modulating property (Guillot et al. 2010). There is an inverse link between inflammation and immune-modulating effect of vitamin D which draw more attention toward MS (Vita et al. 2014). Metabolic syndrome may not be a single disease entity, but rather a constellation of closely related risk factors. The prevalence of the metabolic syndrome is known to increases with menopause (Carr, 2003). Vitamin D deficiency also plays a key role in the pathophysiology of risk factors of metabolic syndrome which affect the cardiovascular system, increases insulin resistance and obesity, stimulate RAAS that cause hypertension. The discovery of vitamin D receptor (VDR) expressed ubiquitously in almost all body cells that suggest an involvement of vitamin D mediated effects on metabolic syndrome (Prasad and Kochhar, 2015). Thus the postmenopausal women were more susceptible to having vitamin D deficiency, are known to be at high risk of having metabolic syndrome, CVD, and T2DM.
The relationship between hypovitaminosis D and the components of metabolic syndrome. (Source: Minambres et al. 2015)

A. Vitamin D deficiency and Abdominal Obesity during menopause:

Menopause is associated with weight gain and altered body fat distribution. Body fat is distributed into two patterns, the accumulation of fat centrally, as intra-abdominal fat (apple shape) and, the accumulation of fat in the gluteo-femoral region (pear shape). The accumulation of fat in a central distribution (intra-abdominal) has emerged as a risk factor for CVD (Kannel et al. 1991). Estrogen promotes the accumulation of gluteo-femoral fat, and the loss of estrogen with menopause is associated with an increase in central fat (Krotkiewski et al. 1983; Poehlman et al. 1995). Although, some cross-sectional and longitudinal studies have believed that menopausal transition was associated with abdominal adiposity, independent of the effect of age and total body adiposity (Zamboni
et al. 1992, Poehlman et al. 1995). In postmenopausal women, low vitamin D status is associated with greater bone turnover, bone loss, and obesity (Macdonald et al. 2008). 25(OH) D concentrations are associated with body composition variables, especially body fat, independently of seasonal variability (Parikh et al. 2004). The link between hypovitaminosis D and obesity has been reported when obesity is defined by using both BMI and waist circumference (Scragg et al. 2004). The metabolic and endocrine status in obese women is quite complex and under the influence of different fat hormones (Lobo, 2008; Hroussalas et al. 2008).

**Effect of 1,25(OH)$_2$D$_3$ on adipose tissue/ lean and obese state**

(Source: Mutt et al. 2014)
In obesity, adipose tissue undergoes hypertrophic enlargement, which results in an imbalanced blood flow leading to hypoxia, inflammation and macrophage infiltration (Goossens, 2008; Trayhurn, 2013). 1, 25(OH)\textsubscript{2}D\textsubscript{3} acts at several levels to modulate the function of the immune system (Lemire, 2000). Recent evidence focused on the involvement of 1, 25(OH)\textsubscript{2}D\textsubscript{3} in the regulation of adipose tissue inflammation by reducing the proinflammatory cytokines secreted from adipose tissue. In differentiated adipocytes from human subcutaneous white adipose tissue 1, 25(OH)\textsubscript{2}D\textsubscript{3} attenuates TNF-\textgreek{a} induced MCP-1 secretion, while it inhibited secretion of adiponectin without affecting its mRNA levels (Lorente-Cebrian et al, 2012). In human subcutaneous adipose tissue fragments 1, 25(OH)\textsubscript{2}D\textsubscript{3} reduced interleukin-1 beta (IL-1\textbeta) induced expression of the inflammatory genes MCP-1, IL-6 and IL-8.

