Studies of the Antenna Effect in Polymer Molecules. 24. Solar Photosynthesis of Previtamin D_3 in Aqueous Solutions of Poly(sodium styrenesulfonate-*co*-2-vinylnaphthalene)

Maria Nowakowska,† Victor P. Foyle,‡ and James E. Guillet*

Contribution from the Department of Chemistry, University of Toronto, Toronto, Ontario, Canada M5S 1A1

Received November 16, 1992

Abstract: The photochemical step in the synthesis of vitamin D_3 in an aqueous solution of poly(sodium styrenesulfonate-*co*-2-vinylnaphthalene) (PSSS-VN) was studied using solar light. The photoisomerization of 7-dehydrocholesterol to previtamin D_3 was found to be exceptionally clean and efficient. The process was shown to be photosensitized by the pendant naphthalene chromophores in the copolymer via singlet–singlet energy transfer. The lack of tachysterol, a major side product formed during photoisomerization in organic solution, was explained by triplet-sensitized isomerization of tachysterol to previtamin. Triplet–triplet energy transfer from excited polymeric naphthalene chromophores to tachysterol is efficient because of the high local concentration of tachysterol in the vicinity of the energy donors.

Introduction

In previous papers in this series, it has been reported that novel water-soluble antenna polyelectrolytes, referred to as "photozymes", act as efficient photocatalysts.[1-10] Repulsive interactions between water and large, hydrophobic, aromatic chromophores present in the macromolecules induce conformations in these polymers resulting in the formation of hydrophobic microdomains. Due to the presence of these microdomains, the polymers solubilize molecules of sparingly water-soluble organic compounds. The aromatic chromophores act as antennas by absorbing light from the near UV–visible spectral region and transferring this excitation energy to the solubilized molecules which may then undergo photochemical reactions. Photozymes have been shown to sensitize several different types of photochemical reactions such as photolysis of undecanone,[1] photooxidation of polynuclear aromatics,[2] 1,3-diphenylisobenzofuran,[4] and styrene,[8] photodechlorination of polychlorinated biphenyls,[9] and photoisomerization of *trans*-stilbene[10] in water solution.

The photochemistry of isomers belonging to the vitamin D family has been the subject of extensive studies for many years.[11-21] This attention originates from scientific interest and also reflects the important biological and medical roles these compounds play. The synthesis of vitamin D is one of the few examples of photochemical reactions of commercial importance.[22] The most important reactions are illustrated in Scheme I based on studies reported by Jacobs and Havinga[11c] in solution at <0 °C. The numbers associated with the arrows represent the relevant quantum yields. The overall process involves two steps: the first photochemical, the second thermal. In the first step, a starting material, 7-dehydrocholesterol (DHC), undergoes photochemical conrotatory ring opening of the cyclohexadiene system to form the (*Z*)-hexatriene derivative, previtamin D, which can then be thermally converted to *cis*-vitamin D by a reversible 1.7 H shift. Unfortunately, the previtamin also undergoes a photochemically induced back ring closure, which both regenerates DHC and creates the 9,10 anti isomer side product, lumisterol. It also displays $Z \rightarrow E$ photoisomerization to the 6,7 isomer, tachysterol. All these photochemical reactions are reversible and all, except $Z \rightarrow E$ isomerization, can occur in the corresponding molecule only through the first excited singlet state. The previtamin $\rightleftharpoons$ tachysterol $Z \rightleftharpoons E$ photoisomerization can occur both in the singlet and triplet states of these molecules. Because intersystem crossing in alkenes is very inefficient, the triplet state can only be significantly populated by energy transfer from a properly chosen donor. Fluorenone and anthracene have been shown to be efficient triplet sensitizers for tachysterol.[16,23] It also shows the thermal step in the formation of vitamin D but does not include other photochemical processes of vitamin D which can be important at higher temperatures where rapid thermal isomerization produces significant quantities of the vitamin. Irreversible

