www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2022; SP-11(6): 2715-2724

www.thepharmajournal.com
Received: 12-03-2022
Accepted: 15-04-2022

Amisha Nakra
School of Agriculture,
Lovely Professional University,
Phagwara, Punjab, India

Pratik Dighe
School of Agriculture,
Lovely Professional University,
Phagwara, Punjab, India

Shruti Singhdeo
School of Agriculture,
Lovely Professional University,
Phagwara, Punjab, India

Corresponding Author
Amisha Nakra
School of Agriculture,
Lovely Professional University,
Phagwara, Punjab, India

Bioavailability of vitamin D and its health promoting activity: A review

Amisha Nakra, Pratik Dighe and Shruti Singhdeo

Abstract
Vitamin D is a fat-soluble vitamin which is mainly required to maintain calcium and phosphorus levels in the body. Although many animal and plant-based sources are known now, still the main source of Vitamin D is sunlight. Vitamin D is present in two forms Vitamin D2 and Vitamin D3 in the body and their absorption and metabolism in the body are found to be slightly different. Vitamin D is also seen to be in synergic relation with many elements which can enhance the bioavailability of Vitamin D and help in improving bone density, cardiovascular health, osteoporosis and overall it maintains adaptable & innate immunity of the body but in contrast, many factors are responsible for hindering the bioavailability of Vitamin D such as antagonistic minerals, skin pigments, drugs, processing of food etc. Different technologies and processes have been developed to enhance the bioavailability of Vitamin D such as UV radiating the food which can enhance the vitamin D content in the food products just like in humans when UV radiation falls on foods products like mushrooms in the range of 290 –330 nm the outer layer produces 7-dehydrocholesterol as it is the precursor of Vitamin and nano-encapsulation improves the solubility of Vitamin D and provides acidic, thermal and enzymatic stability.

Keywords: Vitamin D, bioavailability, vitamin D2, vitamin D3, sources, synergistic, enhancement, nano-encapsulation

1. Introduction
Vitamin D is one of the major fat-soluble vitamins which is involved in calcium metabolism thus it plays a vital role in bone formation and resorption. The active form of vitamin D is 1, 25-dihydroxy vitamin D3. Apart from animal sources of Vitamin D, sun exposure is the main source of vitamin D_3. Presently, vitamin D_3 deficiency has become one of the most common deficiencies because of less exposure of the body to the sun due to a sedentary lifestyle, 95% coverage of the skin and also variable sun hours and other factors responsible for vitamin D deficiency may be variable and limited sources of vitamin D in the diet. The richest source of vitamin D is fatty fish which is not consumed aa s staple food and the o vegetarian population is restricted to consume it (Dary, O., & Hurrell, R. 2016) Most circulating 25-hydroxyvitamin D is produced by the skin in human from exposure to sunlight; however, seasonal changes, living at high latitudes, dark skin pigmentation, ageing, and other factors can hinder this process, necessitating periodic dependency on dietary sources to supply the needed precursor to 25-hydroxyvitamin D. The sources of vitamin D, including vitamin $D_{,2}$ is obtained from plants and fungi whereas vitamin D_3 is obtained from animal sources, fortified foods, and supplements. Low concentrations of 25-hydroxyvitamin have increased the risks of chronic diseases, including diabetes mellitus, cancer, autoimmune disorders, and osteoporosis. (Babu, U. S., & Calvo, M. S. (2011). Natural foods rich in vitamin D are not frequently consumed by most of the population. Also, concentrations of vitamin D in foods are variable. Mushrooms and egg yolks are listed as rich sources of vitamin D, but the concentrations are often very low and variable, which res results poor concentration of serum 25-hydroxyvitamin D of the vitamin D content in these foods.

2. Sources of vitamin D2 and D3
Vitamin D is mainly classified into two forms Vitamin D_2 (ergocalciferol) and Vitamin D_3 (cholecalciferol) Vitamin D2 is mainly known to be present in mothest abundant form mushroom shares Vitamin D3 is found in animal sources and is also significantly produced by Human Skin on being exposed to radia radiation sunlight. (Wilson, *et al*., 2017). The forms of vitamin D differ in many aspects such as structure, bioavailability, sources, side chains, absorption in the body etc. Mostly VitaD-rich rich richrce known till now is fatty fit fish

veterinarian Population is mainly known to be dependent the ous synthesis of Vitamin D by UV radiations falling in the range of 290 –330 nm producing 7-dehydrocholesterol as it is pre the cursor of Vitamin D3. (Adamec, *et al*., 2011) [2]. The retention for all the raw Vitamin D rich foods is found to be 100%. Till date meat, mushroom, orange juice, milk and fortified cheese is considered to contain bioavailable vitamin D. Vitamin D2 present in mushrooms (*Agaricus bisporus or Lentinula edodes*) after UV irradiation treatment is significantly bioavailable. The pig meat contains 60% more significant amount of Vitamin D2 rather than Vitamin D2 supplements. On comparing the Vitamin D2 supplementation of 14 µg with dietary Vitamin D2 present in Mushroom it was found that the Serum 25(OH) D concentration increase at same levels, furthermore the serum levels were comparatively higher than the group without supplementation. The UVB radiation technique used to enhance the Vitamin D2 level is approved by European Union (EU) under the novel food Regulation (EC) No. 258/97 in order to the Vitamin D2 level of <10µg/100g for sale of Button Mushrooms (*Agaricus bisporus*) in Europe. Vitamin D2 concentration in food matter of interest because Vitamin D2 is not found in any other animal source till now. (Tripkovic, *et al*., 2012) [70]. The formulation of Vitamin D matrix also plays a significant role on comparing oil, ethanol and lactose encapsulated, it was observed that lactose encapsulated vitamin D is has the highest bioavailability. The fortified bread prepared with vitamin D2-rich yeast was found to increase the serum level up to 98 nmol/L whereas Rye bread fortified with Vitamin D3 increased serum level upto 16.3 nmol/L in healthy adult women. Both 1000 IU of Vitamin D2 and Vitamin D3 obtain in orange juice and Vitamin D supplements were equally efficient in increasing Serum 25(OH) D. Vitamin D3 was found to be more suitable for fortification of eggs as Vitamin D2 fortified feed in birds consistently showed the lower Serum 25(OH) D levels. (Hohman, *et al*., 2011) [25-26]. Vitamin D2 concentration in decline after 28 wks in case of egg fortification indicating the age related metabolic clearance of Vitamin D2 but still Vitamin D3 is considered relatively much reliable because of its consistent concentrations in long run. Consuming one Vitamin D fortified per day enriches the body with 1.9 to 2.8µg which is 2-3 times more than that of average egg. As mentioned above the retention for raw Vitamin D sources is 100% however the cooking and processing of these sources like boiling, frying, and baking, as well as whether the addition of lemon juice can alter its retention. (Ložnjak, *et al*., 2018). Some Vitamin D rich sources have been shown in Table 1.

Table 1: Sources of Vitamin D and their Vitamin D content

S. No.	Source	Form of Vitamin D	Amount	Reference
Sources of both Vitamin D3				
1.	Fish Oil Capsule	Vitamin D3	50 µg/ 840 mg	Manson *et al*., (2020) [49].
2.	Fortified Egg Yolk	Vitamin D3	25.3 to 33.7 µg/100 g	Mattila *et al*. (2014).
3.	Salmon	Vitamin D3	12.4 µg/100 g	Lamberg-Allardt, *et al*. (2016).
4.	Fortified Orange Juice	Vitamin D3	1.663 µg/100 g	Byrdwell, *et al*., (2011) [10].
Sources of both Vitamin D2 and D3				
5.	Fortified margarine	Vitamin D2 and Vitamin D3	530 IU/100 gm	Lips *et al*., (2014) [44]
6.	Cod liver oil	Vitamin D2/ vitamin D3	250 µg/ 100g	Moulas, *et al*., (2018) [53].
7.	Reindeer lichen	Vitamin D2/ vitamin D3	87 µg/100 g	Baur, *et al*., (2016) [5].
8.	Plant oils Wheat germ oil Avocado oil Sunflower oil	Vitamin D2	22.1–34.2 µg/g 4.2–23.4 µg/g 7.9–17.4 µg/g	Byrdwell, *et al*., (2011) [10].
9.	Herring	Vitamin D2 and Vitamin D3	20 µg/ Herring	Lamberg-Allardt, *et al*. (2016).
10.	Fortified Yogurt	Vitamin D2/ vitamin D3	10 ug/ 100g	Manson *et al*., (2020) [49].
11.	Fortified Milk	Vitamin D2 and Vitamin D3	10 µg/ 1000 ml	Lips *et al*., (2014) [44]
Sources of Vitamin D2				
12.	UV Radiated Mushroom *Agaricus bisporus* *Pleurotus ostreatus* *Lentinula edodes*	Vitamin D2	<10µg/100g 1340 µg/100 g 6900 µg/100g 5390 µg/100g	Kučan *et al*, (2018) [37]
13.	Chocolates Dark Chocolate White Chocolate Chocolate Nut	Vitamin D2	1.90-5.48 µg/100 g 0.19-1.91 µg/100 g 0.15 µg/100 g	Taofiq, *et al*. (2017) [68].
14.	UV radiated yeast in Bread	Vitamin D2	2.25 µg /100 g,	Hohman *et al*.