Signal transduction of inflammatory pathways in adipose tissue involves activation of NF-\textkappaB and translocation of p65 to nucleus mediated by degradation of I\textkappaB\textalpha (Tourniaire et al, 2013). Mutt et al (2012) have demonstrated that 1, 25(OH)\textsubscript{2}D\textsubscript{3} suppressed lipopolysaccharide (LPS) stimulated IL-6 secretion in human isolated mature and MSC differentiated adipocytes. 1, 25(OH)\textsubscript{2}D\textsubscript{3} inhibits the inflammatory markers in adipocytes via the involvement of Nuclear factor kappa-B (NF\textkappaB) or p38 mitogen-activated protein kinase (P38MAPK) classical inflammatory pathway (Marcotorchino et al. 2012; Gao et al. 2013; Ding et al. 2013. Thus, the presence of 1, 25(OH)\textsubscript{2}D\textsubscript{3} inhibited chemokine and cytokine secretion in human adipocytes. 1, 25(OH)\textsubscript{2}D\textsubscript{3} strongly inhibited the activation of the NF-\textkappaB and MAPK signaling pathways, which prevent gene transcription of the proinflammatory factors.
The above figure showed the molecular actions of 1, 25(OH)\(_2\)D\(_3\) in inflammation and energy homeostasis in adipocytes. Stimulation via e.g., (LPS), TNF-\(\alpha\) via specific receptors e.g. Toll-like receptor (TLR), IL-6 receptors(IL-6R) activate NF\(\kappa\)B or P38MAPK signaling dependent transcription of inflammatory genes such as IL-6, TNF-\(\alpha\), and IL-1\(\beta\). 1, 25(OH)\(_2\)D\(_3\) inhibits inflammation by inhibiting Inhibitor kappa-B (I\(\kappa\)B\(\alpha\)) phosphorylation and translocation of NF\(\kappa\)B as well P38MAPK into the nucleus. Furthermore, 1, 25(OH)\(_2\)D\(_3\) affects energy homeostasis through uncoupling proteins (Source: Mutt et al. 2014).

**B. Vitamin D deficiency and Hyperglycemia/ Insulin Resistance during menopause:**

Two of the most important pathophysiological components of the metabolic syndrome are increased visceral fat accumulation and insulin resistance. Insulin resistance, with inadequate compensatory hyperinsulinemia, diminishes the normal suppression of FFA arising from adipose tissue by insulin. The increased levels of FFA may impair peripheral
glucose uptake, increase hepatic gluconeogenesis, and reduce hepatic clearance of insulin (Despres, 1993). Several studies have shown increased fasting insulin and increased fasting glucose levels in postmenopausal compared with premenopausal women, which would imply worsened insulin resistance with the menopause (Razay et al. 1992; Dallongevielle et al. 1995; Poehlman et al. 1997, Lynch and Adams, 2014).

**Effects of estrogen hormone on various organs to influences the glucose metabolism**

Intake of calcium and vitamin D are related to the metabolic syndrome in 10,066 middle-aged or older women from the Women’s Health Study who are free of CVD, cancer, or diabetes and who never used hormonal therapy (Liu et al. 2005). Dietary vitamin D was inversely associated with the prevalence of metabolic syndrome when considering with calcium intake. The vitamin D endocrine system plays an important role in the glucose homeostasis and insulin release mechanism. Vitamin D enhances insulin secretion by interacting with 1, 25(OH)_{2} D_{3}-RXR-VDR complex which binds to vitamin D responsive elements (VDRE) found in the insulin gene promoter region, to enhance the transcriptional activation of the insulin gene and increases insulin synthesis (Wei et al. 2008). It acts as a

**Mechanism of action of 1, 25(OH)$_2$D$_3$ in human-monoecyte-derived Dendritic Cells.**

(Source: Ferreira et al. 2015)

The VDR-bound 1, 25(OH)$_2$D$_3$ activates the PI3K-Akt-mTOR pathway via either forming a complex and phosphorylating the regulatory subunit of PI3K, or by other unknown mechanisms. This releases and activates the catalytic subunit, which unleashes the PI3K downstream pathway. Among other functions, activation of this pathway promotes the expression of different key glycolytic enzymes, which induces glycolysis. Control of surface marker expression and cytokine production by 1, 25(OH)$_2$D$_3$ might arise from its
impact on the PI3K pathway, which can control essential transcription factors (e.g. GSK-3b and NF-kB nuclear translocation) or from the direct regulation of transcription factors, key metabolic bi-functional enzymes, and RNA binding proteins (Ferreira et al. 2015).

