

**Original Article****Formulation of Lichen Based Pill a Natural Source of Vitamin D3 with a High Absorption Rate by Ambrosiya Neo-Medicine Pvt. Ltd**

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## ABSTRACT

*The physiological functioning of the human body depends on vitamin D as a source of minerals. Numerous ailments like bone health, cancer, heart issues, infectious diseases, and CNS are brought on by its shortage. The body's vitamin D insufficiency can be caused by a variety of things, including a person's diet, lifestyle, and geographic location. Blood levels of vitamin D can be maintained through a variety of means, including vitamin D supplements, vitamin D medications, and lifestyle changes.*

*A pharmaceutical company with its main office in India called Ambrosiya Neo-Medicine Pvt. Ltd. has introduced a food-grade vitamin D pill based on lichen to address the global problem of vitamin D deficiency's consequences. In Dr. JAIN's Precision Diagnostic Centre, a comparison study of conventional vitamin D and lichen-based pills was done on ten patients who were vitamin D deficient. The results of the vitamin D level after 48-hours have shown that the lichen-based vitamin D is absorbed at a higher rate than regular vitamin D.*

*We anticipate Ambrosiya's supply of vitamin D supplements made from lichen, and it can make a big difference to the government's attempts to address the problem of vitamin D insufficiency in people everywhere.*

**Keywords:** Lichen based Vitamin D; Natural source; High absorption rate; Herbal product; Food-grade vitamin D pill; Ambrosiya Neo-Medicine Pvt. Ltd