3. Bioavailability Vitamin D3 and Vitamin D2

3.1 Mechanism of Vitamin D3 vs. D2 metabolism

Although the pathway followed by Vitamin D3 and Vitamin D2 is same still there are many differences in their efficiency of being metabolized in the body. There are no evidence based studies explaining why vitamin D2 is less bioavailable, however many studies suggest that it may be due to the fact that Vitamin D2 comprises of an additional double bond at carbon 23 and a methyl group at carbon 24 which shows the factor that Vitamin D3 may have different potency than that of Vitamin D2. (Jones *et al*., 2012). The metabolism of Vitamin D is completed in three steps as follow: first of all Vitamin D when enters the blood stream is not available for body therefore it needs to be transferred to liver through Vitamin D Binding Proteins for the process of Hydroxylation through the enzyme called 25-hydroxylase. The enzyme 25-

hydroxylase prefers Vitamin D3 as substrate rather than Vitamin D2. (Jones, *et al*., 2013) [32]. Moreover, In Liver the enzyme CYP27A1 doesn't hydroxylate Vitamin D2, therefore there are chances that more amount of Vitamin D3 is being converted into 25-hydroxyvitamin D (25OHD) than that of Vitamin D2 by all other enzymes. It has also been noted that Vitamin D Binding Protein has more affinity towards Vitamin D3 rather than Vitamin D2, the structural differences between Vitamin D2 and Vitamin D3 may play a significant role in determining its affinity towards Vitamin D binding Protein. All the reasons might be enough to understand the bioavailability of Vitamin D2 and Vitamin D3. (Tripkovic *et al*., 2012) [70]. Further 25-hydroxyvitamin D (25OHD) is transferred to Kidney with the help of Vitamin D binding Proteins where the enzyme called 1-α- Hydroxylase converts the Vitamin D into its bioactive form known as Vitamin D (1,25 OH2D). The enzyme involved in this process CYP2R1 which equally Hydroxylates 25-hydroxyvitamin D (25OHD2) and 25-hydroxyvitamin D (25OHD3). Three Hormones are known to control this process namely: Parathyroid hormone (PTH), fibroblast (FGF23) and 1,25 OH2D itself. Parathyroid hormone is found to stimulate the Hydroxylation of Vitamin D in the body whereas the other two hormones (fibroblast and 1,25 OH2D) inhibit the process. These hormones work in co-ordination with each other to maintain the Vitamin D serum levels in the body. (Heaney *et al*., 2011). Vitamin D3 vs. Vitamin D2 metabolism has been shown in Figure 1.