![Diagram of Vitamin D and β cell function/insulin secretion]

**Vitamin D and β cell function/insulin secretion**

(Source: Harinarayan, 2014)

C. **Vitamin D deficiency and atherogenic dyslipidemia during menopause:**

Alterations in lipid metabolism with estrogen deficiency are thought to be a substantial component of CVD risk in postmenopausal women (Kannel and Wilson, 1995), but there are also direct effects of estrogen deficiency on body fat distribution (central obesity), insulin action, the arterial wall, and fibrinolysis that may influence cardiovascular risk..

Altered lipid profile due to Estrogen deficiency could lead to increase the prevalence of the metabolic syndrome in postmenopausal women compared with premenopausal women.
Metabolism (Park et al. 2003), and this postmenopausal worsening of the metabolic profile may contribute to the future risk of CVD. A menopausal transition leads to low level of estrogen that influences the manifestation of the metabolic syndrome. High amounts of intra-abdominal fat (IAF) was associated with increased insulin resistance (IR) and free fatty acid (FFA) levels and decreased adiponectin. These factors contribute to increased secretion of the apo B-containing protein, leading to hypertriglyceridemia and increased HL activity, which leads to a predominance of small dense LDL particles and a reduction in the large anti-atherogenic HDL\textsubscript{2} particles (Carr, 2003).

Many longitudinal studies have shown that the prospective transition to postmenopausal period was associated with a 16\% increase in TG and 25\% decrease in HDL (Jensen et al. 1990, Poehlman et al. 1995, Do et al. 2000, Matthews et al. 2001). Menopausal changes in HDL metabolism are more complex because antiatherogenic HDL\textsubscript{2} levels decrease while HDL\textsubscript{3} levels increase (Stevenson et al, 1993; Li et al. 1996; Kuller et al. 1997; Carr and Brunzell, 2003). HDL\textsubscript{2} particles are the large, buoyant, and more cardioprotective subspecies of total HDL. Endogenous estrogen levels are inversely associated with HL activity (Tikkanen et al. 1986). Hepatic lipase hydrolyzes the TG and phospholipid in LDL and HDL and is one factor that determines the size and density of LDL and HDL particles (Santamarina-Fojo et al. 1998). The higher the HL activity, the more TG, and phospholipid hydrolyzed, resulting in smaller, denser more atherogenic lipoprotein particles. Lipoprotein lipase hydrolyzes TG in triglyceride-rich lipoproteins, generating FFA that can serve as an energy source or can be stored in adipocytes. CETP catalyzes the exchange of cholesterol ester in HDL and LDL particles for TG in VLDL, and high CETP concentrations are
associated with reduced HDL levels. Menopausal status does not appear to affect CETP activity (Lewis-Barned et al. 1999).

Hypovitaminosis D has been reported to be associated not only with lowered insulin secretion and sensitivity but also with adverse effects on both total cholesterol and LDL-C concentrations. The Woman’s Health Initiative calcium/vitamin D randomized controlled trial found a 4.46 mg/dl decrease in LDL-C after daily supplementation with 1000 mg of calcium and 400 IU of vitamin D3 (Schnatz et al. 2014). Some study reported that the plasma vitamin D concentration was positively associated with HDL-C, concluding that lower vitamin D would be associated with a more atherogenic lipid profile, which is a major risk factor for progression toward coronary artery atherosclerosis (Schnatz et al. 2011). In a process called reverse cholesterol transport, large HDL particles are known to carry cholesterol from atherosclerotic plaques (Rye et al. 2009), and these large HDL particles are driven from cholesterol-loaded macrophages by cholesterol efflux.