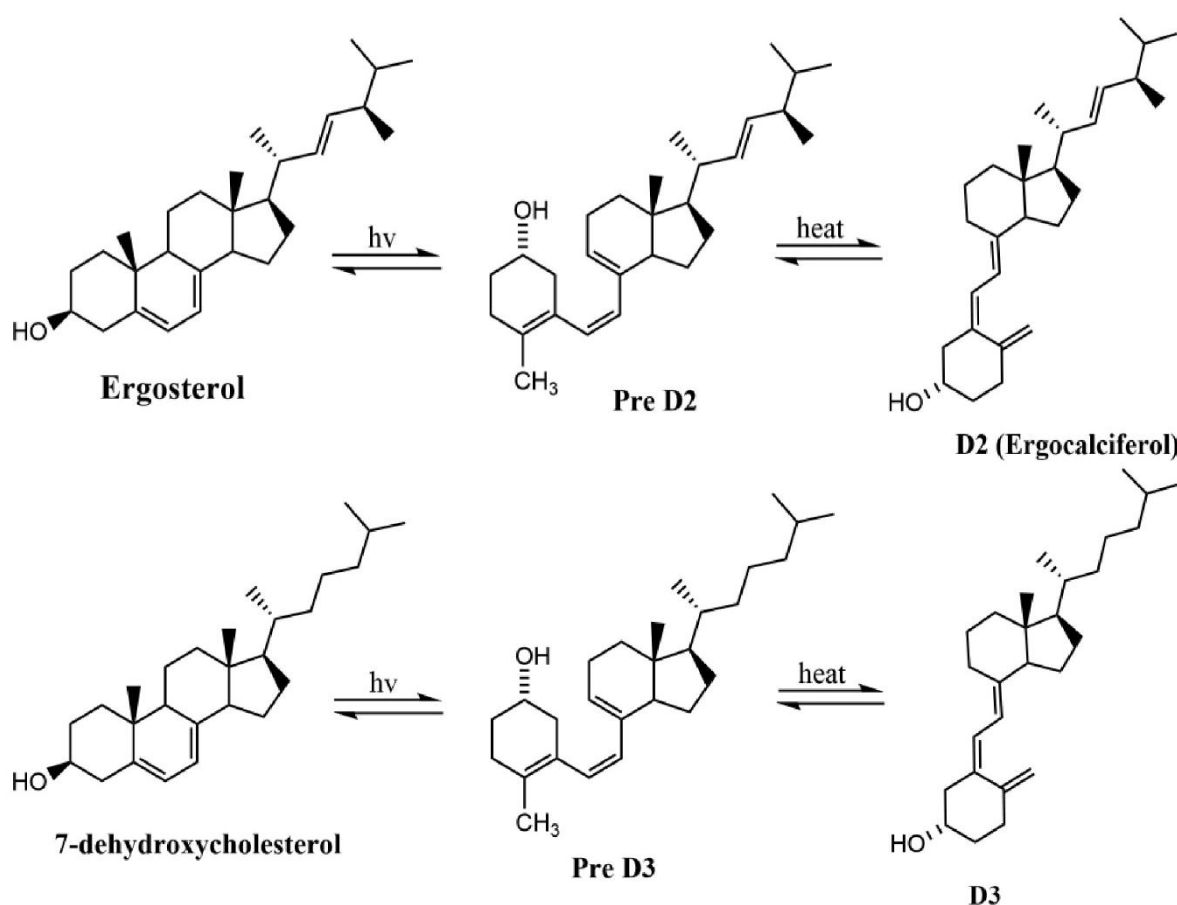
**Introduction**

As a fat-soluble vitamin, vitamin D is essential to maintaining healthy human physiology. It is essential for maintaining appropriate levels in the blood of calcium and phosphate, which are necessary for all body cells to operate normally, including muscular contraction, nerve conduction, and general cellular function. Additionally, it becomes apparent to be crucial for the immune system, inflammation, cell growth, and differentiation [1].

Vitamin D is mostly produced by the skin in the human body following prolonged exposure to ultraviolet light band B (UVB). The skin's 7-dehydrocholesterol is

transformed into the inactive precursor cholecalciferol (Vitamin D<sub>3</sub>). 90% of the total amount of circulating vitamin D is produced by skin exposure to sunshine, while the remaining 10% is obtained through diet (fatty fish, cod liver oil, egg yolk, and fortified dairy products) [2].

The production and metabolism of D<sub>2</sub> and D<sub>3</sub>, In the skin, D<sub>3</sub> is created from 7-DHC through a non-enzymatic process in which UVB radiation breaks the B ring, and the resulting pre-D<sub>3</sub> is then isomerized to D<sub>3</sub> through a thermo-sensitive process (Figure 1).



**Figure 1:** Production of vitamin D2 and D3.

There are several enzymes that convert D<sub>3</sub> to 25OHD<sub>3</sub> in the liver and elsewhere, but CYP2R1 is the most significant. This step's monitoring is, at best, minimal. The active metabolite 1, 25(OH)<sub>2</sub>D<sub>3</sub> is produced after metabolization of 25OHD in kidney and other tissues or the first step in the catabolic process 24,25(OH)<sub>2</sub>D<sub>3</sub>, CYP27B1 and CYP24A1 respectively both enzymes are responsible (Figure 2).

In the kidney, CYP27B1 is promoted by PTH and inhibited by FGF23 as well as high calcium (Ca) and phosphate (P) levels, despite the fact that the regulation varies depending on the tissue. The regulation of CYP24A1 is just the opposite. 1, 25(OH)<sub>2</sub>D<sub>3</sub> also regulates its own production directly and by inhibiting PTH production, stimulating FGF23 production, and inducing CYP24A1. Both UVB (280-320 nm) intensity and skin pigmentation level contribute to the rate of D<sub>3</sub> formation [3].