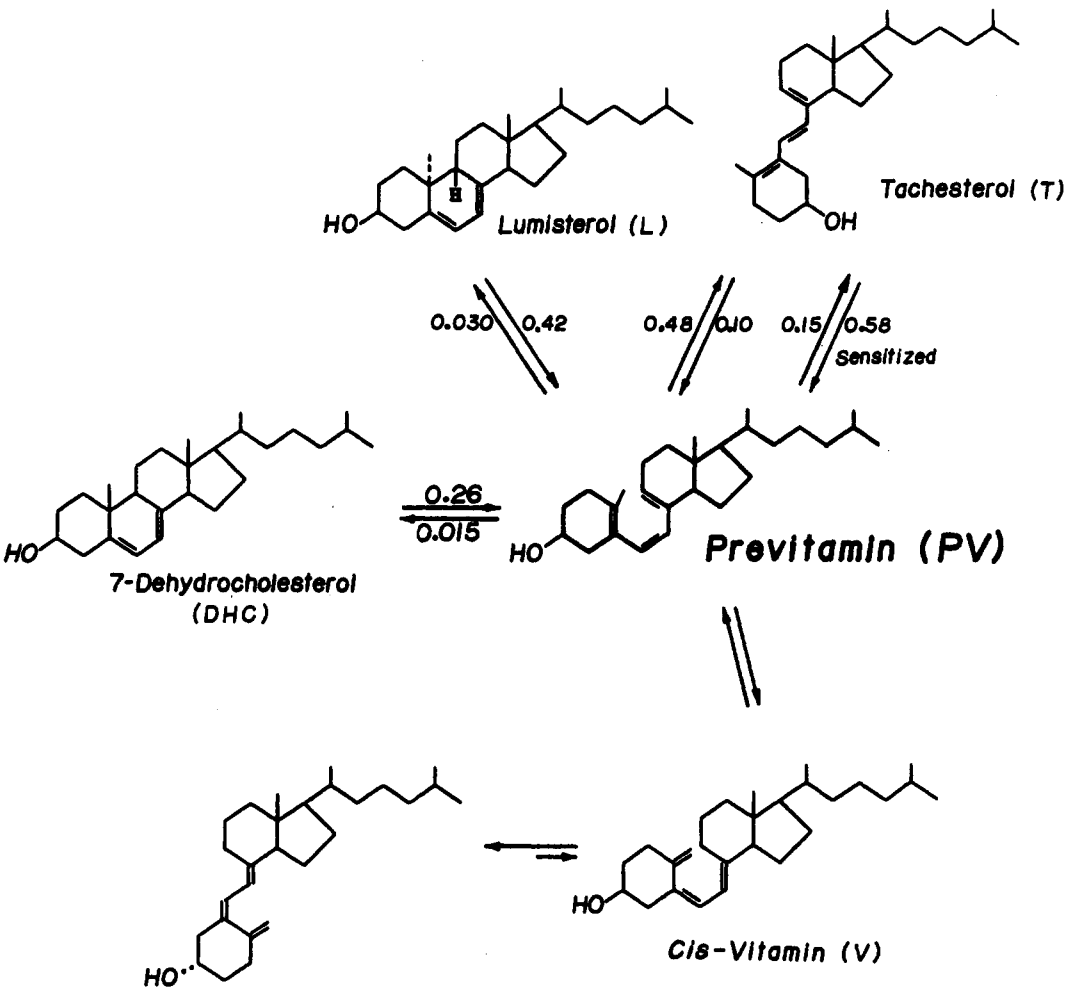
† Current address: Faculty of Chemistry, Jagiellonian University, Krakow, Poland.

‡ Current address: Surpass Chemicals Ltd., 10 Chemical Court, West Hill, Ontario, Canada M1R 4C7.

(1) Nowakowska, M.; White, B.; Guillet, J. E. *Macromolecules* **1988**, *21*, 3430.
(2) Nowakowska, M.; White, B.; Guillet, J. E. *Macromolecules* **1989**, *22*, 2317.
(3) Nowakowska, M.; White, B.; Guillet, J. E. *Macromolecules* **1989**, *22*, 3903.
(4) Nowakowska, M.; Guillet, J. E. *Univ. Jagiell. Acta Chim.* **1991**, *34*, 37.
(5) White, B.; Nowakowska, M.; Guillet, J. E. *J. Photochem. Photobiol., A* **1989**, *50*, 147.
(6) Sustar, E.; Nowakowska, M.; Guillet, J. E. *J. Photochem. Photobiol., A* **1990**, *53*, 233.
(7) Nowakowska, M.; White, B.; Vogt, S.; Guillet, J. E. *J. Polym. Sci., Part A: Polym. Chem.* **1992**, *30*, 271.
(8) Nowakowska, M.; Guillet, J. E. *Macromolecules* **1991**, *24*, 474.
(9) Nowakowska, M.; Sustar, E.; Guillet, J. E. *J. Am. Chem. Soc.* **1991**, *113*, 253.
(10) Nowakowska, M.; Storsberg, J.; Guillet, J. E. Manuscript in preparation.
(11) See reviews and citations therein: (a) Senders, G. M.; Pot, J.; Havinga, E. *Fortschr. Chem. Org. Naturst.* **1969**, *27*, 131. (b) Havinga, E. *Experientia* **1973**, *29*, 1181. (c) Jacobs, H. J. C.; Havinga, E. *Adv. Photochem.* **1979**, *11*, 305.
(12) Windaus, A.; Hess, A.; Rosenheim, O.; Pohl, R.; Webster, T. A. *Chem. Ztg.* **1927**, *51*, 113.
(13) Havinga, E.; Koevoet, A. L.; Verloop, A. *Recl. Trav. Chim. Pays-Bas* **1955**, *74*, 1230.
(14) Havinga, E.; De Kock, R. J.; Rappoldt, M. P. *Tetrahedron* **1960**, *11*, 276.
(15) Snoeren, A. E. C.; Daha, M. R.; Lugtenburg, J.; Havinga, E. *Recl. Trav. Chim. Pays-Bas* **1970**, *89*, 261.
(16) Eyley, S. C.; Williams, D. H. *J. Chem. Soc., Chem. Commun.* 1975, 858.
(17) von Pfoertner, K.; Weber, J. P. *Helv. Chim. Acta* **1972**, *55*, 921.
(18) Jacobs, H. J. C.; Gielen, J. W. J.; Havinga, E. *Tetrahedron Let.* **1981**, *22*, 4013.
(19) Malatesta, V.; Willis, C.; Hackett, P. A. *J. Am. Chem. Soc.* **1981**, *103*, 6781.
(20) Gliesing, S.; Reichenbächer, M.; Ilge, H.-D.; Fassler, D. *J. Prakt. Chem.* **1987**, *2*, 311.
(21) Gorman, A. A.; Hamblett, I.; Lambert, C.; Prescott, A. L. *Photochem. Photobiol.* **1990**, *51*, 29.
(22) Pfoertner, K. H. In *Photochemistry in Organic Synthesis*; Coyle, J. D., Ed.; The Royal Society of Chemistry: London, 1986.
(23) Stevens, R. D. S. U.S. Patent 4,686,023, 1987.