**D. Vitamin D deficiency and Hypertension during menopause:**

Vitamin D is a negative endocrine regulator of the RAAS. Increased activation of the RAAS, which is the main regulator of electrolyte and volume homeostasis, contributes to
the development of arterial hypertension (Connell et al. 2008). Renin is mainly synthesized by the juxtaglomerular cells of the kidney and stimulates the production of angiotensin II and of aldosterone, which increases blood pressure directly by vasoconstriction and indirectly by salt and water retention and other mechanisms (Connell et al. 2008). Inappropriate, increased activation of the RAAS has been reported in VDR and 1α-hydroxylase enzyme activity (Li et al. 2002; Xiang et al. 2005; Simpson et al. 2007; Zhou et al. 2008). Vitamin D receptors are distributed on vascular smooth muscle, endothelium, cardiomyocytes, and activated 1, 25(OH)_2 D that suppresses renin gene expression, regulating the growth and proliferation of vascular smooth muscle cells, cardiomyocytes, and inhibiting cytokine release from lymphocytes. Therefore, the absence of vitamin D receptor activation leads to tonic up-regulation of the renin-angiotensin system, eventually leading to hypertension and left ventricular hypertrophy (Wang et al. 2008).

Importantly, VDR and 1α-hydroxylase developed arterial hypertension and myocardial hypertrophy, which were present even after normalization of calcium homeostasis; however, blocking of the RAAS system with angiotensin-converting -enzyme inhibitors normalized blood pressure and cardiac abnormalities (Li et al. 2002; Xiang et al. 2005; Simpson et al. 2007; Zhou et al. 2008). Furthermore, increased RAAS activation, arterial hypertension, and myocardial abnormalities could be successfully treated with 1, 25(OH)_2 D in 1α-hydroxylase knockout mice (Zhou et al. 2008). The molecular effects of vitamin D on the RAAS have been clarified by the finding that liganded VDR suppresses renin expression by binding to the transcription factor cAMP response element binding protein (CREB) (Yuan et al. 2007). As a result, stimulation of renin transcription is inhibited because CREB is no longer able to stimulate renin transcription by binding to cAMP.
response elements in the promoter region of the renin gene (Yuan et al. 2007). In patients with arterial hypertension, renin activity has been inversely associated with 1, 25(OH)₂ D levels (Resnik et al. 1986; Burgess et al. 1990). Importantly, decreased renin and angiotensin II levels were observed in several, but not all, studies that examined the activity of the RAAS after treatment with vitamin D, 1, 25(OH)₂ D or active vitamin D analogs (Resnick et al. 1984; Lind et al. 1989; Kimura et al. 1999; Park et al. 1999; Sugden et al. 2008; Freundlich et al. 2008).

**A possible mechanism of vitamin D deficiency and development of hypertension, obesity, and insulin resistance (Source: Grober and Kisters, 2012).**
**Vitamin D and Inflammation:**

Recently, vitamin D plays an important role in the modulation of the immune/inflammation system via regulating the production of inflammatory cytokines and inhibiting the proliferation of proinflammatory cells (Yin and Agarwal, 2014). Vitamin D regulates the immune system via the vitamin D receptor (VDR) which is present in most immune cells types, particularly in antigen-presenting cells (APCs) such as monocytes, macrophages and dendritic cells (White, 2008). Vitamin D receptor (VDR), a member of the superfamily of nuclear receptors and functions as a ligand-activated transcription factor (Nagy et al. 2012). It is now well recognized that CYP27B1 and VDR are expressed in cells involved in the immune/inflammation system in the human body (Brennan et al. 1987), which provides the biological basis for the role of vitamin D in low-grade inflammatory diseases. The increase of CYP27B1 results in the accumulation of 1, 25(OH)\_2 D, which further binds and activates the vitamin D receptor (VDR), leading to the target gene transcription via vitamin D response elements located in the regulatory regions of 1, 25(OH)\_2 D target genes (Korf et al. 2012). Chen et al found that 1,25(OH)\_2 D\_3 can regulate TLR signaling via stimulating SOCS1 by downregulating miR-155 in macrophages, which provide a novel negative feedback regulatory mechanism for vitamin D to control innate immunity (Chen et al. 2013). In a recent study, both forms of vitamin D- 1, 25(OH)\_2 D and 25(OH) D dose-dependently inhibited lipopolysaccharide-induced p38 phosphorylation, IL-6, and TNF\_\alpha production by human monocytes via histone H4 in an acetylation-dependent manner (Zhang et al. 2012). Moreover, 1,25(OH)\_2 D or its analogs have been shown to initiate the differentiation of myeloid progenitors into macrophages (Ohta et al. 1985), and to reduce
MCP-1 and IL-6 expression via inhibiting the activation of nuclear factor-κB (NF-κB) in macrophages (Sanchez-Nimo et al. 2012).