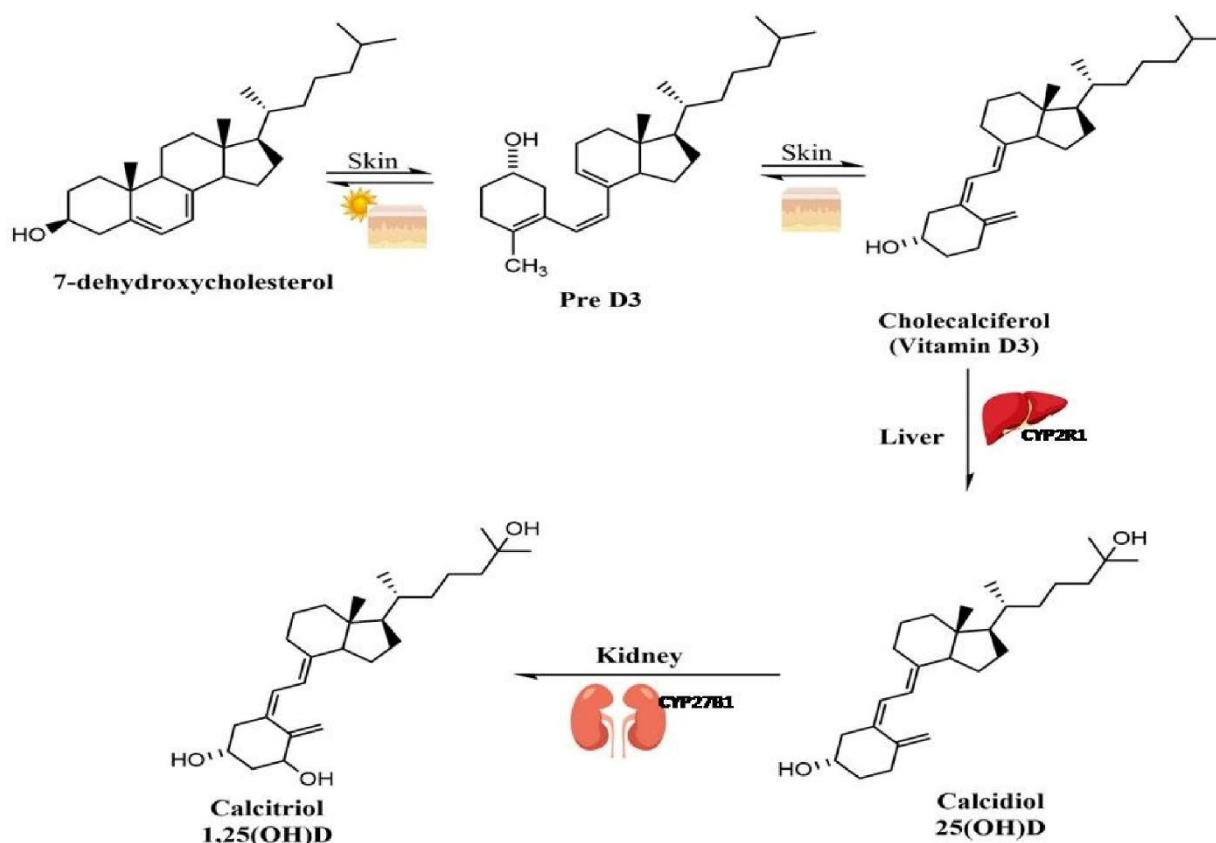
The first hydroxylation, at the C-25 site, happens in the liver, and the second hydroxylation, at the C-1α site, occurs mostly in the kidney. Only after these two hydroxylations does vitamin D<sub>3</sub> become physiologically active. Vitamin D<sub>3</sub> undergoes

hydroxylation in hepatic cells, resulting in its main circulating metabolite [4].

The lack of vitamin D's effects: The different disease which developed due to deficiency of vitamin D is summarized in table 1.

A 2018 study that was published in the Journal of Family Medicine and Primary Care found that up to 90% of Indians may be vitamin D deficient. According to the study, vitamin D insufficiency was more prevalent in urban and rural areas. In other parts of the world, particularly in places with little sun exposure, vitamin D deficiency is a common problem. A 2017 research article in the Journal of Endocrinology and Metabolism found that the prevalence of vitamin D deficiency varies widely around the globe, from 40% to 90%.

In this manuscript, the lichen-based vitamin D manufacturing process and absorption rate are reported. From this study, it is concluded that Ambrosiya Neo-Medicine PVT. LTD. developed this tablet, and the finding of a vitamin D absorption rate after forty-eight hours indicate that it could fight the vitamin D crisis in the world.