0002-7863/93/1515-5975$04.00/0

Scheme I



photoreactions of vitamin D in long-term irradiation result in the formation of so-called "overirradiation products". The cited values for the quantum yields were found to be the same, within experimental error, as those for reactions initiated with radiation at 254 or 313 nm.[11c]

Scheme I presents only part of the photochemistry of vitamin D isomers since the process is quite complicated, and it is practically impossible to obtain a pure, final product or even to generate a product at high conversion by direct irradiation of DHC. Interestingly, although each of the isomers has a distinct absorption spectrum in the UV region,[13,20] it is not possible to excite each selectively due to their spectral overlap (see Figure 1).

This paper reports studies of the photochemical step in the synthesis of vitamin D_3 in a medium consisting of an aqueous solution of antenna polyelectrolyte poly(sodium styrenesulfonate-*co*-2-vinylnaphthalene) (PSSS-VN) using solar light. The aim of this report is to present the new concept of applying photozymes as microphotoreactors in which solar energy can be used to drive useful and specific syntheses of organic compounds in water solution.

Experimental Section

Materials. Poly(sodium styrenesulfonate-*co*-2-vinylnaphthalene) was synthesized and purified according to the method previously described.[1] The polymer consisted of about 60 mol % 2-vinylnaphthalene and 40 mol % sodium styrenesulfonate groups.

7-Dehydrocholesterol (Aldrich, 98%) and lumisterol (kindly provided by Dr. K. Pfoertner, Hoffman-La Roche & Co., Basel) were recrystallized three times from ethyl acetate in an argon atmosphere. Vitamin D_3 (Aldrich, 99+%) was used as received. Naphthalene (Aldrich, 99%) and *trans*-stilbene (Aldrich, 96%) were purified by triple crystallization from methanol.

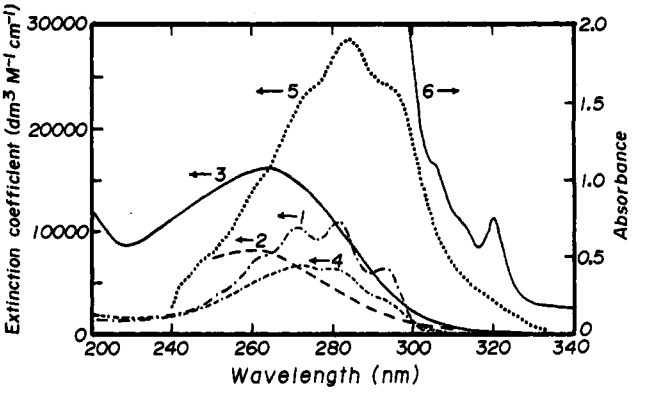


Figure 1. UV absorption spectra of (1) DHC, (2) previtamin, (3) vitamin D_3, (4) lumisterol, and (5) tachysterol and (6) absorption spectrum of an aqueous solution of PSSS-VN (c = 1 g dm^{-3}, l = 1 cm) in a near-UV region.

Polymer solutions were prepared using deionized water. Ethyl acetate (Aldrich, 99.5+%), tetrahydrofuran (Caledon, HPLC), methanol, cyclohexane, chloroform (all ACS Spectro Grade), and ethanol (Aldrich, anhydrous, denaturated) were used as received.

Procedures. Solubilization of DHC. Solubilization of DHC in aqueous polymer solutions was achieved by slowly injecting microliter quantities of DHC (5×10^{-2} M) dissolved in tetrahydrofuran to milliliter quantities of polymer solution. Typically, 40–50 μL of DHC solution was injected to 10 mL of PSSS-VN solution of concentration 1 g dm^{-3}. Both the polymer and the stock DHC solutions were carefully degassed and protected against light and heat. When solubilization was completed, the resulting solution was shaken for 5 min, bubbled with argon for 15 min to remove the THF introduced with the probe, and equilibrated for 30 min in the dark at room temperature.

Irradiation of Samples. Natural source irradiations were carried out using solar light. These experiments were performed in Toronto on bright, sunny days in August and September (1990). The temperature of the samples was maintained between 15 and 20 °C. Under these conditions, PSSS-VN absorbs less than 3% of the total visible and UV radiation of the sun.

Artificial source irradiations were performed by using a deep-UV, constant intensity control system (Optical Associates, Model 780) described earlier in detail.[1] Interference filters (313 and 254 nm) were used to obtain monochromatic light. The incident light intensity was determined by using a ferrioxalate actinometer: $I_0 = 4 \times 10^{-9}$ Einstein s^{-1} (at 313 nm) and $I_0 = 7.3 \times 10^{-9}$ Einstein s^{-1} (at 254 nm). The solutions were mixed during irradiation using a small magnetic stirring bar.

Ultraviolet Spectra. UV spectra of the samples were measured using a Hewlett-Packard 8451A diode array spectrophotometer.

Fluorescence Spectra. Steady-state emission spectra of the samples were recorded at room temperature using an SLM spectrofluorimeter.

