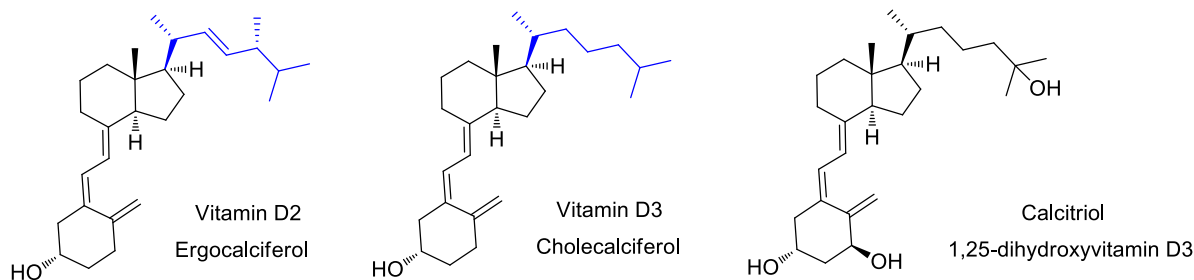


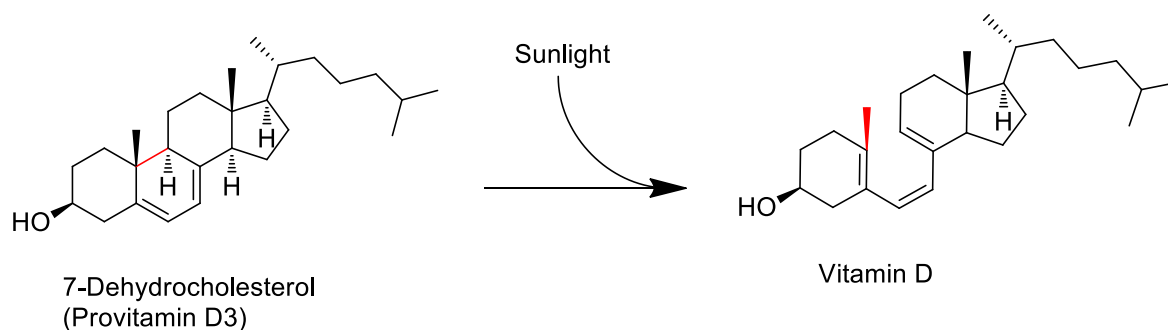
Chemical Identity & Function of Vitamin D

Vitamin D, also referred to as calciferol, is a fat-soluble vitamin that promotes calcium absorption by ensuring that the body maintains proper concentrations of serum calcium and phosphate which is necessary for bone growth.¹ The IUPAC name for vitamin D is (3 β ,5Z,7E)-9,10-secocholesta- 5,7,10(19)-trien-3-ol.^{1,5} This vitamin has two forms, vitamin D₃ and vitamin D₂ also known as cholecalciferol and ergocalciferol, respectively (Scheme 1). Of the two different forms of vitamin D, studies have proven vitamin D₃ to be the most potent and effective form of vitamin D in humans in regards to effectively binding to vitamin D receptors.² The mechanism for vitamin D₃ includes the synthesis from pro-vitamin D₃ and ultraviolet light from the sun (Scheme 2). Vitamin D is often metabolized and stored more commonly in the blood, liver, kidney, lungs, skin and other body tissues.² Studies show that detectable levels of vitamin D can circulate in the blood stream for two to three months.² The recommended daily amount is 10 mcg (400 IU) for children under one year old, 15 mcg (600 UI) for those between 1-70 years old, and 20 mcg (600 UI) for those over 71 years old.¹ Adequate levels of vitamin D can prevent rickets in children. Vitamin D in conjunction with calcium is effective in osteoporosis prevention in older adults.¹ The upper level, UL, for vitamin D intake is 100 mcg (4000 IU). While the body cannot overproduce vitamin D, excessive consumption in a short period of time can have several harmful side-effects.³ These side effects can include headaches, vomiting, and loss of appetite. If taken in excess for prolonged periods of time, vitamin D can cause high levels of calcium in the blood which, in turn, can lead to kidney stones, kidney disease, and even atherosclerosis (hardening of the arteries).³