**Figure 2: Production and metabolism of vitamin D3**

**Table 1: Different consequences of vitamin D.**

S. No.	Consequences	Cause and effects	References
1	<b>Skeletal manifestations</b>	Adults with osteomalacia and osteoporosis, these disorders are brought on by inadequate vitamin D intake, which results in poor calcium absorption from the diet and excessive calcium outflow from the bones and kidneys. Children who have rickets have poor mineralization of their bones because of an imbalance between calcium and phosphorus in their bones. As a result, there are apparent bone abnormalities. It also causes muscle weakness and bone pain. It is hypothesised that vitamin D might strengthen muscles, preventing falls. Numerous studies have linked low vitamin D levels to an increased risk of fractures and falls in elderly people.	[5,6]
2	<b>Depression</b>	Patients with vitamin D deficiencies needed much more time to recover than others without deficiencies. It means that addressing hypovitaminosis D is crucial for the efficient treatment of depression. In the rodents' and humans' CNS, vitamin D receptor VDR and the enzyme 25-hydroxyvitamin D <sub>3</sub> 1-hydroxylase are present in both neurons and glial cells. Vitamin D may play a role in psychiatric diseases given its significance to numerous brain functions, such as neuroimmunomodulation, oxidative stress, and neuroplasticity.	[7,8]

3	<b>Parkinson's disease</b>	Parkinson's disease (PD) patients had vitamin D deficiencies. Evidence that VDR may be a hereditary risk factor for Parkinson's disease (PD) indicates the possible significance of vitamin D in the disease. Vitamin D functions as a potential preventive/therapeutic method for this disease because vitamin D level is a controllable factor. It has been demonstrated that vitamin D3 administration considerably improves both the motor and non-motor symptoms of Parkinson's disease and improves quality of life.	[9,10]
4	<b>Suicide</b>	In individuals who are suicidal, both peripheral and central inflammation is enhanced. Levels of vitamin D were founded significantly lower than those in healthy controls and depressive patients without suicidality. The psychiatric patients' have high inflammatory markers (specifically IL-1 $\beta$ and IL-6) and low plasma vitamin D levels have a related, reported by researchers. It has been proposed that the brain pathways behind depression and suicidal behaviour directly involve inflammation. Finding and treating these patients' vitamin D insufficiency should be of the utmost significance.	[11,12]
5	<b>Infectious disease</b>	There is increasing confirmation that vitamin D signalling plays a physiological role in defending the human host against viral and bacterial invaders and that it is active across the immune system. The synthesis of cytokines, antimicrobial proteins, and pattern recognition receptors are a few of the mechanisms of vitamin D-innate immunological signalling.	[13]
6	<b>Cancer</b>	Tumour angiogenesis inhibited, promoting cell adhesion to one another, and improving intercellular communication through gap junctions by vitamin D and its metabolites that lower the incidence of many types of cancer. This strengthens the inhibition of proliferation brought on by close physical contact between adjacent cells within a tissue.	[14,15]
7	<b>Autoimmune disease</b>	Autoimmune disorders may begin to develop due to deficient vitamin D showing and/or too little consumption of vitamin D brought on by genetic predisposition (such as VDR polymorphisms) and/or environmental factors (such as not enough sunlight exposure in high-latitude areas or during the cold season). Vitamin D's distinctive capacity to attach to VDR and operate as a transcriptional factor allows it to control gene expression and further the immune cells' immunomodulatory actions. It has been demonstrated to decrease Th1 and promote Th2 cytokine production, inhibit Th17 cytokine production, increase Treg activity, activate NKT cells, and stimulate NKT cell activities, skewing T cells towards Th2 polarisation. A novel role for vitamin D is to modulate immunological processes, having implications for the emergence or avoidance of autoimmune disorders.	[16]