Product Analysis. Analysis of the products formed during irradiation of DHC in aqueous photozyme and in organic solvents was carried out using a Waters HPLC system with a Whatman Partisil 5 silica gel analytical column (4.6-mm i.d., 25 cm) and equipped with a Hitachi Model 100-40 spectrophotometer UV detector. A Hewlett-Packard 8451A diode array UV spectrophotometer equipped with a flow-through quartz cell (1-cm path length, 8-μL volume) connected to the effluent stream of the HPLC was used to take on-line spectra of each component. By use of a simple BASIC program, we set the spectrophotometer to take 60 spectra in 60 s as the eluting component passed through the cell. The monitoring of the spectra of the product through its elution was important to check the precision of separation and to determine the purity of the compound present in the eluting volume. This was especially important for the case of the previtamin because of its poor separation from lumisterol. A mixture of chloroform, cyclohexane, and tetrahydrofuran (50:50:1) was used as the mobile phase. The products were detected by monitoring their absorption at 254 nm. The samples for analysis were prepared as follows. The product mixture formed during irradiation of the polymer/DHC solution was extracted with chloroform under a stream of argon. The combined organic layers were washed with water, the solvent was removed under reduced pressure at room temperature, and residual solid material was dissolved in 0.5 mL of chloroform for analysis. The qualitative analyses were based on a comparison of the retention times and UV absorption spectra of the compounds from the reaction mixtures with the corresponding pure substances and literature data.[13,20] For quantitative analyses, the HPLC system was calibrated with solutions of known concentrations of these compounds containing toluene as internal standard.

Results and Discussion

It was observed that PSSS-VN in aqueous solution efficiently solubilizes DHC. The solubility of DHC in pure water was determined to be $(7.2 \pm 0.5) \times 10^{-6}$ M. Using a simple two-phase model for the system, previously described in detail,[24] we determined the distribution coefficient of DHC, defined as the ratio of the weight fraction of DHC in the polymeric pseudophase to that in the aqueous phase, to be $(4.1 \pm 0.5) \times 10^4$. Due to the preferential solubility of DHC in the hydrophobic polymeric pseudophase, the local concentration of this compound in the vicinity of naphthalene chromophores is high.

Irradiation of the PSSS-VN aqueous solution containing solubilized DHC with solar light results in efficient and selective production of previtamin D_3. Figure 2a shows HPLC traces obtained for the reaction mixture formed during irradiation of the PSSS-VN solution containing solubilized DHC ($c_0^{DHC} = 1.7 \times 10^{-4}$ M) with solar light for 90 min. For comparison, an HPLC trace for the mixture formed during solar irradiation of DHC in cyclohexane is presented in Figure 2b. Note that in the first spectrum there are only two distinct peaks: one of retention time 15 min, assigned to the starting material, and the second of retention time 7.3 min, characteristic of previtamin D_3. The assignment of the previtamin was based on the UV spectrum of the respective fraction and confirmed by thermal rearrangement (heated for 30 min at 60 °C under argon atmosphere in the dark) to vitamin D_3. In the second spectrum (Figure 2b), additional peaks appeared. They are characteristic for lumisterol ($t_R = 7.8$ min), tachysterol ($t_R = 9.8$ min), vitamin ($t_R = 10.7$ min), and an unidentified side product ($t_R = 13.5$ min) with a UV spectrum similar to DHC.

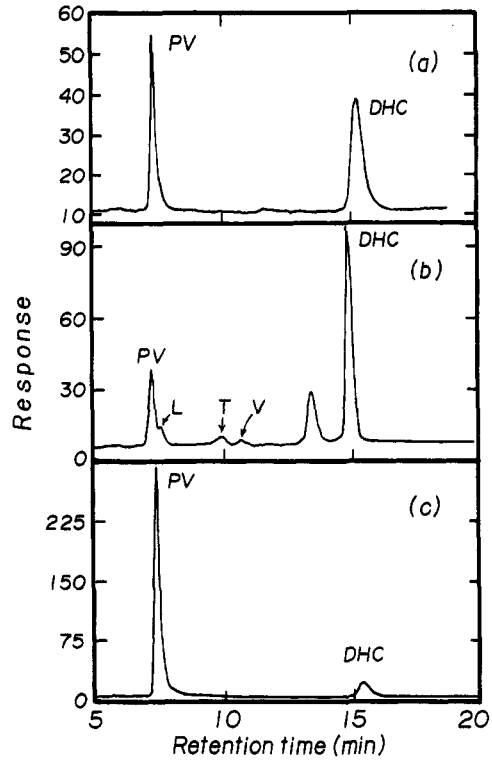


Figure 2. HPLC traces for the reaction mixtures obtained (a) after 90 min of solar irradiation of an aqueous solution of PSSS-VN containing solubilized DHC, (b) for DHC in cyclohexane solution irradiated 90 min with solar light, and (c) after 30 min of irradiation of an aqueous solution of PSSS-VN containing solubilized DHC ($c_{DHC} = 1.7 \times 10^{-4}$ M) with light at 313 nm.

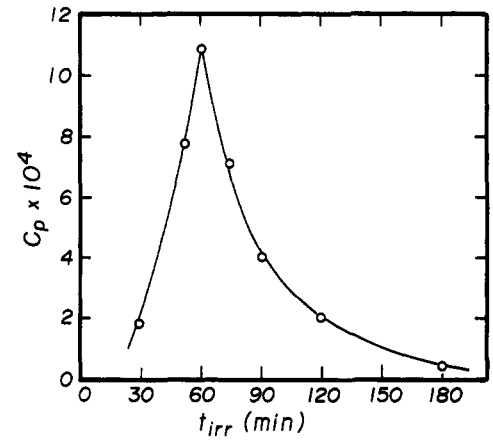


Figure 3. Dependence of the concentration of previtamin formed in a photozyme on the time of exposure to solar light ($c_{pol} = 1$ g dm^{-3}, $c_{DHC} = 1.7 \times 10^{-4}$ M).