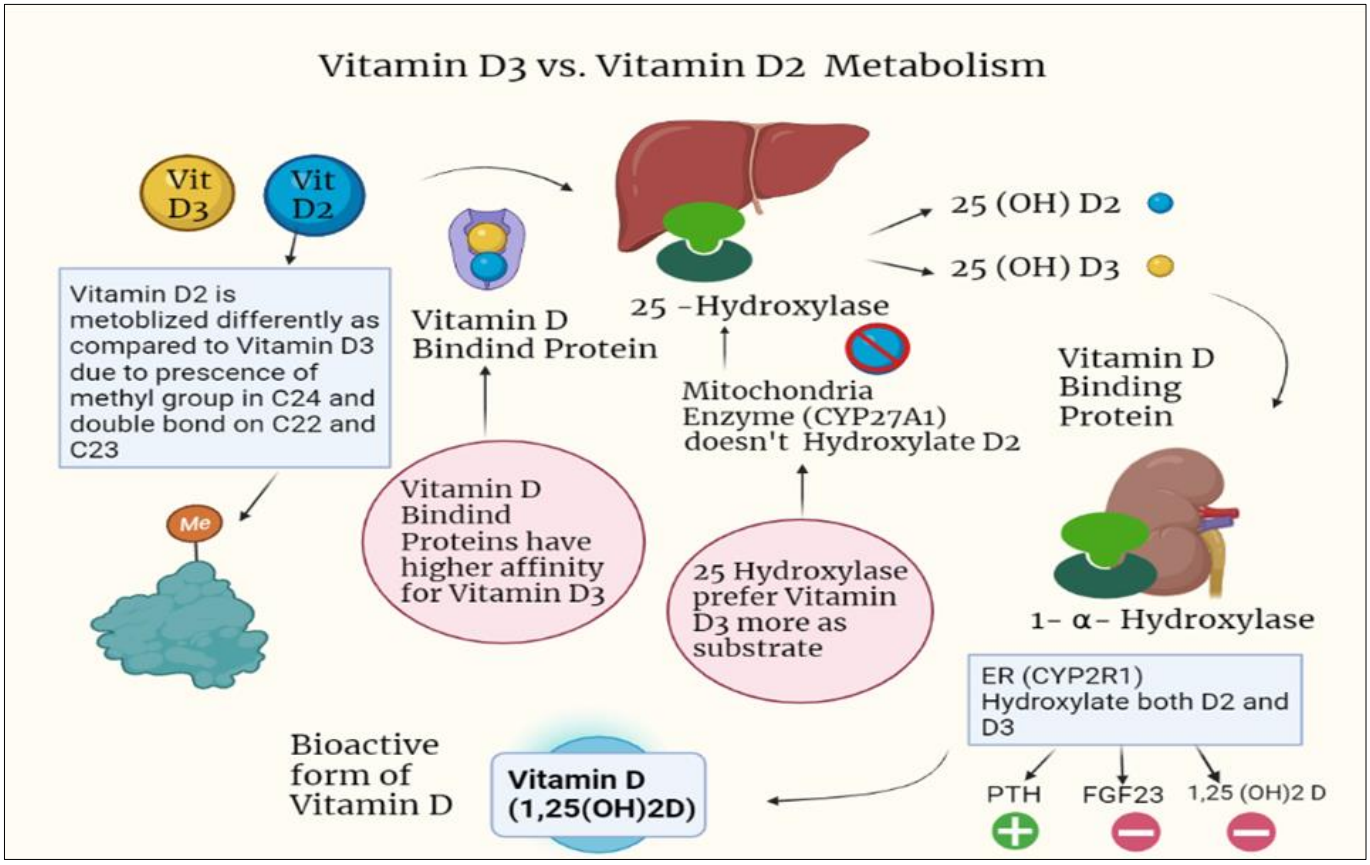

Fig 1: Mechanism of Vitamin D2 vs. Vitamin D metabolism

3.2 Evidence based studies

The higher bioavailability of Vitamin D3 may be much related to lower serum levels of 25(OH) D3 on addition of vitamin D2. Seven studies claim that Serum 25(OH) D significantly increased after the supplementation of vitamin D3. (Lehmann *et al*., 2013). In studies equal 500 IU of vitamin D2 and Vitamin D3 supplemented to a healthy individual showed similar effects on the serum levels of 25(OH) D within 3 days but in long run vitamin D2 has more chances of getting depleted as compared to Vitamin D3 in 60 Days of interval. Another studies also showed a rapid decline in vitamin D2 within 14 days. The serum level of Vitamin D3 was approx 5 times more than that of Vitamin D2 i.e 512 nmol.d/L and 151 nmol.d/L Vitamin D2 and Vitamin D3 respectively. (Heaney *et al*., 2011). Vitamin D3 was known to increase the Serum 25(OH) D 56-87% higher than that of Vitamin D2. A consistent bioavailability for both Vitamin D2 and Vitamin D3 was observed in both supplemented and fortified form in Orange Juice. In contrast many studies indicated that Vitamin D3 was 2 times more bioavailable than its other form Vitamin D2. (Adamec, *et al*., 2011) [2]. It may be stated that bioavailability of any form of Vitamin D is not just dependent on the form of vitamin D but also on form in which it is consumed. The Vitamin D3 was more bioavailable in supplement rather than Vitamin D2 consumed in oil. The human health condition also plays a crucial role in determining the bioavailability of both the forms. The subjects suffering from cystic fibrosis had lower absorption rates. (Tripkovic *et al*., 2012) [70]. However, bio equivalency of both forms of Vitamin D is still debatable. The contrasting results obtained in bioavailability of both forms of Vitamin D2 and Vitamin D3 may dependent on multiple factors as absorption of both forms Vitamin D2 and Vitamin D3 is independent from its hydroxylation rather it is more dependent on the fact that both the forms differ in their affinity towards VDR for different class of Vitamin D.

4. Synergic Effects of Vitamin D and other micronutrients on Human Health

4.1 Vitamin K and Vitamin D

Vitamin K is found to play synergic role with Vitamin D in curing many diseases. It has been found in a study that Vitamin K supplemented along with Vitamin D and Calcium can improved Bone Mineral Density of postmenopausal women. (Je *et al*., 2011) [31]. It has been found that insufficient amount of vitamin K in the body reduced the affinity of Calcium to bone matrix as calcium levels are significantly controlled by Vitamin D. A therapy including intake of combination of Vitamin k2 and Vitamin D3 can elevate BMD and lower serum UcOC. (Van Ballegooijen *et al*., 2017) [76-77]. In study it was clearly shown that taking Vitamin D+ K can significantly lower baseline eGFR (22.4 ±10.1 ml/min/1.73m2 vs 30.2 ±12.6 ml/min/1.73m2) lower serum uric acid levels (6.8 ±1.7 vs 8.5 ±1.9 mg/dl) and higher calcium × phosphate product (3.3 ±1.06 vs 2.7 ±0.6 mmol2/l2). The BMD after 6 months treatment of postmenopausal women significantly in increased from 0.01 ± 0.03 g/cm^2 vs -0.008 ± 0.04 g/cm^2 as shown in Table 3. A proved that by intake of 1000 µg/day vitamin K1 + 320 IU vitamin D in combination can improve cardiovascular health cardiovascular health maintaining the characteristic of wall of vessels in cartid artery along with balancing CAC and lowering carotid intima-media thickness (CCA-IMT) in the heart as compared to the patients taking alone vitamin D. (Van Ballegooijen *et al*. 2017) [76-77]. The mechanism behind Vitamin K and Vitamin D interaction can be studied in the figure, where it is shown that Vitamin D Binding Receptors undergo gamma Carboxylation which further results in altering the biochemical properties and increasing Osteocalcin which maintains the bone health as shown in Fig 2. The interaction has also been seen to play important role in glycoxidation can be counterbalanced by adding Vitamin D + Vitamin K which can enhance the maturation of Osteoblastic cells. The role of Vitamin K and Vitamin D can be understood in maintaining the cardio vascular health by favoring γ-glutamate carboxylation to prevent vascular calcification which is not desirable for CVD Patient.

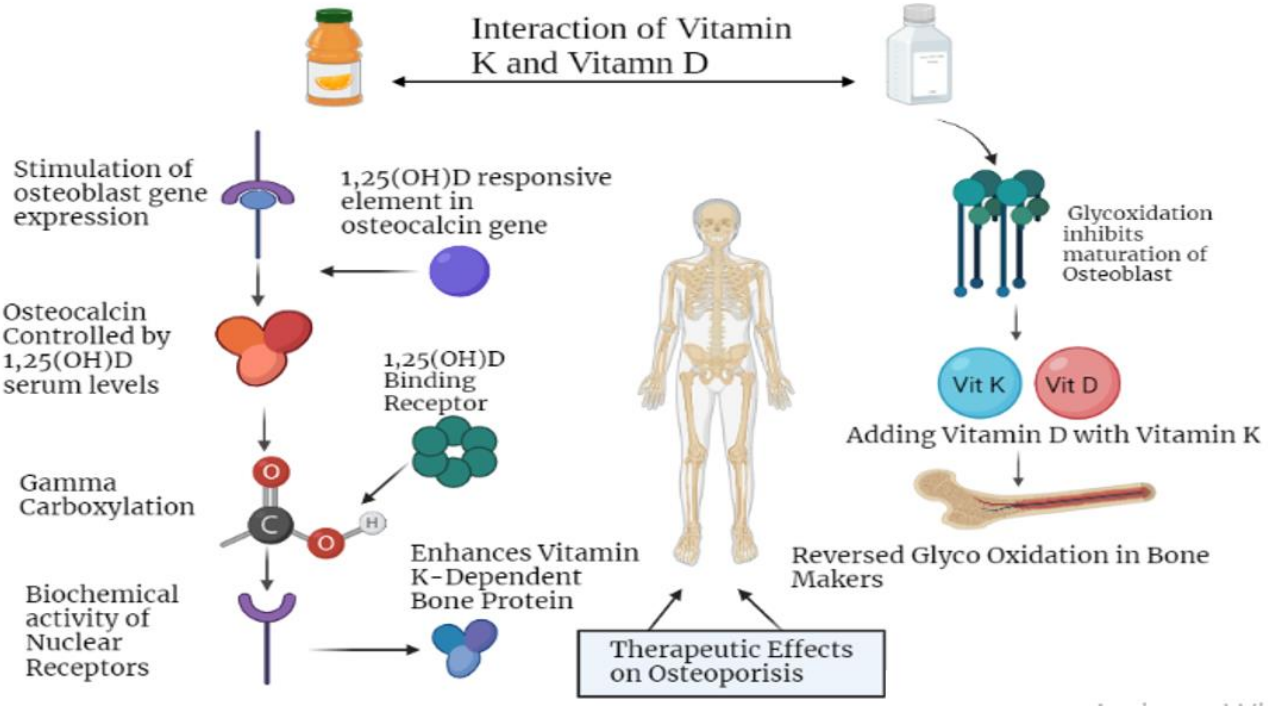

Fig 2: Mechanism of Vitamin D and Vitamin K interaction in maintaining Bone Health

4.2 Vitamin D, Vitamin C and Zinc

Vitamin D, Vitamin C and Zinc are seen to play synergic role in improving the adaptive as well as innate immunity in the body by enhancing the function of epithelial barriers, cellular defense and antibodies constituting the three main lines of immune defense. An increase in respiratory infections has been observed even with deficiency or insufficiency of one of them. They work such that the role and activity of each of them is stimulated as well as the functions performed by them is divided in the body. Vitamin C plays its role in minimizing the symptoms of Common Cold or Influenza and reduces the stress level in human reducing the level of cortisol by 50%. On the other hand Zinc in the form of lozenges plays role in relieving the symptom of Common Cold or Infection if consumed within 24. Zinc is another co-factor for vitamin D in maturation and activity of osteoblast cells. (Schwalfenberg, *et al*. 2015) [62]. It has been observed that vitamin D caused significant (159%) increase in zinc transport in cultured Caco-2 cells which improves anti oxidative properties of both the elements. Therefore, consumption of combination of 1000 mg ascorbic acid plus 10 mg zinc is recommended for Rhinorrhea. Simultaneously, Vitamin D enhances the activity of potent antimicrobial peptides, synthesized in neutrophils, monocytes, NK cells and in epithelial cells lining which protect the lungs from getting infected. It has been suggested that Vitamin D deficiency can increase the risk of respiratory infection by 12%. (Maggini *et al*., 2017) [47].