8	<b>Insulin resistance</b>	Decreased insulin sensitivity, decreased insulin turnover, and poor glucose tolerance are all symptoms of vitamin D insufficiency. In addition, vitamin D repletion enhances glucose clearance in vitamin D-deficient mice without regard to other dietary components.	[17,18]
9	<b>Diabetes</b>	It has been demonstrated that calcitriol has a role in the production and secretion of insulin, the control of calcium trafficking in beta islet cells, and the effects it has on the activity of insulin. Insufficient amounts of serum 25-hydroxyvitamin D are related with increased risks, however confounding by obesity and behavioural variables cannot be completely ruled out.	[19]
10	<b>Cardiovascular Diseases</b>	Cells all over the entire circulatory system express the vitamin D receptor. The enzyme 1-hydroxylase is produced by a variety of cell types, including vascular smooth muscle cells, endothelial cells, and cardiomyocytes, and it transforms 25-hydroxyvitamin D into calcitriol, the organic ligand for the vitamin D receptor. It has been demonstrated that calcitriol possesses anti-inflammatory characteristics, regulates the renin-angiotensin system, inhibits the growth of vascular smooth muscle cells, and decreases coagulation.	[20]

## Experimental Work

### Materials

Lichen samples (*Bulbothrix Setschwanensis*) were collected from the hills of Uttarakhand region, India. Liquid CO<sub>2</sub>, Extraction vessel, High-pressure pump, Back pressure regulator, Collection vessel, Solvent trap, Rotary evaporator were used to carry out the extraction. Analytical balance was used to measure weight of sample and the obtained extract. UV-Vis spectrophotometer was used to determine the concentration of vitamin D3 in the extract.

Lichen Extract based Vitamin D3 (Food Grade) was developed by Ambrosiya Neo Medicine Pvt. Ltd. The extract samples contain *Bulbothrix Setschwanensis* (Zahlbr). Hale was identified by Dr. Lalit M. Tiwari (Professor), Department of Botany, D.S.B. Campus, Kumaun University, Nainital (Uttarakhand), 263002, India.

### Method of Preparation

#### 1. Preparation of Lichen Samples:

High quality lichen samples rich in vitamin D3 content were selected. Lichen samples were cleaned by removing any visible impurities or debris from them. It was then powdered finely using mortar and pestle or a

grinder. 1kg of powdered sample was transferred to the extraction vessel.

#### 2. Supercritical Fluid Extraction:

The extraction vessel was closed and filled with liquid CO<sub>2</sub>. Pressure inside the extraction vessel was increased to 300-350 bar using the high-pressure pump. Extraction vessel was heated to 50-60°C and this temperature and pressure was maintained for 2 hours to allow the CO<sub>2</sub> to extract the vitamin D3 from the lichen sample. A back pressure regulator was used to lower the pressure and allow the CO<sub>2</sub> to flow into the collection vessel. The extraction process was repeated with fresh CO<sub>2</sub> until the desired amount of vitamin D3 is extracted. Typically, 70-80 g of extract (Based on lichen quality) can be obtained from 1 kg of lichen powder.

#### 3. Solvent Recovery:

CO<sub>2</sub> and vitamin D3 mixture was collected in the collection vessel then transferred to a solvent trap to separate the CO<sub>2</sub> from the vitamin D3. Rotary evaporator was used to evaporate the remaining CO<sub>2</sub> and obtain the Lichen based vitamin D3 extract.

#### 4. Analysis:

Lichen vitamin D3 extract was weighed using an analytical balance. The concentration of vitamin D3 in



the extract was determined using a UV-Vis spectrophotometer. Which contain around 23,000 IU/gm of Lichen based vitamin D3 extract.

Ambrosiya has created a special method for extracting and manufacturing vitamin D3 from lichen, allowing them to create premium supplements at a competitive price. Their tablets don't include any artificial colours or preservatives, are vegan, or contain any gluten.

### Clinical Study

Lichen Extract- based Vitamin D3 (Food Grade) was developed in Ambrosiya Neo-Medicine PVT. LTD., C/O Karan Joshi, H.N 64, Cheenpur, Kusumkhara, Haldwani, Distt. Nainital, 263139, India.