Scheme 1. Chemical Structure of Calcitriol, Vitamin D₂ and Vitamin D₃



Scheme 2. Mechanism of Action for Vitamin D₃



- (1) Vitamin D Health Sheets for Health Professionals. (<http://ods.od.nih.gov/factsheets/VitaminD-HealthProfessional/>) Accessed 3/3/2014
- (2) Heymann, W. Metabolism and Mode of Action of Vitamin D. *J. Biol. Chem.* **1937**, 118, 371-376.
- (3) *Vitamin D*. (<http://www.webmd.com/vitamins-supplements/ingredientmono-929-VITAMIN%20D.aspx?activeIngredientId=929&activeIngredientName=VITAMIN%20D>) Accessed 3/3/2014

Spectroscopic Characterization of Vitamin D

The ^1H -NMR and ^{13}C -NMR spectra recorded for vitamin D_3 are shown in Figures 1 and 2, respectively. Both NMR spectra were taken on a 500 MHz spectrometer with CDCl_3 as the solvent. In the ^1H -NMR spectrum, two peaks are observed at 0.5 and 0.9 ppm which are indicative of the two C-H hydrogens. The ^{13}C -NMR spectrum shows C=C double bond peaks from 100-150 ppm; cholecalciferol contains 3 C=C double bonds as observable in the ^{13}C -NMR. An ultraviolet absorbance spectrum with an alcohol reagent indicates that vitamin D has absorbance between 200 and 320 nm (Figure 3). The highest peak wavelength is at 270 nm with the highest absorbance value of 0.58. The second highest peak is observed at 218 nm with an absorbance value of 0.50. Looking at an IR spectrum, the hydroxyl group is indicated by the O-H stretch at 3400 cm^{-1} (Figure 4). The peaks found at 1610 , 1620 , and 1700 cm^{-1} are indicative of the alkenes. At 2900 cm^{-1} , an alkane H peak is observed. These pertinent observations support the hypothesis that the spectra samples can be identified as vitamin D_3 .