4.3 Magnesium and Vitamin D

Magnesium and Vitamin D have been seen to be synergic to each other as magnesium is reported to play an important role in activation of Vitamin D and vice versa. Magnesium is one the cofactors for vitamin D binding proteins whereas magnesium serum levels have been seen to elevate with

increased consumption of Vitamin D to 2000 IU for 6 months in diabetic patients. In addition to this, hepatic and renal metabolism of Vitamin D dependent on magnesium levels in the body. (Uwitonze *et al*., 2018) [75]. Lower levels of magnesium in the body also results in impaired PTH function, which one of the major stimulant of Vitamin D metabolism. It plays significant role in bone mineralization by activating vitamin D metabolites. Higher consumptions of magnesium lower the risk of Vitamin D deficiency. Apart from Vitamin D and Calcium, magnesium can also reduce the chances of Osteoporosis. Oral supplementation of magnesium gave some clinically important results as it elevated the osteocalcin levels by 44% in women and lowered Urine deoxypyridinoline levels by 41%. (Dai, *et al*.2018).

4.4 Other Synergic factors

The combined effect of Vitamin D and Metformin was studied and it was observed that insulin IGFBP-3 level increased and increased Phosphorylation of Akt which inhibited the activity of mTOR, P70S6K and S6P proteins; this improves the chemo preventive activity of vitamin D3 (Li *et al*., 2015) [42]. The proteins c-Myc and Cylin D1 significantly increased which enhanced the expression of VDR and CYP27B1 as compared to taking Vitamin D or metformin alone in colorectral Cancer patients. Vitamin D was also found to be in positive correlation with estradiol (E2) in women having metabolic Syndrome. Low estradiol (E2) elevated the risk of Vitamin D defeiciency in postmenopausal women. Soyisoflavone and Vitamin D increase the BMD in menopausal women as Soyisoflavones stimulate the expression of VDR consequently increasing osteoblast activity in maintaining Bone density. Not only isoflavones and Vitamin D increase the BMD but also prevents the Bone resorption by releasing Estrogen Receptor which inhibits osteoclast maturation thereby increasing the OPG formation. (Park, *et al*., 2012) [56]. Monounsaturated fatty acids consumption in the diet can improve the bioavailability of vitamin D3 in healthy older adults compared to polyunsaturated fatty acid. Phytosterols also enhanced the absorption of Vitamin D being a fat soluble Vitamin.

5. Factors Affecting the Prevalence of Vitamin D Bioavailability

Vitamin D deficiency has become a common micronutrient deficiency not only in India but in many other countries. The prevalence of Vitamin D deficiency depends on multiple factors; some of those factors are discussed below and the table 2. Below shows prevalence of Vitamin D deficiency throughout many countries of world on based of different life cycles.

Table 2: Prevalence of Vitamin D deficiency in World and India

S. No.		New borns -infants	>5 years	12-18 years	18-50 years	Pregnant and Lactating Mothers	Elderly People	References
1.	India	86%	61.4%	94.8%	83%	74%	91.2%	Mehlawat, *et al*.,(2014) [51]
2.	US	70%	40%	21%	80%	50%	>40%	Parva, *et al*., (2018) [57], Lee, *et al*. (2017), Dawodu, *et al*., (2012) [14]. Kunadian, *et al*. (2014) [39]. Forrest, *et al*., (2011) [16]. Turer, *et al*. (2013) [72]. Andıran *et al*. (2012) [3].
3.	China	36.3%	50%	40% 90%	76%	90.5%	70-90%	Zhang *et al*., (2013). Yu, *et al*., (2014) [83], Tao, *et al*. (2012) [67]. Lu *et al*., (2012) [45].
4.	Australia	40-57%	10%	68%	20%	60-80%	75%	Paxton *et al*. (2013) [58]. Nowson *et al*., (2012) [54]. Malacova, *et al*. (2019) [48]. Tran *et al*., (2013) [69].
5.	Europe	57%	80%	55%	45.8%	74.1%	61%	Hribar, *et al*., (2021) [27]. Vandevijvere *et al*., (2012) [78]. Saraf *et al*. (2016) [61].
6.	Africa	65%	7%	14-25%	36%	44%	48%	Mogire, *et al*. (2020) [52], Principi *et al*. (2013) [60], Green *et al*. (2015) [19], Kalkwarf, *et al*. (2012) [33], Braegger *et al*. (2013) [9].
7.	Japan	25%	-	90%	60%	73.2%	60.1%	Kanatani *et al*. (2019) [85], Okazaki *et al*. (2017) [84], Mackawy *et al*. (2013), Kubota *et al*. (2018) [86], Inamo *et al*. (2011).
8.	Korea	48.7%	58.6%	71.8%	83.0%	79.2%	62.1%	Choi, *et al*. (2015) [87], Jung, *et al*. (2013) [88], Nah, *et al*. (2015) [89], Seo, *et al*. (2013) [90], Kim, *et al*. (2018) [91], Chung, *et al*. (2014), Choi, *et al*. (2013) [92].
9.	Sri Lanka	63%	35%	13.2%	58.8%	17%	-	Anusha, *et al*. (2019) [93], Jayatissa, *et al*. (2019) [94], Subasinghe, *et al*. (2019) [95], Godawita *et al*. (2021) [96].

5.1 Processing of Food

Cooking eggs in three different ways can also eliminate the Vitamin D in significant amount. About 10% of Vitamin D was lost by boiling the egg for 10 mins and 20% loss was observed by scrambling the eggs for about 3mins. Vitamin D in UHT fortified milk was lost upto 10% every month when stored at 4 ºC. Loss in Vitamin D during pasteurized milk, UHT milk, dry milk and dry milk desserts was noticed to be 20%, 30%, 40% and 20%. Jakobsen *et al*., (2014) [29.30].

5.2 Antagonistic Vitamins and Minerals

The absorption of Vitamin D in GI tract was reduced to 17% with higher concentration of Vitamin E in the body. Vitamin A also interfered with Vitamin D absorption and reduced by 30%. In a study it was found that S- calcium decreased after intake of retinyl palmitate with 1,25 $(OH)_2D_3$ whereas PTH levels remained unaffected. The antagonistic nature of both Vitamins is expected to be due to fat soluble behavior of them.

5.3 Drugs

Bile acid sequestrants like colestipol and cholestyramine bind to fat soluble Vitamins like Vitamin D. Therefore over excretion of Bile acid from the body can lead to reduced

stores of Vitamin D. Orlistat used for weight loss inhibits the absorption of dietary fats in the body corresponding to low Vitamin D levels in the body. Statins is another drug used for checking the cholesterol levels also interferes with Vitamin D metabolism as Vitamin D is derived from Cholesterol. Vitamin D and Statins compete for CYP3A4 activity which establishes the antagonistic relationships between them.

5.4 Age

Age is also seen to play important role in Vitamin D metabolism in the body due change is lifestyle, Dietary (28%). 19% of menopausal women suffering from osteoporosis discontinued the intake of calcium + vitamin D due to lack of motivation being the most common reason, followed by fear of side effects being the other factor. (Liyanage, *et al*. (2011). The fortified milk when stored in glass bottle (71.81%) showed higher stability rather than the one stored in LDPE pouches. (Moulas, *et al*. 2018) [53].

5.5 Skin Pigments: The skin pigments play significant role in Vitamin D production. It was found in the study that on exposing the individuals from different races to same UVB radiation fair skinned individuals showed increased serum Vitamin D levels whereas black individuals showed no significant difference in serum Vitamin D levels within 2 weeks. The melenocytes from lower epidermis layer moved to middle epidermis increasing melenocytes in black individuals 4 times more than that of fair skinned individual. The dietary intake for fair skinned and black skinned children was different 6 and 14 µg vitamin D/d . (Mostafa *et. al*., 2015).