Each gram of Lichen-Based Vitamin D3 extract contains 23,000 IU/gm (0.580 mg/gm or 580.237mcg/gm) of Vitamin D, making it the cheapest potent and absorbable source of Vitamin D.

A study was conducted in DR. JAIN'S PRECISION DIAGNOSTIC CENTRE. Ten patients which have deficiency of Vitamin D were first identified. Out of ten five patients (P1-P5) were given Normal Vitamin D (Marketed product) and the remaining five patients (P6-P10) were given Lichen-Based Vitamin D3. After 48 hours the absorption of lichen-based Vitamin D3 in patients (P6-P10) was higher observed.

Vitamin D normal range is 30-100 mg/ml, deficiency below 20 mg/ml, insufficiency 20-30 mg/ml, and toxicity above 100 mg/ml.

### Discussion

Rickets, osteomalacia, and osteoporosis are health issues linked to vitamin D insufficiency. Additionally, it has been connected to a higher risk of a number of illnesses, including autoimmune disorders, diabetes, cardiac issues, infectious infections, and several forms of cancer. Limited sun exposure, dark complexion, obesity, advanced age, and certain illnesses like inflammatory bowel disease and kidney disease are risk factors for vitamin D deficiency [21].

Depending on age and location, a daily dosage of vitamin D is advised. The Recommended Dietary Allowance (RDA) for adults in India is 400–600 IU of vitamin D per day, whereas in the US, it is 600–800 IU. Sunlight exposure, eating foods high in vitamin D, and taking vitamin D supplements are the three basic approaches to combat vitamin D insufficiency [22].

When skin is exposed to sunlight, the body naturally creates vitamin D. Sunlight exposure aids in the transformation of a skin substance called 7-dehydrocholesterol into Vitamin D<sub>3</sub>. Depending on characteristics including skin tone, latitude, and time of day, a person may require a different quantity of sun exposure to create adequate Vitamin D. But getting 5–30 minutes of sun exposure to the face, arms, legs, or back twice a week is advised [23].

Several foods naturally contain vitamin D, such as egg yolks, beef liver, and fatty fish like salmon, mackerel, and tuna. Vitamin D is added to several meals like milk, orange juice, and cereal. For those who get little sun exposure or can't take vitamin D pills, eating these foods can help enhance vitamin D consumption.