Figure 1. ^1H -NMR Spectrum of Vitamin D_3 ⁴

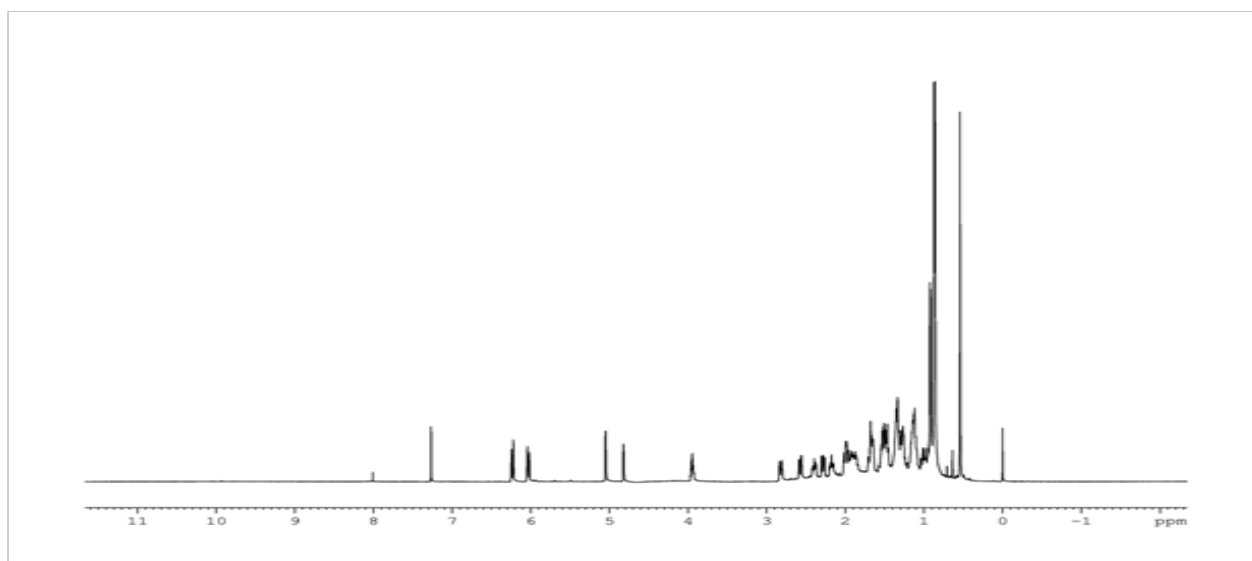


Figure 2. ^{13}C -NMR Spectrum of Vitamin D₃⁴