6. Enhancing bioavailability of Vitamin D:

6.1 Vitamin D Fortification and Food Enhancement:

Vitamin D fortification is simply adding Vitamin D to foods and that food is claimed to have additional amounts of Vitamin D. Recently, Vitamin D is fortified by feeding the livestock animals with Vitamin D Rich Diet or Feed which can increase Vitamin D content up to significant amount. This process is also known as Vitamin D bio-addition. Different countries have different laws for fortification of Vitamin D, for example in Germany Vitamin D fortifications is restricted to margarines only whereas in Finland fluid milks, margarines/fat spreads are recommended to be fortified. (Pilz *et. al*., 2018) [59]. The countries like U.S, Canada and Finland follow the strategy of mass fortification. The mean serum levels increased to 7.6 nmol/L from the year 2000 to 2011 in Finland. WHO recommends a Dietary intake of Vitamin D to be 10 µg (400 IU) per day so as to reach serum level of 40-50 nmol/L. If more than 2.5% population is seen to be Vitamin D deficient then the need for Vitamin D fortification is set up. Same recommendations in India are suggested by ICMR i.e. 400 IU consumption of Vitamin D by 97.5% population of the country. (Calvo *et al.*, 2013) [11]. Additional Intake through fortified foods may reach up to 1000-4000 IU per day. The Vitamin D concentration in fortified foods should not exceed 20 IU/100 Calories. In elderly Vitamin D deficiency is common because surface area of skin decreases with age. The evidences indicate that supplement vitamin D has dosage effect on improving bone health and muscle strength and in the elderly 25(OH)-D serum concentrations of 75110 mol/L provide an adequate status for fall and fracture prevention. It is also claimed to inhibit the division of cells in colorectal cancer, works as immune system regulator and modulate the autoimmune diseases. The foods which are commonly fortified with vitamin D are juices, bread, milk and dairy products. (Gupta, A. (2014) [20] It has been proposed that 8001,000 IU of vitamin D per day may be needed to achieve a satisfactory status in all adults of 75100 nmol/L of 25(OH)-D. It has been shown that 49% of adults (57-85 years) consume dietary supplements mostly in the form of multivitamin (28%). 19% of menopausal women suffering from osteoporosis discontinued the intake of calcium + vitamin D due to lack of motivation being the most common reason, followed by fear of side effects being the other factor. (Liyanage, *et al*. (2011). The fortified milk when stored in glass bottle (71.81%) showed higher stability rather than the one stored in LDPE pouches. (Moulas, *et. al*. 2018) [53].

6.2 UV radiations on Vitamin D

Mushrooms are one of the richest vegetarian sources of Vitamin D. It is consumed worldwide as part of their common diet when it is cooked in combination with other vegetables. Usually, the Vitamin D in mushrooms is present in the form of Vitamin D_2, whereas other forms of Vitamin D such as Vitamin D_3 and Vitamin D_4 are present in small amounts as vitamin D_3 is mostly present in animal based sources. Mushrooms when exposed to UV radiations by using UV lamps can be enhanced with significant amounts of vitamin D. (Keegan *et al*., 2013) [34]. The amount of Vitamin D in fresh mushrooms is most likely to be 10µg/100g. On exposing the mushroom to UV radiation lamps the vitamin D content in mushroom can increase up to 40µg/g dried mass. (Ahlawat, O *et al*. 2016) The most effective range of wavelength of radiations used is 280-315 nm. Although the fixed temperature range for the radiations is still not known but some studies suggest the 25-35 ^{o}C to be most ideal temperature range for exposure of the mushrooms. UV pulsed lamp can also be used for this purpose it can enhance the vitamin D content in the mushroom in relatively very short period of time (1-3s). High energy pulsed lamp can penetrate deep into the flesh of mushroom and produce higher Vitamin D content in the mushroom. Vitamin D content in fresh mushrooms exposed to UV radiation may depend on whether they are sliced or whole, the distance from the lamp housing, the size of the mushroom, and the total number of pulses received. On being exposed to UVB radiations hens produced eggs in which Vitamin D increased from 1.1 µ/100g to 1.7 µ/100g.

6.3 Nano-encapsulation of Vitamin D

Nanoencapsulation is a technology of entrapping a bioactive component within a nano–capsule to enhance the solubility and bioavailability of an element. This technology overcomes the drawbacks of functions food as the vitamin D fortified foods are usually seen to lose vitamin D content overtime during processing of the food. (Haham, *et al*. 2012) [21]. In a study starch nanoparticles were used to entrap the Vitamin D using ultrasonic technique. Starch is considered as potential nanocarrier of Vitamin D as it has amorphous polysaccharides in the center which are stable to acid hydrolysis and this sets the biocompatibility between both the molecules as shown in figure no. 3. Another reason for using Starch is its ability to release the attached molecule slowly. Using nanoencapsulation improves the solubility of Vitamin D as smaller the size small is the Brownian movement due to gravity and also nanoparticles smaller in size have high dissolution which is favorable for poor water soluble compound. Encapsulation efficiency was as high as 71% and

Load efficiency also increase due formation of Hydrogen Bonding between Vitamin D and Starch which resulted in slow release of Vitamin D (Hasanvand *et al*., 2015) [22]. The technology is highly stable in acidic and enzymatic condition because only 3.5% of Vitamin D was released within 2 hours. Thermostability was also observed in the Vitamin D as no peaks of Vitamin D at 87 and 286 °C (melting point of Vitamin D) were observed in thermogram. Usually nanoemulsion prepared at low frequency and high intensity is more suitable for fish oil and reduces the toxicity of nanoencapsulation because less surfactant is required. Fish oil nanoemulsion was found to be stable even after 45 days when kept undisturbed. The encapsulation efficiency was higher in nanocapsules having low fish oil (95.7%) as compared the nanocapsules having higher fish oil (98.2%), because per unit Vitamin D particles decrease in higher Fish oil content. The thick layer of oil in encapsulated Vitamin D prevented it from getting degraded in mouth (due to salivary amylase) and on stomach (acidic condition). (Walia *et al*., 2017) [79]. Vitamin D3 entrapped using lactoglobulin showed encapsulation efficiency of 94.5% whereas only 45.8 ± 1.55% to 67.6 ± 2.76% EE was observed for alginate based nanotechnology. However, almost similar EE results were obtained using zein-based hydrogel beads (96.9%). Free vitamin D when stored under refrigerator for 5 weeks lost 91% of Vitamin D whereas the lactoglobulin entrapped Vitamin D3 was retained significantly with loss of 8.3 ± 1.7% only because of combine effect of anti-oxidative properties of thiol group and coaglum matrix blocking the accessibility oxidative agents. Vitamin D3 was retained successfully in gastric tract as lactoglobulin structure remained consistent in the stomach as the pH of stomach remained same as that of lactoglobulin-Vitamin D complex. Only 3.7% of Vitamin D was lost as compared to unprotected Vitamin D 90.0 ± 1.3%. Loss was Vitamin D from lactoglobulin and casein was less as compared to nano particles of ethanol water mixture. (Diarrassouba *et al*. 2015) [15].

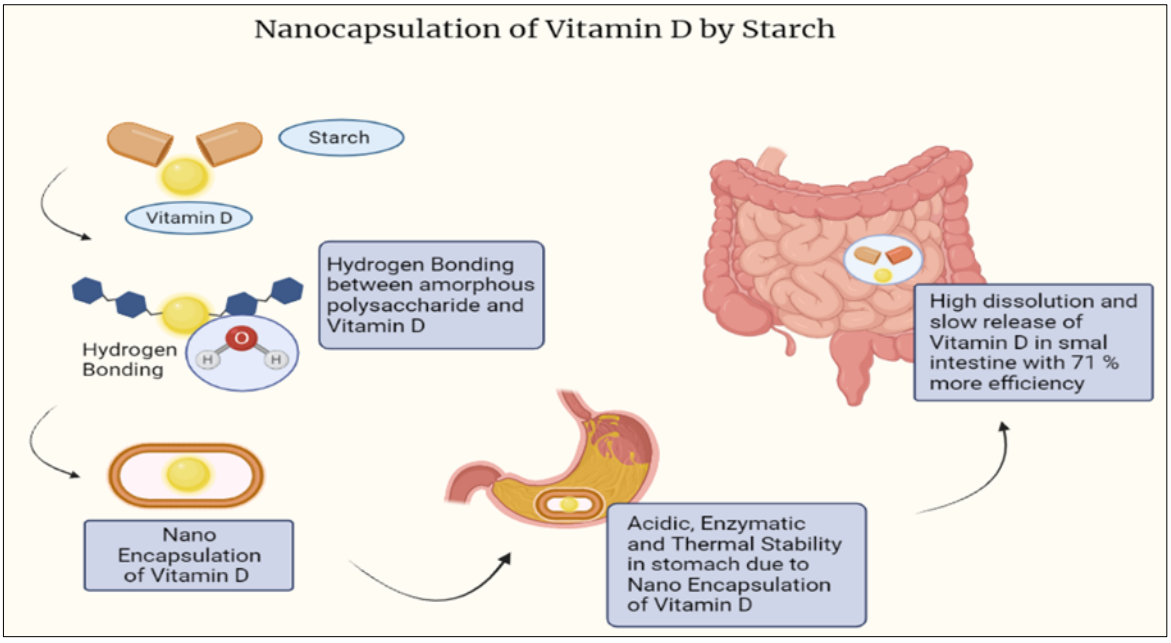

Fig 3: Nano-Encapsulation of Vitamin D by Starch to enhance the Bioavailability of Vitamin D