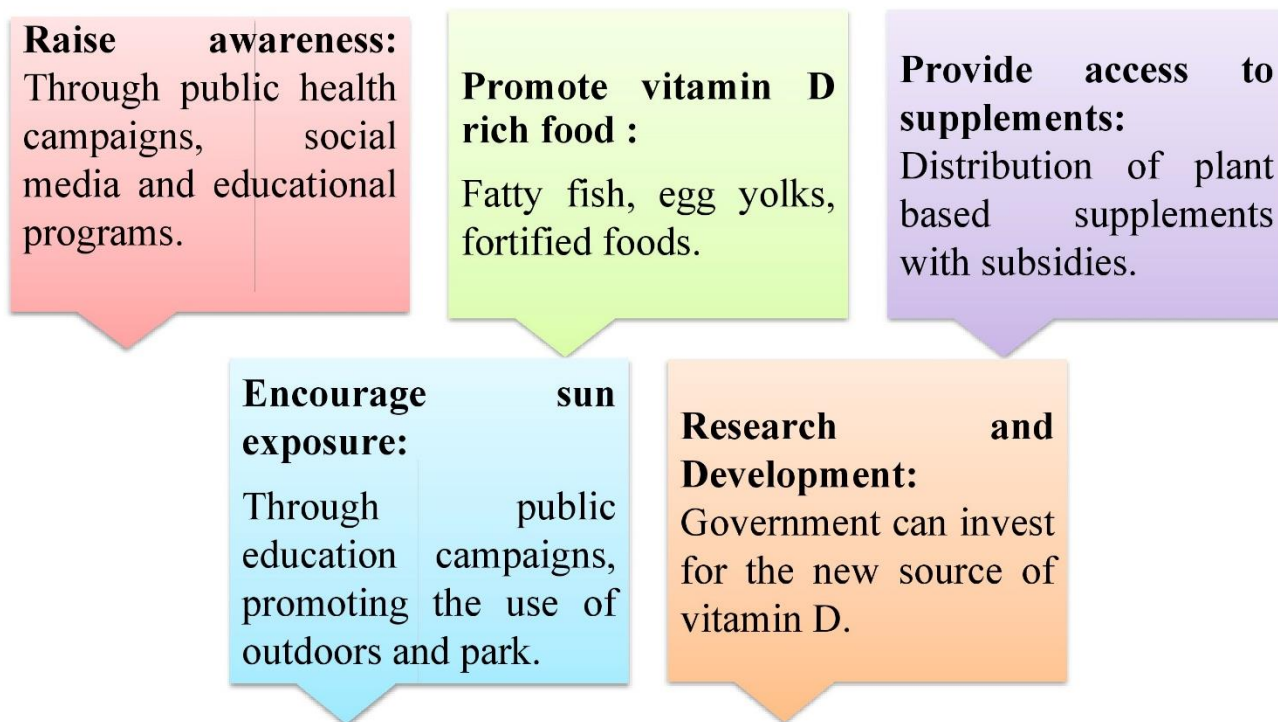
There are two types of vitamin D supplements: vitamin D2 and vitamin D3. The more powerful vitamin D3 is the one that is made in the skin when it is exposed to sunshine. There are many different dosages of supplements, ranging from 400 IU to 5000 IU per day. The necessary dosage will vary according on a person's age, health, and vitamin D levels [23].

Vitamin D supplements made from plants, such as lichen extract pills, can aid in the fight against global and Indian vitamin D insufficiency. Fungi that grow on rocks, trees, and other surfaces are known as lichens. Several lichen species are organic sources of vitamin D. People who don't consume animal products, such as vegetarians and vegans, can acquire a rich supply of plant-based vitamin D by taking lichen extract pills.

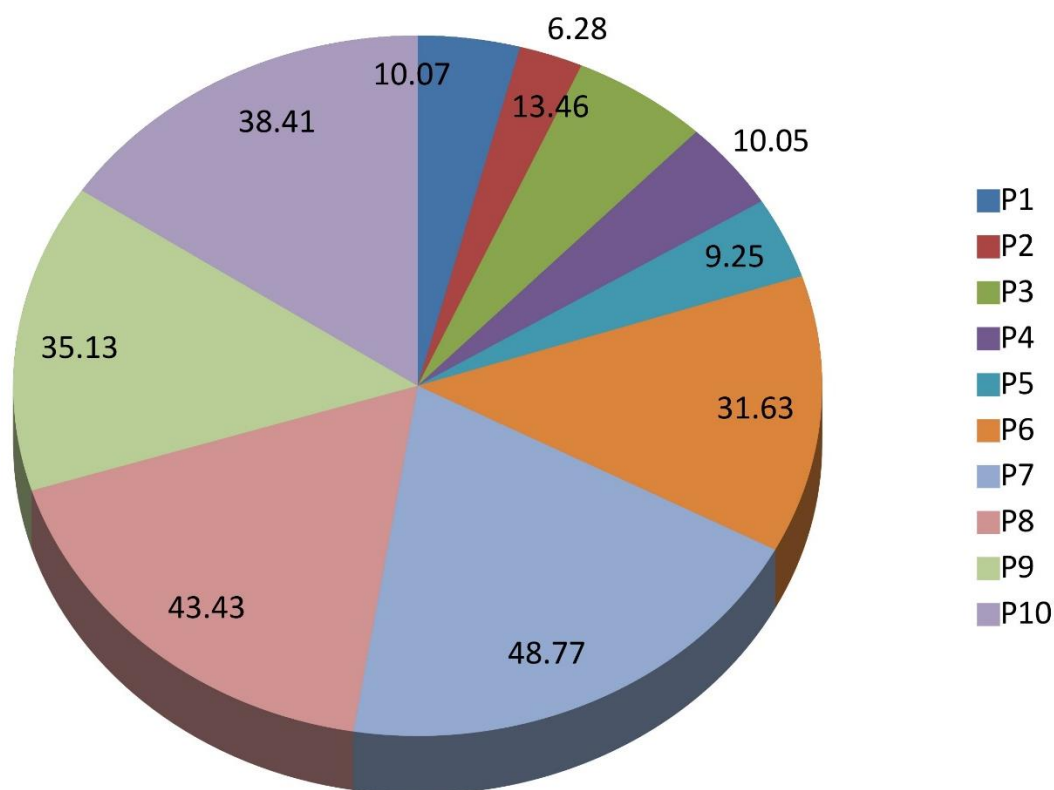
Vitamins D3 and D2 are both present in lichen extract tablets. Vitamin D2 is less effective than vitamin D3, vitamin D2 is still effective at increasing the level of vitamin D in the blood. Lichen extract tablets provide vitamin D3 that absorbs similarly to animal sources of vitamin D3.