The conversion of DHC to previtamin is more efficient in the photozyme than in organic solvent. After 1 h of solar irradiation, about 65% of the DHC is converted to previtamin in the photozyme, while only a 4.5% yield is obtained in cyclohexane after the same period of time. Interestingly, it was observed that the content of previtamin in the reaction mixture increases for irradiation times up to 60 min and then decreases after this (Figure 3). This is caused by the fact that the previtamin formed can undergo both thermal and photochemical rearrangements.

(24) Nowakowska, M.; White, B.; Guillet, J. E. *Macromolecules* **1990**, *23*, 3375.

Table I. Analytical Results for Photosensitized Reactions of DHC in Organic Solvent and Aqueous Polymer Solution (λ_{ex} = 313 nm)

system	$c_0^{DHC} \times 10^4$ M	t_{irr}, min	$c_{pre} \times 10^5$ M	$c_{tach} \times 10^6$ M	$I_a \times 10^3$ Einstein	$\gamma_{pre} \times 10^3$	$\gamma_{tach} \times 10^3$
DHC in cyclohexane	3.2	15	0.99	0	2.7	3.7 ± 0.2	0
		30	1.93	2.58	5.4	3.6 ± 0.2	0.48 ± 0.02
DHC in aqueous PSSS-VN solution	1.7	30	7.5	0	6.7	12.0 ± 0.5	0
		45	12.1	0	10.1	12.0 ± 0.5	0

Both of these processes become important after longer irradiation time. Due to infrared heating during exposure to solar light, the temperature of the sample gradually increases which induces thermal isomerization to vitamin D_3 followed by its photochemical transformation to other products. The efficiency of the photoisomerization of the previtamin to the byproducts was found to be much lower in the photozyme than in organic solvent.

The optimal exposure time to achieve the maximal conversion of DHC to previtamin depends upon the experimental conditions, including the initial concentrations of DHC and polymer, the temperature of the exposed sample, and the intensity of solar radiation. While the first three variables can be defined quite precisely and adjusted according to the demands of the experiment, it is very difficult to control the dose of solar radiation absorbed by the system for a series of nonsimultaneous experiments. In order to obtain more information on the dependence of DHC conversion on the radiation dose absorbed, experiments with monochromatic light at 313 nm were performed.

Irradiation of the photozyme system using an OAI lamp with a 313-nm interference filter and the same initial concentrations of polymer and DHC as those used in solar irradiation results in exceptionally clean photoisomerization of DHC to previtamin (see Figure 2c). In this case, the maximum conversion (70%) was achieved after 45 min of irradiation. The quantum efficiency of DHC photoisomerization in aqueous solution of PSSS-VN was determined to be $(2.0 \pm 0.5) \times 10^{-2}$ (see Table I). Irradiation of DHC + naphthalene in cyclohexane solution at 313 nm also results in photosensitized isomerization of DHC. The quantum efficiency of the process was found to be $(3.7 \pm 0.1) \times 10^{-3}$ at an initial concentration of DHC equal to 3.2×10^{-4} M and a naphthalene concentration 5.0×10^{-3} M (see Table I). Note that prolonged irradiation of DHC in organic solvents in the presence of naphthalene results in the appearance of tachysterol. The apparent quantum efficiency of tachysterol formation was determined to be $(4.8 \pm 0.2) \times 10^{-4}$. Tachysterol was not formed when the reaction was carried out in the photozyme.

Mechanistic Studies of Photoisomerization of DHC in Photozyme. Based on the absorption and emission spectra of DHC in ethanol at 77 K, the energy level for DHC in its first excited singlet state was determined to be about (93 ± 2) kcal mol^{-1} (307 nm). Solar radiation in the erythmal region (290–310 nm) should be absorbed by DHC and initiate its isomerization. This process occurs in human skin exposed to solar light and is responsible for production of vitamin D_3 in the human body. Due to the low total concentration of DHC in the photozyme system used and the higher absorption of PSSS-VN in the near-UV spectral region (see Figure 1), the direct excitation of DHC with solar light is negligible. For example, at 313 nm, DHC absorbs about 0.6% of the incident radiation. The photoisomerization of DHC in the photozyme system is therefore sensitized by excited polymeric naphthalene chromophores. Measurements of the steady-state fluorescence spectra of PSSS-VN in aqueous solution in the absence and presence of DHC show that DHC quenches the excited singlet state of naphthalene chromophores. Model studies performed using naphthalene/DHC solutions in THF show that the quenching can be described by Stern–Volmer kinetics (Figure 4). The bimolecular rate constant for the process in THF solution at room temperature was determined to be $(3.0 \pm 0.5) \times 10^9$ M^{-1} s^{-1}. The quenching could involve participation of an exciplex intermediate, since it has been shown that naphthalene can form exciplexes with a series of steroidal dienes.[25]

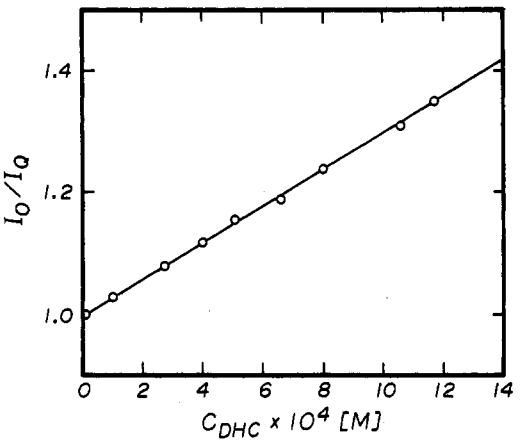


Figure 4. Stern–Volmer plot of the ratio of naphthalene fluorescence intensities (I_0/I_Q) in the absence (I_0) and presence of quencher (I_Q) versus concentration of DHC in THF solution.