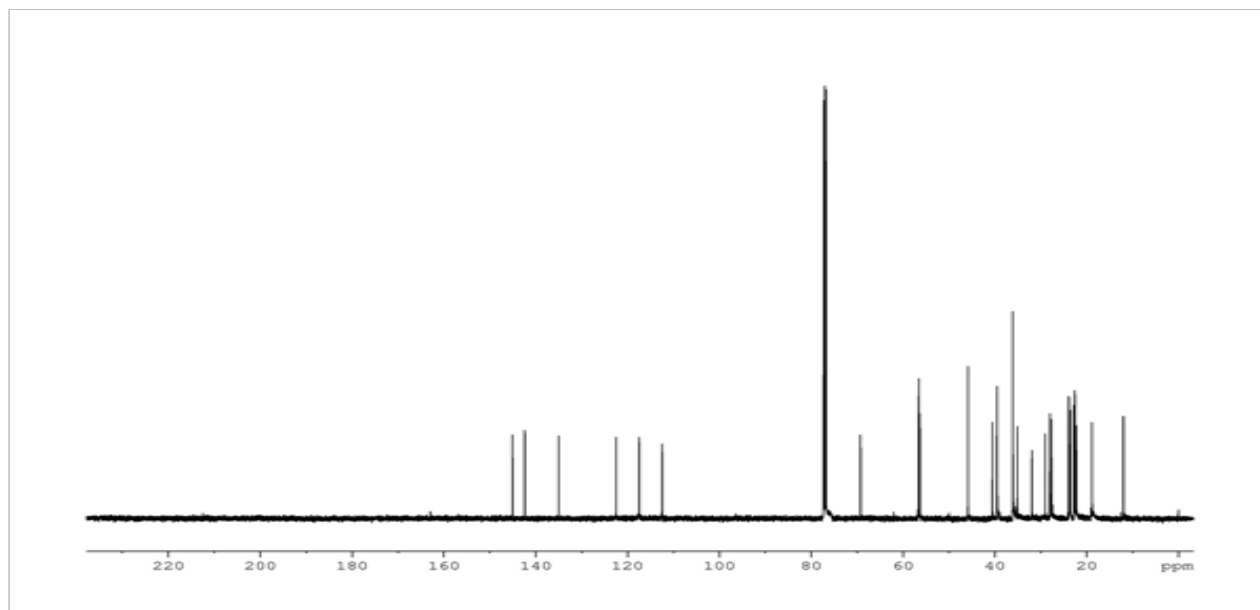


Figure 3. UV/Vis Spectrum of Vitamin D₃⁵

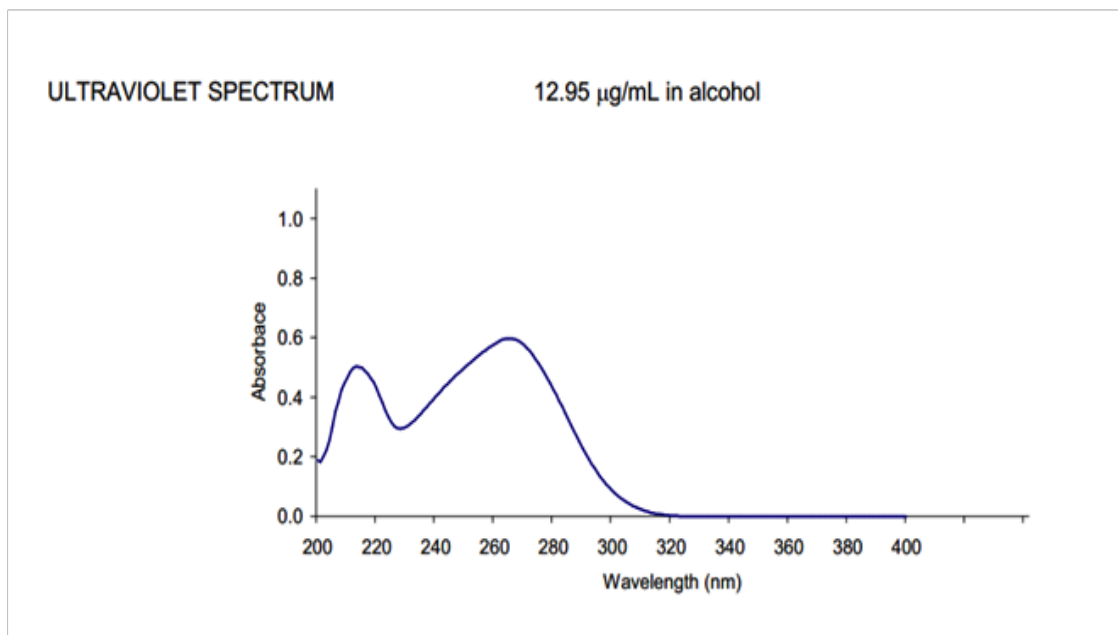
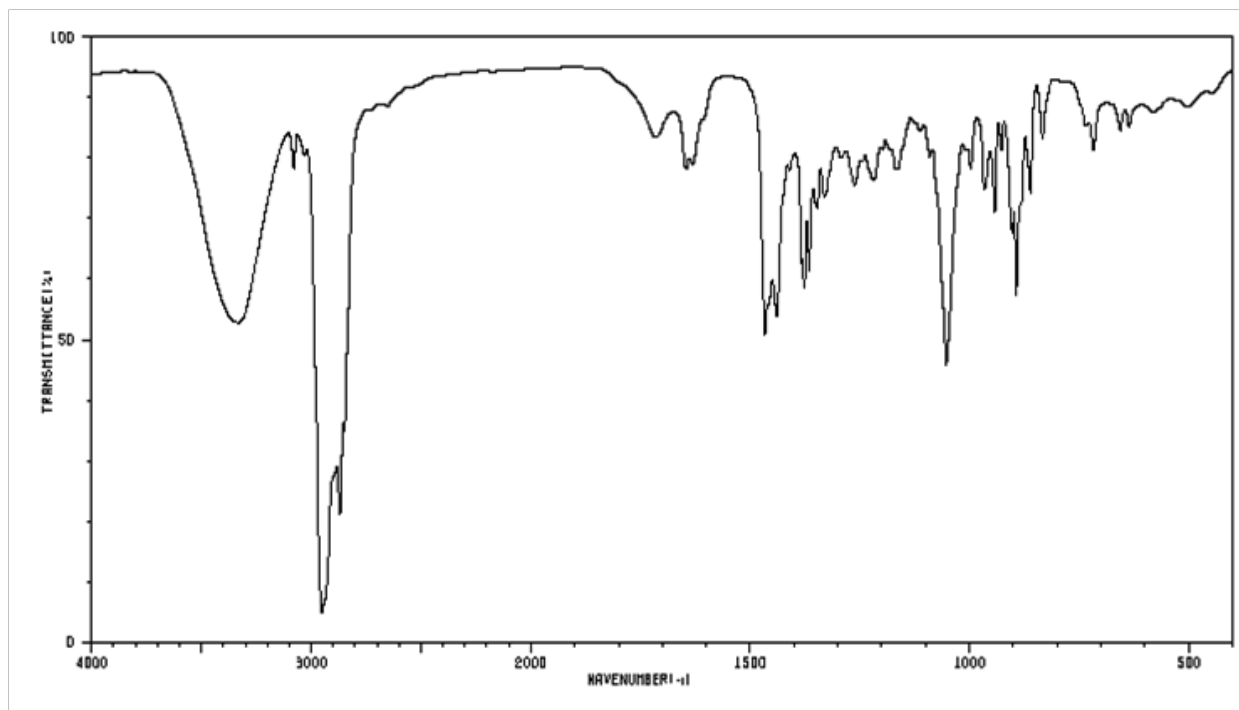