Vitamin D metabolites and immune modulation: endocrine, paracrine, and autocrine responses (Source: Norman and Powell, 2014)

In addition, vitamin D induces an anti-atherogenic monocyte/macrophage phenotype via regulating endoplasmic reticulum stress (Riek et al. 2012) and can selectively suppress key effector functions of interferon (IFN)-γ-activated macrophages (Helming et al. 2005). Vitamin D deficiency causes increased proinflammatory cytokine expression in epicardial adipose tissue, which is coupled with increased inflammatory cellular infiltrate, suggesting the anti-inflammation effect of vitamin D in epicardial adipose tissue is a novel mechanism for athero-protection (Gupta et al. 2012). Most verified mechanisms of the anti-atherogenic
effect of vitamin D are regulated by an inflammatory response. First, vitamin D exerts protective effects against endothelial dysfunction, an inflammatory process that precedes atherosclerosis, via multiple mechanisms, including stimulating nitric oxide production and inhibiting oxidative stress (Wong et al. 2010; Kassi et al. 2013). Vitamin D has been found to inhibit contractions, which were endothelium-dependent through inhibiting cyclooxygenase-1 expression and reactive oxygen species production (Wong et al. 2010; Kassi et al. 2013). In addition, calcitriol significantly repressed the expression of cyclooxygenase 2 and promoted prostaglandin catabolism, both of which reduce the level of prostaglandins and suppress proinflammatory cytokine expression in endotheliocytes (Krishnan et al. 2009). Second, 1, 25(OH)$_2$D may alter macrophage function and gene expression, which is crucial in the formation of foam cells and vascular inflammation response that promote the process of atherosclerosis (Riek et al. 2012). In patients with type 2 diabetes mellitus, 1, 25(OH)$_2$D can inhibit the foam-cell formation and suppresses macrophage cholesterol uptake via reducing peroxisome proliferated-activated receptor-$\gamma$-dependent CD36 expression (Takeda et al. 2010). Third, 1,25(OH)$_2$D inhibits the proliferation of vascular SMCs (VSMCs) and exerts protective effects against VSMC morphological changes, which further inhibit the secretion of inflammatory molecules (Carthy et al. 1989; Tukaj et al. 2012).

Interestingly, in the presence of 1,25(OH)$_2$D, VDR has also been found to repress gene transcription via displacing the deoxyribonucleic acid-bound nuclear factor of activated T-cells, thus repressing inflammatory cytokine expression (White, 2012). Epidemiological studies suggest an inverse association between circulating levels of 25-hydroxyvitamin D and inflammatory markers, including CRP and interleukin IL-6 (Hopkins et al. 2011).
Supplemental vitamin D and calcium have been found to decrease the biomarkers of inflammation (Hopkins et al. 2011; Bjorkhem-Bergman et al. 2013). Thus, an effective immune response is heavily dependent on the vitamin D endocrine system which performs a balancing act of inflammation versus anti-inflammation. Recent data from the Women’s Health Initiative revealed that higher baseline CRP and IL-6 levels predicted cardiovascular outcomes in apparently healthy older women (Pradhan et al. 2002). Several studies have shown higher IL-6 levels in postmenopausal compared with premenopausal women (Pfeilschifter et al. 2002).

Schematic representation of the primary mechanisms through which vitamin D regulates macrophage-mediated innate immune response.
(Source: Yin and Agarwal, 2014).