Singlet–singlet energy transfer from naphthalene to DHC induces photoisomerization of DHC to previtamin D_3. Such a naphthalene-photosensitized isomerization of quadricyclene to norbornadiene has also been reported by Murov and Hammond.[26] In the photozyme system, the hydrodynamic radius of PSSS-VN (molecular weight $M_n \sim 120\,000$) is about 140 Å and each polymer chain contains, on average, 430 naphthalene chromophores. The average naphthalene concentration within each polymer coil is therefore $\sim 6 \times 10^{-2}$ mol L^{-1}, of the same order as those used in Figure 4. We envisage a mechanism whereby the migrating singlet exciton moving along the polymer chain hunts for a possible exciplex site where a DHC molecule is in close proximity to the excited naphthalene.

Photochemical Reactions of the Previtamin. As shown in Figure 2b, the formation of the previtamin in cyclohexane is accompanied by the creation of other isomers. The composition of the product mixture strongly depends on the excitation wavelength (Figure 5). Excitation at 254 nm mainly produces tachysterol while exposure to radiation of longer wavelength, 313 nm, results in formation of considerable amounts of lumisterol. This difference can be explained by taking into account the absorption characteristics of the respective isomers at the excitation wavelengths and the quantum yields for their isomerizations (compare Figure 1 and Scheme I). The formation of tachysterol and lumisterol is preceded by primary excitation of the previtamin to the singlet state. The overall kinetic scheme is quite complicated because it involves a set of parallel reversible processes.

Interestingly, it was shown in model studies that although naphthalene photosensitizes the formation of previtamin, it inhibits the creation of tachysterol and lumisterol. For example, during irradiation of DHC/naphthalene solutions in cyclohexane ($c_0^{DHC} = 1.6 \times 10^{-4}$ M, $c_{naph} = 2.1 \times 10^{-3}$ M) at 254 nm (90% of light absorbed by naphthalene), the rate of previtamin formation is four times higher than that during irradiation of DHC solution, while the increase in tachysterol concentration was two times slower in the system containing naphthalene. Similarly, the presence of naphthalene in a system ($c_0^{DHC} = 3.2 \times 10^{-4}$ M, c_{naph}

(25) Pusset, J.; Beugelmans, R. *Tetrahedron* **1976**, *32*, 791.
(26) Murov, S.; Hammond, G. S. *J. Am. Chem. Soc.* **1968**, *72*, 11.

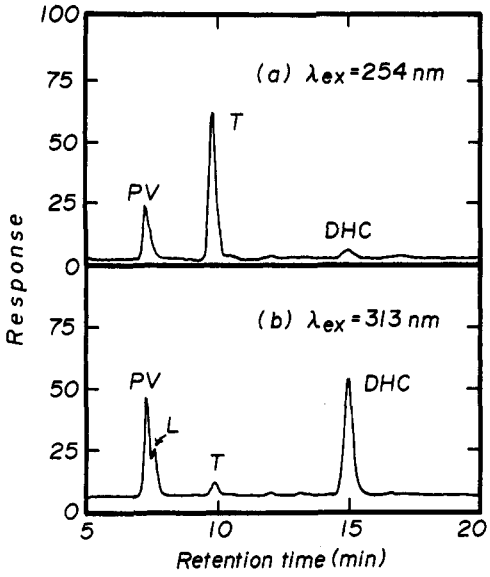


Figure 5. HPLC traces for DHC in cyclohexane solution irradiated for 120 min at (a) 254 and (b) 313 nm.

$= 5 \times 10^{-3}$ M) sensitizes previtamin formation and inhibits the appearance of lumisterol when 313-nm radiation is used to initiate reaction (97% of radiation absorbed by naphthalene). The formation of lumisterol is four times slower in the presence of naphthalene. In a system containing naphthalene, any photoisomerization of previtamin is induced by photosensitization because the amount of direct excitation is negligible. As was shown experimentally, however, the efficiency of this process is limited, which can be explained by taking into account two factors: the maximum of the absorption spectrum for the previtamin is shifted deeper toward the UV in comparison to those of the other photoisomers and the geometry of the molecule is not favorable for effective interaction with naphthalene.

The mechanism of singlet–singlet energy transfer in the aromatic hydrocarbon/triene system is still not fully explained. One might expect that the long-range Förster mechanism or a mechanism involving specific interactions can operate. Quenching according to the Förster mechanism cannot be efficient in the naphthalene–previtamin system because of poor overlap between the emission spectrum of the donor and absorption spectrum of the acceptor. On the other hand, to ensure efficient, specific interactions, both energetic and geometrical requirements have to be fulfilled. It has been shown that the efficiency of quenching of naphthalene by dienes is correlated with the position of the diene singlet absorption maxima and increases with a decrease in the excitation energy of the diene.[27] It has also been demonstrated that dienes possessing molecular geometries which allow extended physical overlap between the diene and the naphthalene moiety display the highest quenching activity. This explains the lower than expected quenching characteristics for cyclic dienes, which result from poor physical overlap with naphthalene. This might also hold for the previtamin triene, which possesses a rigid cyclic structure.

Considering the above, it seems that the naphthalene group plays an unusual double role in the photochemistry of the previtamin; it does act as a photosensitizer although the efficiency of the photosensitized isomerization of previtamin is lower than that induced by direct excitation, but, more importantly, it acts as a radiation screen. At higher local concentrations of naphthalene, such as in a photozyme system, the screening effect becomes increasingly important. This is one of the reasons that there are no detectable amounts of side products in the synthesis of the previtamin in a photozyme system up to high degrees of

(27) Stephenson, L. M.; Hammond, G. S. *Pure Appl. Chem.* **1968**, *16*, 125.