Figure 4. IR Spectrum of Vitamin D₃⁶

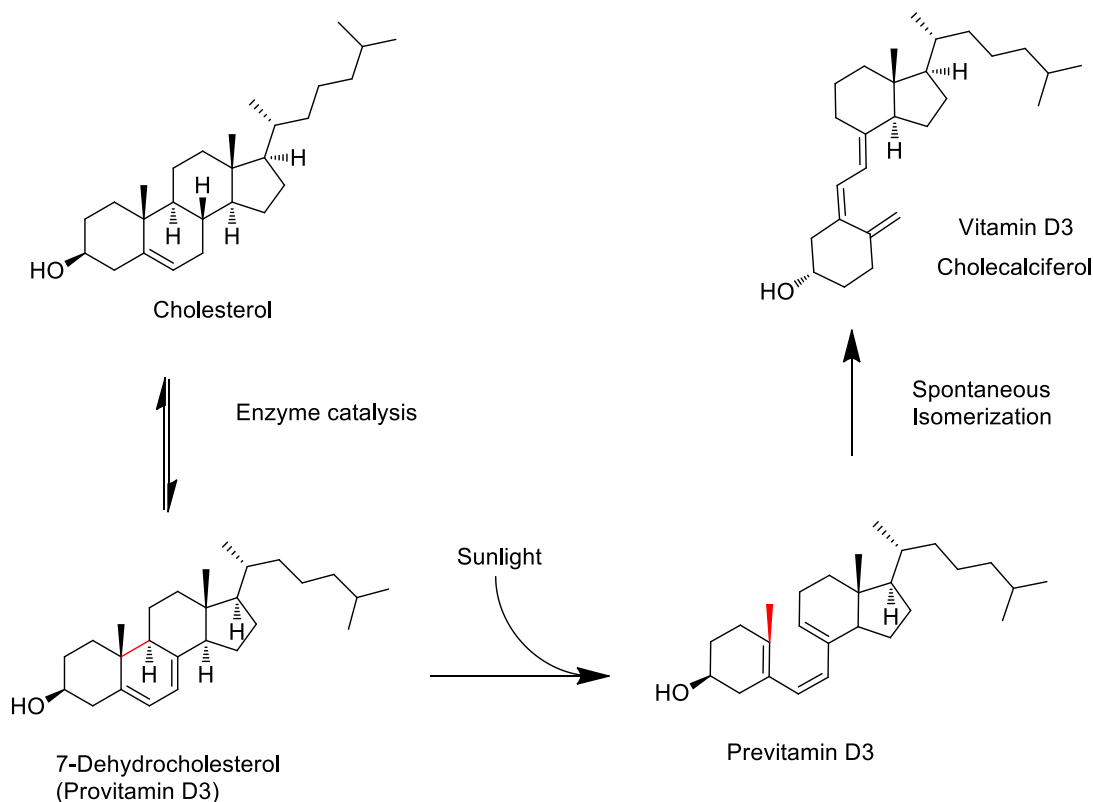


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- (4) ¹H-NMR and ¹³C-NMR Spectra for Vitamin D₃. *Biological Magnetic Resonance Data Bank*. http://www.bmrwisc.edu/metabolomics/mol_summary/show_data.php?molName=cholecalciferol&id=bmse000507. Accessed 3/3/2014
- (5) Cholecalciferol Product Information. <http://www.sigmaaldrich.com/content/dam/sigmaaldrich/docs/Sigma-Aldrich/Datasheet/c9774-076k8807dat.pdf>. Accessed 3/3/2014

Structure Search & Synthesis of Vitamin D

Vitamin D₃ is synthesized in a naturally occurring reaction where 7-dehydrocholesterol (pro-vitamin D) reacts with sunlight (specifically UV B radiation) to form what is called pre-vitamin D₃. Pre-vitamin D₃ undergoes spontaneous isomerization to form Vitamin D₃. This is then extracted from either plant or animal products and used in supplements. While the reaction from pro-vitamin D to vitamin D₃ is well defined, the steps leading up to synthesis are not quite as well known. Most research suggests that there is an enzymatic reaction that oxidizes cholesterol into 7-dehydro-cholesterol (Scheme 3).⁶ This reaction can also go backwards in order to prevent the buildup of excess vitamin D, by converting the pro-vitamin to inactive metabolites. It is also possible to increase the levels of pro-vitamin D through inhibition of enzymes such as 7-dehydrocholesterol reductase which would convert pro-vitamin D into other sterols.⁷ The mechanism by which cholesterol is converted to pro-vitamin D is still being researched as well as the location in the body at which it takes place. Some researchers have suggested that this reaction takes place in the intestinal wall, however due to the large concentration of 7-dehydrocholesterol reductase in the intestines, pro-vitamin D would likely rapidly be converted back to cholesterol.^{6,7} While lacking concrete proof, the reaction most likely occurs in the skin.

Scheme 3. Synthesis of Vitamin D₃ from Cholesterol



-Illustrates the predicted synthesis pathway that cholesterol undergoes to form Vitamin D₃⁸⁹

Leading References

- (6) Masterjohn, C. Vitamin D is Synthesized from Cholesterol and Found in Cholesterol Rich Foods. Accessed 3/3/2014
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- (9) Keverling Buisman, J. A.; Stevens, W.; Vliet, J. V. D. Investigations on sterols: I. A new synthesis of 7-dehydrocholesterol (provitamin D). *Recl Trav Chim Pay B* **2010**, *66*, 83-92.