Conclusion

It can be concluded that Vitamin D is available in our body in two forms Vitamin D2 and Vitamin D3. The two forms of Vitamin D have different sources Vitamin D3 is found in animal based sources whereas Vitamin D2 is found in plant based sources. The absorption and metabolism of Vitamin D in the body is slightly different from one another as both the forms have different structure and DBP have more affinity for Vitamin D3 rather than that of Vitamin D2. More enzymes are involved in metabolism of Vitamin D3 rather than Vitamin D2. Evidence based study is available for Vitamin D3 vs. Vitamin D2 bioavailability. Vitamin D is seen to be in synergic relation with Vitamin K as both enhance the activity of one another and assist in treatment of bone density, cardiovascular diseases and hypertension. Vitamin D, zinc and Vitamin C are seen to improve immunity. Magnesium and Vitamin D are seen to improve the activity of osteoblasts and magnesium is major mineral in enzymes involved in Vitamin D metabolism. Many factors like processing, antagonist elements, aging, skin color, impaired absorption in GI tract can reduce the bioavailability of Vitamin D. Vitamin D can be enhanced in food by treating the food with UVB radiation and by encapsulating the Vitamin D with suitable matrix which results in slow release of Vitamin D consequently preventing it from being degraded during processing or being lost in GI tract.

References

1. Abbasi A, Emam-Djomeh Z, Mousavi MAE, Davoodi, D. Stability of vitamin D3 encapsulated in nanoparticles of whey protein isolate. Food chemistry. 2014;143:379-383.
2. Adamec J, Jannasch A, Huang J, Hohman E, Fleet JC, Peacock M, *et al*. Development and optimization of an LC-MS/MS-based method for simultaneous quantification of vitamin D2, vitamin D3, 25-hydroxyvitamin D2 and 25-hydroxyvitamin D3. Journal of separation science. 2011;34(1):11-20.
3. Andıran N, Celik N, Akca H, Doğan G. Vitamin D deficiency in children and adolescents. Journal of clinical research in pediatric endocrinology. 2012;4(1):25.
4. Awad AB, Alappat L, Valerio M. Vitamin d and

metabolic syndrome risk factors: evidence and mechanisms. Critical reviews in food science and nutrition. 2012;52(2):103-112.
5. Baur AC, Brandsch C, König B, Hirche F, Stangl GI. Plant oils as potential sources of vitamin D. Frontiers in nutrition. 2016;3:29.
6. Benedik E. Sources of vitamin D for humans. International Journal for Vitamin and Nutrition Research, 2021.
7. Bikle DD. Vitamin D metabolism, mechanism of action, and clinical applications. Chemistry & biology. 2014;21(3):319-329.
8. Borel P, Caillaud D, Cano NJ. Vitamin D bioavailability: state of the art. Critical reviews in food science and nutrition. 2015;55(9):1193-1205.
9. Braegger C, Campoy C, Colomb V, Decsi T, Domellof M, Fewtrell M, *et al*. Vitamin D in the healthy European paediatric population. Journal of pediatric gastroenterology and nutrition. 2013;56(6), 692-701.
10. Byrdwell WC, Exler J, Gebhardt SE, Harnly JM, Holden JM, Horst RL, *et al*. Liquid chromatography with ultraviolet and dual parallel mass spectrometric detection for analysis of vitamin D in retail fortified orange juice. Journal of Food Composition and Analysis. 2011;24(3):299-306.
11. Calvo MS, Whiting SJ. Survey of current vitamin D food fortification practices in the United States and Canada. The Journal of steroid biochemistry and molecular biology. 2013;136:211-213.
12. Cashman KD. Vitamin D: dietary requirements and food fortification as a means of helping achieve adequate vitamin D status. The Journal of steroid biochemistry and molecular biology. 2015;148:19-26.
13. Daly RM, Gagnon C, Lu ZX, Magliano DJ, Dunstan DW, Sikaris KA, *et al*. Prevalence of vitamin D deficiency and its determinants in Australian adults aged 25 years and older: A national, population-based study. Clinical endocrinology. 2012;77(1):26-35.
14. Dawodu A, Wagner CL. Prevention of vitamin D deficiency in mothers and infants worldwide—a paradigm shift. Paediatrics and international child health. 2012;32(1):3-13.
15. Diarrassouba F, Garrait G, Remondetto G, Alvarez P, Beyssac E, Subirade M. Improved bioavailability of vitamin D3 using a β-lactoglobulin-based coagulum. Food chemistry. 2015;172:361-367.
16. Forrest KY, Stuhldreher WL. Prevalence and correlates of vitamin D deficiency in US adults. Nutrition research. 2011;31(1):48-54.
17. Giménez VMM, Bergam I, Reiter RJ, Manucha W. Metal ion homeostasis with emphasis on zinc and copper: Potential crucial link to explain the non-classical antioxidative properties of vitamin D and melatonin. Life Sciences, 2021, 119770.
18. Giménez VMM, Bergam I, Reiter RJ, Manucha W. Metal ion homeostasis with emphasis on zinc and copper: Potential crucial link to explain the non-classical antioxidative properties of vitamin D and melatonin. Life Sciences. 2021;281:119770.
19. Green RJ, Samy G, Miqdady MS, El-Hodhod M, Akinyinka OO, Saleh G, *et al*. Vitamin D deficiency and insufficiency in Africa and the Middle East, despite year-round sunny days. SAMJ: South African Medical Journal. 2015;105(7):603-605.
20. Gupta A. Fortification of foods with vitamin D in India. Nutrients. 2014;6(9):3601-3623.
21. Haham M, Ish-Shalom S, Nodelman M, Duek I, Segal E, Kustanovich M, *et al*. Stability and bioavailability of vitamin D nanoencapsulated in casein micelles. Food & function. 2012;3(7):737-744.
22. Hasanvand E, Fathi M, Bassiri A, Javanmard M, Abbaszadeh R. Novel starch based nanocarrier for vitamin D fortification of milk: Production and characterization. Food and Bioproducts Processing. 2015;96:264-277.
23. Haussler MR, Whitfield GK, Kaneko I, Haussler CA, Hsieh D, Hsieh JC, *et al*. Molecular mechanisms of vitamin D action. Calcified tissue international. 2013;92(2):77-98.
24. Hill TR, Granic A, Aspray TJ. Vitamin D and ageing. Biochemistry and Cell Biology of Ageing: Part I Biomedical Science, 2018, 191-220.
25. Hohman EE, Martin BR, Lachcik PJ, Gordon DT, Fleet JC, Weaver CM. Bioavailability and efficacy of vitamin D2 from UV-irradiated yeast in growing, vitamin D-deficient rats. Journal of agricultural and food chemistry. 2011;59(6):2341-2346.
26. Hohman EE, Martin BR, Lachcik PJ, Gordon DT, Fleet JC, Weaver CM. Bioavailability and efficacy of vitamin D2 from UV-irradiated yeast in growing, vitamin D-deficient rats. Journal of agricultural and food chemistry. 2011;59(6):2341-2346.
27. Hribar M, Hristov H, Lavriša Ž, Koroušić Seljak B, Gregorič M, Blaznik U, *et al*. Vitamin D Intake in Slovenian Adolescents, Adults, and the Elderly Population. Nutrients. 2021;13(10):3528.
28. Hu B, Huang QR. Biopolymer based nano-delivery systems for enhancing bioavailability of nutraceuticals. Chinese Journal of Polymer Science. 2013;31(9):1190-1203.
29. Jakobsen J, Knuthsen P. Stability of vitamin D in foodstuffs during cooking. Food chemistry. 2014;148:170-175.
30. Jakobsen Jette, Knuthsen Pia. Stability of vitamin D in foodstuffs during cooking. Food Chemistry. 2014;148:170-175. DOI:10.1016/j.foodchem.2013.10.043
31. Je SH, Joo NS, Choi BH, Kim KM, Kim BT, Park SB, *et al*. Vitamin K supplement along with vitamin D and calcium reduced serum concentration of undercarboxylated osteocalcin while increasing bone mineral density in Korean postmenopausal women over sixty-years-old. Journal of Korean medical science. 2011;26(8):1093-1098.
32. Jones G. Extrarenal vitamin D activation and interactions between vitamin D2, vitamin D3, and vitamin D analogs. Annual review of nutrition. 2013;33:23-44.
33. Kalkwarf HJ, Denburg MR, Strife CF, Zemel BS, Foerster DL, Wetzsteon RJ, *et al*. Vitamin D deficiency is common in children and adolescents with chronic kidney disease. Kidney international. 2012;81(7):690-697.
34. Keegan RJH, Lu Z, Bogusz JM, Williams JE, Holick MF. Photobiology of vitamin D in mushrooms and its bioavailability in humans. Dermato-endocrinology. 2013;5(1):165-176.
35. Kienreich K, Tomaschitz A, Verheyen N, Pieber T, Gaksch M, Grübler MR, *et al*. Vitamin D and