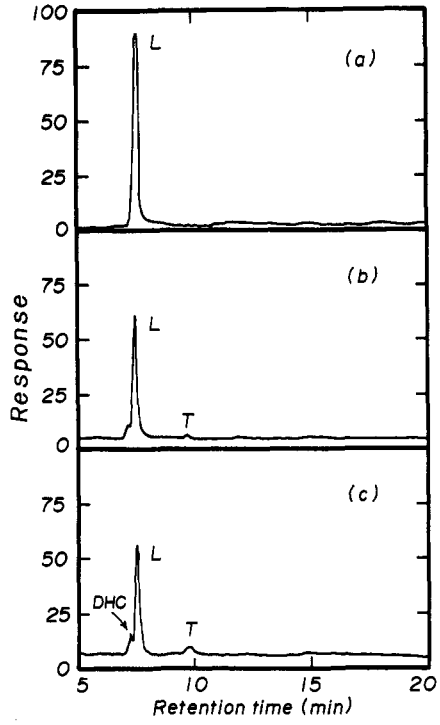


Figure 6. HPLC traces for lumisterol irradiated in the presence of naphthalene at 313 nm for various periods of time: (a) 0 min, (b) 15 min, and (c) 30 min.

DHC conversion; the other reason relates to the photosensitized back isomerization of lumisterol and tachysterol to previtamin.

Photosensitized Isomerization of Lumisterol. It was demonstrated that irradiation of a lumisterol/naphthalene solution ($c_0^{Lu} = 7.6 \times 10^{-5}$ M, $c_0^{naph} = 4.3 \times 10^{-3}$ M) with radiation at 313 nm (98% of radiation absorbed by naphthalene) results in the consumption of lumisterol accompanied by the appearance of previtamin in the first stage and later, tachysterol (Figure 6). During prolonged irradiation, the ratio between the concentration of previtamin and tachysterol increases and a trace amount of DHC appears. Unfortunately, due to the overlap between lumisterol and previtamin peaks, it is difficult to obtain precise quantitative data.

Triplet-Photosensitized Isomerization of Tachysterol. In the next step of this work, the effect of naphthalene on the photochemical reaction of tachysterol was studied. It is known that very efficient isomerization of previtamin to tachysterol (see Scheme I) during irradiation of the reaction mixture with short-wavelength, highly energetic radiation (254 nm) is a major problem in the commercial synthesis of vitamin D. The reaction mixture in the photostationary state is exceptionally rich in tachysterol (tachysterol content ≈75%).

DHC in cyclohexane solution ($c_{DHC} = 1 \times 10^{-4}$ M) was irradiated for 120 min at 254 nm. This leads to formation of a substantial amount of tachysterol ($c_{tach} = 6 \times 10^{-5}$ M) and consumption of 85% of the DHC (Figure 7a). The quantum yield of tachysterol formation in cyclohexane was found to be 0.45 which is in good agreement with the literature value.[11c] In the next step, naphthalene was added to the reaction mixture ($c_{naph} = 8.5 \times 10^{-3}$ M) and the system was irradiated with light at 313 nm. HPLC analysis was performed every 5 min. It was observed that the content of tachysterol decreased and the amount of previtamin increased (Figures 7b–d), unequivocally showing that the isomerization of tachysterol is photosensitized by naphthalene. The quantum efficiency of this process was determined to be $(2 \pm 0.3) \times 10^{-2}$.

It is possible that the process can be sensitized by both singlet–singlet and triplet–triplet energy transfer. In order to distinguish between these two mechanisms, the effect of triplet quencher

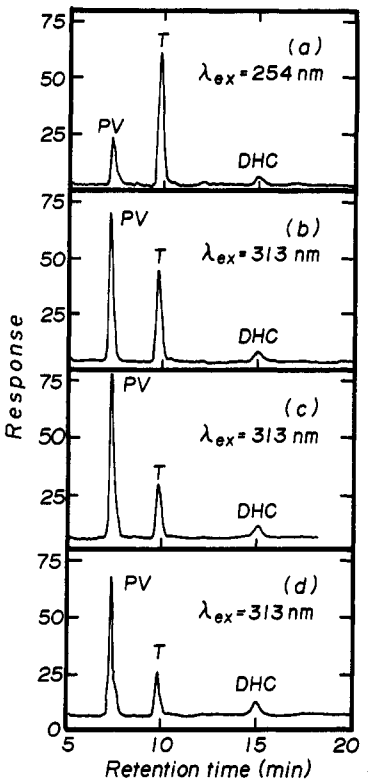


Figure 7. HPLC traces for DHC in cyclohexane irradiated for 120 min at (a) 254 nm and for mixtures obtained following the first-step naphthalene-sensitized reaction induced by irradiation at 313 nm for different periods of time: (b) 5 min, (c) 15 min, and (d) 30 min.

trans-stilbene ($E_T < 50$ kcal mol^{-1})[28] on the naphthalene-sensitized isomerization was studied. It was found that *trans*-stilbene ($c = 5 \times 10^{-3}$ M) inhibited the naphthalene-sensitized conversion of tachysterol to previtamin. *trans*-Stilbene is not expected to be an efficient quencher for the naphthalene singlet state because its singlet energy level is higher than that for naphthalene ($E_{naph} = 92$ kcal mol^{-1}, $E_{trans\text{-}st} = 94.2$ kcal mol^{-1}).[28] Based on the above observation, it can be concluded that the naphthalene-photosensitized isomerization of tachysterol occurs via triplet–triplet energy transfer. This explains the high efficiency and observed selectivity of this process. It is known that *cis–trans* isomerization of tachysterol ⇌ previtamin is the only isomerization of the vitamin D family which can be induced by triplet sensitization.[11c] Triplet sensitization by naphthalene is favored by the high efficiency of naphthalene triplet formation (quantum efficiency of intersystem crossing is 0.82) and the relatively long lifetime (~0.1 s) of the naphthalene triplet. It is worth noting that although this photosensitized process is reversible, the quantum yield for previtamin formation is four times higher than that for the back reaction and almost six times higher than those in the case of the processes induced by direct excitation (see Scheme I).