Vitamin D supplements made from plants are generally well-tolerated and secure. For those who are lactose intolerant or allergic to dairy products, they are a good substitute. Tablets containing lichen extract are a reliable source of vitamin D. They can be produced without the use of animal products, and their growth is environmentally friendly. Vitamin D supplements made from plants are practical and simple to take. They can be taken with or without food and come in tablet form [24-40].

Government and different NGO can approach the population to fight vitamin deficiency diseases with various approaches (Figure 3).



**Figure 3:** Fight Vitamin D Deficiency: Government approach.



**Figure 4:** Percentage of vitamin D3 level in blood after 48 hours.

## Conclusion

Our body needs vitamin D, a vital mineral, for a variety of processes, including the functioning of the immune system, good bone condition, and mental well-being. However, a lot of people lack enough vitamin D, which can cause a number of health issues. Inadequate sun exposure, dietary limitations, and other reasons may all contribute to this insufficiency. Ambrosiya Neo-Medicine PVT. LTD. has developed a solution for this issue.

Launched Lichen-Based Vitamin D3, the most efficient, cost-efficient, and environmentally friendly form of Vitamin D3. This vitamin D3 version is the simplest to use and is vegan, vegetarian, and all-natural. Lichen-Based Vitamin D3 is a vegan source of Vitamin D3 that is extracted from lichens, a combination of algae and fungus. A 60,000 IU of chemical based which equivalent to 2.6 grams of Lichen-Based Vitamin D3 extract contains 60,000 IU of Vitamin D3 were given to the patients have vitamin D deficiency, making it the most cheaply potent and absorbable source of Vitamin D3. Lichen-Based Vitamin D3 is the ideal source of vitamin D for vegans and vegetarians because it is natural, sustainable, and free of animal ingredients. This product is toxin and contamination free and is the cheapest source of Vitamin D, making it accessible to everyone who needs it. The finding of comparative study of normal vitamin D3 and Lichen based D3 indicated that the rate of absorption of vitamin D3 in lichen based compared to normal vitamin is higher (Figure 4).

A sustainable source of vitamin D that doesn't hurt the environment or use animal products is lichen-based vitamin D3. We can produce 1800 corer tablets of lichen-based vitamin D3 per quarter, assuring a steady and on-going supply.

Lichen-Based Vitamin D3 is performed by giving the body the essential quantity of Vitamin D for a variety of processes, including bone health, immune system, and mental wellness. Lichen-based Vitamin D3's potency and absorbability make sure that the body can efficiently absorb the mineral, lowering the danger of Vitamin D insufficiency and the related health issues. Because this medicine is accessible as a tablet, it is simple to use and included in your everyday routine. In addition to enhancing your health, choosing lichen-based vitamin D3 also helps a sustainable and vegan-friendly source of the vitamin.

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## Conflict of Interest

The author declares no conflict of interest.

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