cardiovascular disease. Nutrients. 2013;5(8):3005-3021.
36. Kristensen H, Rosenqvist E, Jakobsen J. Increase of vitamin D2 by UV-B exposure during the growth phase of white button mushroom (*Agaricus bisporus*). Food & nutrition research. 2012;56(1):7114.
37. Kučan R, Soltirovska Šalamon A, Andronikov D, Benedik E. Dietary Sources of Vitamin D, Vitamin D Supplementation, and Its Bio-Viability. Central European Journal of Paediatrics. 2018;14(2):115-122.
38. Kühn J, Schröter A, Hartmann BM, Stangl GI. Cocoa and chocolate are sources of vitamin D2. Food chemistry. 2018;269:318-320.
39. Kunadian V, Ford GA, Bawamia B, Qiu W, Manson JE. Vitamin D deficiency and coronary artery disease: A review of the evidence. American heart journal. 2014;167(3):283-291.
40. Lee JM, Smith JR, Philipp BL, Chen TC, Mathieu J, Holick MF. Vitamin D deficiency in a healthy group of mothers and newborn infants. Clinical pediatrics. 2007;46(1):42-44.
41. Lehmann U, Hirche F, Stangl GI, Hinz K, Westphal S, Dierkes J. Bioavailability of vitamin D2 and D3 in healthy volunteers, a randomized placebo-controlled trial. The Journal of Clinical Endocrinology & Metabolism. 2013;98(11):4339-4345.
42. Li W, Wang QL, Liu X, Dong SH, Li HX, Li CY, *et al*. Combined use of vitamin D3 and metformin exhibits synergistic chemopreventive effects on colorectal neoplasia in rats and mice. Cancer Prevention Research. 2015;8(2):139-148.
43. Libon F, Cavalier E, Nikkels AF. Skin color is relevant to vitamin D synthesis. Dermatology. 2013;227(3):250-254.
44. Lips P, van Schoor NM, de Jongh RT. Diet, sun, and lifestyle as determinants of vitamin D status. Annals of the New York Academy of Sciences. 2014;1317(1):92-98.
45. Lu HK, Zhang Z, Ke YH, He JW, Fu WZ, Zhang CQ, *et al*. High prevalence of vitamin D insufficiency in China: relationship with the levels of parathyroid hormone and markers of bone turnover. PloS one. 2012;7(11):e47264.
46. Mager DR, Qiao J, Turner J. Vitamin D and K status influences bone mineral density and bone accrual in children and adolescents with celiac disease. European Journal of Clinical Nutrition. 2012;66(4):488-495.
47. Maggini S, Maldonado P, Cardim P, Newball CF, Latino ES. Vitamins C, D and zinc: synergistic roles in immune function and infections. Vitam Miner. 2017;6(167):2376-1318.
48. Malacova E, Cheang PR, Dunlop E, Sherriff JL, Lucas RM, Daly RM, *et al*. Prevalence and predictors of vitamin D deficiency in a nationally representative sample of adults participating in the 2011–2013 Australian Health Survey. British Journal of Nutrition. 2019;121(8):894-904.
49. Manson JE, Bassuk SS, Cook NR, Lee IM, Mora S, Albert CM, *et al*. Vitamin D, marine n-3 fatty acids, and primary prevention of cardiovascular disease current evidence. Circulation research. 2020;126(1):112-128.
50. Maurya VK, Aggarwal M. Factors influencing the absorption of vitamin D in GIT: an overview. Journal of food science and technology. 2017;54(12):3753-3765.
51. Mehlawat U, Singh P, Pande S. Current status of Vitamin-D deficiency in India. Innov Pharm Pharmac other. 2014;2(2):328-35.
52. Mogire RM, Mutua A, Kimita W, Kamau A, Bejon P, Pettifor JM, *et al*. Prevalence of vitamin D deficiency in Africa: a systematic review and meta-analysis. The Lancet Global Health. 2020;8(1):e134-e142.
53. Moulas AN, Vaiou M. Vitamin D fortification of foods and prospective health outcomes. Journal of biotechnology. 2018;285:91-101.
54. Nowson CA, McGrath JJ, Ebeling PR, Haikerwal A, Daly RM, Sanders KM, *et al*. Vitamin D and health in adults in Australia and New Zealand: a position statement. Medical journal of Australia. 2012;196(11):686-687.
55. O'Mahony L, Stepien M, Gibney MJ, Nugent AP, Brennan L. The potential role of vitamin D enhanced foods in improving vitamin D status. Nutrients. 2011;3(12):1023-1041.
56. Park CY, Weaver CM. Vitamin D interactions with soy isoflavones on bone after menopause: A review. Nutrients. 2012;4(11):1610-1621.
57. Parva NR, Tadepalli S, Singh P, Qian A, Joshi R, Kandala H, *et al*. Prevalence of vitamin D deficiency and associated risk factors in the US population (2011-2012). Cureus, 2018, 10(6).
58. Paxton GA, Teale GR, Nowson CA, Mason RS, McGrath JJ, Thompson MJ, *et al*. Vitamin D and health in pregnancy, infants, children and adolescents in Australia and New Zealand: a position statement. Medical Journal of Australia. 2013;198(3):142-143.
59. Pilz S, März W, Cashman KD, Kiely ME, Whiting SJ, Holick MF, *et al*. Rationale and plan for vitamin D food fortification: a review and guidance paper. Frontiers in endocrinology. 2018;9:373.
60. Principi N, Bianchini S, Baggi E, Esposito S. Implications of maternal vitamin D deficiency for the fetus, the neonate and the young infant. European Journal of Nutrition. 2013;52(3):859-867.
61. Saraf R, Morton SM, Camargo Jr CA, Grant CC. Global summary of maternal and newborn vitamin D status–a systematic review. Maternal & child nutrition. 2016;12(4):647-668.
62. Schwalfenberg GK, Genuis SJ. Vitamin D, essential minerals, and toxic elements: exploring interactions between nutrients and toxicants in clinical medicine. The Scientific World Journal, 2015.
63. Scrimshaw NS, SanGiovanni JP. Synergism of nutrition, infection, and immunity: an overview. The American journal of clinical nutrition. 1997;66(2):464S-477S.
64. Song SJ, Zhou L, Si S, Liu J, Zhou J, Feng K, *et al*. The high prevalence of vitamin D deficiency and its related maternal factors in pregnant women in Beijing. PLoS One. 2013;8(12):e85081.
65. Syama MA, Arora S, Gupta C, Sharma A, Sharma V. Enhancement of vitamin D2 stability in fortified milk during light exposure and commercial heat treatments by complexation with milk proteins. Food Bioscience. 2019;29:17-23.
66. Szabó A, Gyepes A, Nagy Á, Abrankó L, Győrfi J. The effect of UVB radiation on the vitamin D2 content of white and cream type button mushrooms (*Agaricus bisporus* Lange/Imbach) and oyster mushroom (Pleurotus ostreatus (Jacq.) P. Kumm). Acta Alimentaria. 2012;41(Supplement-1):187-196.
67. Tao M, Shao H, Gu J, Zhen Z. Vitamin D status of pregnant women in Shanghai, China. The Journal of