After longer irradiation times, trace amounts of lumisterol appear in the reaction mixture. This is probably due to the singlet-photosensitized isomerization of previtamin. Because the quantum yield for lumisterol formation is much lower than that for the back reaction (see Scheme I), this process becomes important only at high concentrations of the previtamin, formed in this system after longer irradiation times.

Mechanism of Photochemical Reactions in the Photozyme. Based on the above experimental results, it is possible to explain the observed, highly specific formation of previtamin during irradiation of the PSSS-VN photozyme containing solubilized DHC. The mechanism of the process involves three different photosensitization steps: (1) singlet-photosensitized isomerization of DHC to previtamin, (2) singlet-photosensitized isomerization of lumisterol to previtamin, and (3) triplet-sensitized isomerization of tachysterol to previtamin. Although these processes are reversible, the equilibrium is shifted toward formation of previtamin. It should be noted that these processes are expected to be efficient in the photozyme system due to the high local concentration of DHC and its isomers in the vicinity of the naphthalene chromophores acting as energy donors. This is particularly important in case of a triplet-sensitized process because close contact between donor and acceptor molecules is required for efficient triplet–triplet energy transfer.

Based on the experimental data presented above, the following scheme can be proposed to describe the photochemical formation of previtamin D_3 in the photozyme system:

$$^1N_0 + h\nu \rightarrow {}^1N^* \quad (1)$$

$$^1N^* \rightarrow {}^1N_0 + h\nu' \quad (2)$$

$$^1N^* \rightarrow {}^1N_0 \quad (3)$$

$$^1N^* + {}^1(DHC)_0 \rightarrow {}^1N_0 + {}^1(DHC)^* \quad (4)$$

$$^1(DHC)^* \rightarrow {}^1P_0 \quad (5)$$

$$^1N^* + {}^1P_0 \rightarrow {}^1P^* \quad (6)$$

$$^1P^* \rightarrow {}^1T_0 \quad (7)$$

$$^3N^* + {}^1T_0 \rightarrow {}^1N_0 + {}^3T^* \quad (8)$$

$$^3T^* \rightarrow {}^1P_0 \quad (9)$$

$$^3N^* + {}^1P_0 \rightarrow {}^1N_0 + {}^3P^* \quad (10)$$

$$^3P^* \rightarrow {}^1T_0 \quad (11)$$

$$^1P^* \rightarrow {}^1L_0 \quad (12)$$

$$^1N^* + {}^1L_0 \rightarrow {}^1L^* + {}^1N_0 \quad (13)$$

$$^1L^* \rightarrow {}^1P_0 \quad (14)$$

where N, P, T, and L stand for naphthalene polymeric chromophores, previtamin, tachysterol, and lumisterol, respectively; left superscripts show multiplicity of the electronic states of the respective molecules; and right subscripts and superscripts denote the ground (0) and excited (*) states, respectively.

Conclusions

It has been shown that exceptionally clean synthesis of previtamin D_3 in aqueous solution using solar and artificial radiation can be performed using the novel antenna polyelectrolyte PSSS-VN. The reaction occurs in the interior of hydrophobic microdomains created within the polymeric chains due to the clustering of naphthalene units in contact with water. The first step involving isomerization of DHC to previtamin is sensitized via singlet–singlet energy transfer from excited naphthalene chromophores to DHC. The absence of lumisterol and tachysterol at detectable levels in the reaction mixture can be explained by taking into account the fact that the naphthalene chromophore plays a double role in the photochemistry of isomers belonging to the vitamin D family. It acts both as a photosensitizer and as a screen. It photosensitizes both the formation and back isomerization of lumisterol and tachysterol, but the efficiencies of back reactions toward the previtamin are noticeably higher. Thus, the reaction mixture becomes progressively richer in previtamin. The increase in previtamin concentration during irradiation of DHC in an organic solvent in the absence of naphthalene is a sort of "self-destructive" phenomenon because

(28) Murov, S. L. *Handbook of Photochemistry*; Marcel Dekker: New York, 1973.

it inevitably leads to an increase in the rate of side product formation. This process is greatly inhibited in the photozyme system because the direct excitation of previtamin is limited by the screening provided by the naphthalene polymeric chromophores present at high concentrations and because the efficiency of photosensitized isomerization of previtamin is much lower than that for the back reactions. Due to the optimal combination of photosensitizing and screening effects in the naphthalene photozyme/vitamin D_3 isomeric system, high degrees of DHC conversion are possible while still obtaining a relatively pure product.

Acknowledgment. The authors gratefully acknowledge a generous gift of lumisterol sample from Dr. K. Pfoertner (Hoffmann-LaRoche & Co. Ltd., Basel). The financial support of the Natural Sciences and Engineering Research Council of Canada and the Ontario Centre for Materials Research is gratefully acknowledged.