Maternal-Fetal & Neonatal Medicine. 2012;25(3):237-239.
68. Taofiq O, Fernandes Â, Barros L, Barreiro MF, Ferreira, IC. UV-irradiated mushrooms as a source of vitamin D2: A review. Trends in food science & technology. 2017;70:82-94.
69. Tran B, Armstrong BK, McGeechan K, Ebeling PR, English DR, Kimlin MG, *et al*. Predicting vitamin D deficiency in older Australian adults. Clinical endocrinology. 2013;79(5):631-640.
70. Tripkovic L, Lambert H, Hart K, Smith CP, Bucca G, Penson S, *et al*. Comparison of vitamin D2 and vitamin D3 supplementation in raising serum 25-hydroxyvitamin D status: a systematic review and meta-analysis. The American journal of clinical nutrition. 2012;95(6):1357-1364.
71. Tripkovic L, Wilson LR, Lanham-New SA. Vitamin D2 vs. vitamin D3: They are not one and the same, 2017.
72. Turer CB, Lin H, Flores G. Prevalence of vitamin D deficiency among overweight and obese US children. Pediatrics. 2013;131(1):e152-e161.
73. Urbain P, Jakobsen J. Dose–response effect of sunlight on vitamin D2 production in *Agaricus bisporus* mushrooms. Journal of agricultural and food chemistry. 2015;63(37):8156-8161.
74. Urbain P, Singler F, Ihorst G, Biesalski HK, Bertz H. Bioavailability of vitamin D 2 from UV-B-irradiated button mushrooms in healthy adults deficient in serum 25-hydroxyvitamin D: A randomized controlled trial. European journal of clinical nutrition. 2011;65(8):965-971.
75. Uwitonze AM, Razzaque MS. Role of magnesium in vitamin D activation and function. Journal of Osteopathic Medicine. 2018;118(3):181-189.
76. Van Ballegooijen AJ, Cepelis A, Visser M, Brouwer IA, Van Schoor NM, Beulens JW. Joint association of low vitamin D and vitamin K status with blood pressure and hypertension. Hypertension. 2017;69(6):1165-1172.
77. Van Ballegooijen AJ, Pilz S, Tomaschitz A, Grübler MR, Verheyen N. The synergistic interplay between vitamins D and K for bone and cardiovascular health: a narrative review. International journal of endocrinology, 2017.
78. Vandevijvere S, Amsalkhir S, Van Oyen H, Moreno-Reyes R. High prevalence of vitamin D deficiency in pregnant women: a national cross-sectional survey, 2012.
79. Walia N, Dasgupta N, Ranjan S, Chen L, Ramalingam C. Fish oil based vitamin D nanoencapsulation by ultrasonication and bioaccessibility analysis in simulated gastro-intestinal tract. Ultrasonics sonochemistry. 2017;39:623-635.
80. Wang L, Evatt ML, Maldonado LG, Perry WR, Ritchie JC, Beecham GW, *et al*. Vitamin D from different sources is inversely associated with Parkinson disease. Movement Disorders. 2015;30(4):560-566.
81. Wilson LR, Tripkovic L, Hart KH, Lanham-New SA. Vitamin D deficiency as a public health issue: using vitamin D2 or vitamin D3 in future fortification strategies. Proceedings of the Nutrition Society. 2017;76(3):392-399.
82. Wimalawansa SJ. Non-musculoskeletal benefits of vitamin D. The Journal of steroid biochemistry and molecular biology. 2018;175:60-81.
83. Yu X, Wang W, Wei Z, Ouyang F, Huang L, Wang X, *et al*. Vitamin D status and related factors in newborns in Shanghai, China. Nutrients. 2014;6(12):5600-5610.
84. Okazaki Ryo, Ozono Keiichi, Fukumoto Seiji, Inoue Daisuke, Yamauchi Mika, Minagawa Masanori, *et al*. Assessment criteria for vitamin D deficiency/insufficiency in Japan: proposal by an expert panel supported by the Research Program of Intractable Diseases, Ministry of Health, Labour and Welfare, Japan, the Japanese Society for Bone and Mineral Research and the Japan Endocrine Society [Opinion]. Journal of Bone and Mineral Metabolism. 2017;35(1):1-5. DOI:10.1007/s00774-016-0805-4
85. Kanatani KT, Nakayama T, Adachi Y, Hamazaki K, Onishi K, Konishi Y, *et al*. High frequency of vitamin D deficiency in current pregnant Japanese women associated with UV avoidance and hypo-vitamin D diet. PloS one. 2019;14(3):e0213264.
86. Kubota Takuo, Nakayama Hirofumi, Kitaoka Taichi, Nakamura Yosikazu, Fukumoto Seiji, Fujiwara Ikuma, *et al*. Incidence rate and characteristics of symptomatic vitamin D deficiency in children: A nationwide survey in Japan. Endocrine Journal, 2018. DOI:10.1507/endocrj.EJ18-0008.
87. Choi R, Kim S, Yoo H, Cho YY, Kim SW, Chung JH, *et al*. High prevalence of vitamin D deficiency in pregnant Korean women: the first trimester and the winter season as risk factors for vitamin D deficiency. Nutrients. 2015;7(5):3427-3448.
88. Jung IK. Prevalence of vitamin D deficiency in Korea: Results from KNHANES 2010 to 2011. Journal of nutrition and health. 2013;46(6):540-551.
89. Nah EH, Kim S, Cho HI. Vitamin D levels and prevalence of vitamin D deficiency associated with sex, age, region, and season in Koreans. Laboratory Medicine Online. 2015;5(2):84-91.
90. Seo JA, Eun CR, Cho H, Lee SK, Yoo HJ, Kim SG, *et al*. Low vitamin D status is associated with nonalcoholic Fatty liver disease independent of visceral obesity in Korean adults. PLoS One. 2013;8(10):e75197.
91. Kim SH, Oh JE, Song DW, Cho CY, Hong SH, Cho YJ, *et al*. The factors associated with Vitamin D deficiency in community dwelling elderly in Korea. Nutrition research and practice. 2018;12(5):387-395.
92. Choi YJ, Kim MK, Jeong SJ. Vitamin D deficiency in infants aged 1 to 6 months. Korean journal of pediatrics. 2013;56(5):205.
93. Anusha K, Hettiaratchi U, Gunasekera D, Prathapan S, Liyanage G. Maternal vitamin D status and its effect on vitamin D levels in early infancy in a tertiary care centre in Sri Lanka. International journal of endocrinology, 2019.
94. Jayatissa R, Lekamwasam S, Ranbanda JM, Ranasingha S, Perera AG, De Silva KH. Vitamin D deficiency among children aged 10-18 years in Sri Lanka. Ceylon Medical Journal. 2019;64(4):146-154.
95. Subasinghe CJ, Gunawardana K, Ediriweera D, Somasundaram NP. Prevalence of vitamin D deficiency/Insufficiency and its metabolic associations in an urban setting in Sri Lanka: Data from Colombo Urban study, 2019.
96. Godawita R, Nayanamali A, Silva R, Nanayakkara N. MO534 Vitamin D Deficiency In Patients With Chronic Kidney Disease Of Uncertain Etiology In Wilgamuwa, Sri Lanka. Nephrology Dialysis Transplantation. 2021;36(Supplement_1):gfab087-0054.