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The degradation of vitamin A exposed to ultraviolet radiation

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Summary

The degradation of vitamin A palmitate exposed to UV light was studied. Radiance energy was measured using a U.V. radiation monitor. Degradation in aqueous solutions was non-exponential and could be described by a polynomial equation. Degradation rate was dependent to a small extent on the initial concentration of vitamin A. Results suggested that the non-exponential nature of the degradation curve might be due to the protective effect of degradation products. Breakdown of vitamin A was similar in glass and PVC containers, but degradation was significantly slower in amino acid infusions. The monitor could be used to measure UV radiance from sunlight. This information can be used to estimate the losses of vitamin A palmitate after addition to total parenteral nutrition regimes. The clinical significance of the tailing phenomenon of the degradation process are discussed.

Introduction

Vitamin A is very sensitive to ultraviolet radiation (DeRitter, 1982) and the esters are rapidly inactivated in aqueous solution during exposure to daylight. Liquid pharmaceutical preparations containing vitamins are packed in daylight-protective containers. Multivitamin injections are essential components of total parenteral nutrition and are usually added to the infusion prior to administration. It has been shown that vitamin A is rapidly degraded if these infusions are exposed to daylight

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(Allwood, 1982). The extent and rate of degradation depend on the degree of exposure to daylight. The irradiance to which the container is exposed depends on a number of factors such as the direction of radiation, time of day, climatic conditions and the position of the infusion relative to the radiation source. It is the purpose of this study to examine the correlation between degradation rates of vitamin A in aqueous solution and ultraviolet irradiance. The protective effect of the container and other ingredients of the infusion were also assessed. The relevance of these observations to vitamin A stability in total parenteral nutrition infusion is discussed.

Materials and Methods

Vitamin A palmitate was a gift from Roche Products, Welwyn Garden City, Herts. The preparation contained 100,000 IU/ml in a water-miscible base. Dextrose 5% w/v (Viaflex), Synthamin and PVC (Viaflex) bags were obtained from Travenol Laboratories, Thetford, Norfolk, Aminoplex 12 from Geistlich Sons, Chester and Vamin-Glucose from Kabi-Vitrum, Uxbridge, Middlesex.

A wide-band ultraviolet radiation monitor based on that described by Mountford and Pepper (1981) has been used incorporating a gallium arsenide phosphide (GaAsP) photodiode.

The variation of sensitivity of photodiode was measured using a high-pressure 900 W xenon arc lamp and an Applied Photophysics f/4 single grating monochromator as described elsewhere (Mountford and Pepper, 1981). Using a bandwidth of 2 nm, the detector responded only in the range 300–400 nm with a peak sensitivity at 365 nm.

Using an optical bench and an unfiltered medium-pressure mercury lamp, the linearity of the meter was investigated. The voltage (V) produced at the output of the meter was measured at several distances (d) of the outer face of the filter from a fixed point on the lamp housing. A graph of $V_{0.5}$ against d produced a straight line indicating the detector had a linear response to changes in irradiance. The range of irradiances used was about 0.5–7.5 $mW \cdot cm^{-2}$.

The detector was placed on a turntable on an optical bench and irradiated with radiation from an unfiltered mercury arc lamp. The voltage at the output of the detector was recorded at intervals of 10° up to 90° from the normal. The meter decreased more rapidly than a cosine response. The response of the meter was zero at an angle of 71° from the normal. This compared with a calculated [1] acceptance angle of 70° about the normal.

The monitor was calibrated by exposing it to the radiation from a medium-pressure mercury arc lamp operated from a stabilizing power supply. The radiation from the lamp passed through a Schott UG1 glass filter and a WG335 filter resulting in essentially monochromatic radiation at 365 nm and some infrared wavelengths. The voltage at the output of the detector was recorded and the total irradiance was measured at the same position by a calibrated wide-angle Rank Hilger FT17

[1] Data from Hamamatsu Catalogue No. sc-3-2; Hamamatsu TV Co. Ltd., Japan.

thermopile using the method of Diffey and Challoner (1978). The measurement was repeated with a RG630 filter in front of the thermopile since the thermopile responded to radiation with wavelengths greater than 680 nm. The measured sensitivity of the detector to 365 nm radiation was 25.4 ± 1.2 mV · mW^{-1} · cm^{-2}.

Irradiation method

In preliminary experiments vitamin A solutions were irradiated using sunlight as the source of ultraviolet radiation and ultraviolet-sensitive film badges (Davies et al., 1976) in which 8-methoxypsoralen was the chromophore. These were unsuccessful because of: (a) the relatively slow rate of change of the transmission of the chromophore compared with the relatively fast rate of change of vitamin A concentration; and (b) the variation in the spectral distribution of ultraviolet global irradiation due to solar altitude and the total amount of atmospheric ozone.

All further irradiations were carried out in a darkened environment at room temperature. The source of ultraviolet radiation used to irradiate vitamin samples was a Mineralight UVSL25. This device contains two 4 W fluorescent tubes the radiation from which is filtered to produce a spectrum in the range 254–390 nm [2]. The detector was placed at a known distance from the front face of the housing of the lamp and the output of the meter was recorded on a chart recorder. Initial experiments showed that the output from the meter and therefore the source of radiation were constant over typical irradiation times. A device which measures and integrates ultraviolet irradiance (Pepper and Mountford, 1983) would have proved more appropriate and is currently under construction. The detector was replaced by the vitamin samples located in a glass or plastic container. A mercury thermometer was used to measure the temperature of the solution during irradiation. Solutions of vitamin A were prepared in 150 ml glass beakers. In most experiments the beaker contained 100 ml of a 4 IU/ml solution (equivalent to 10,000 IU in 2.5 litres total parenteral nutrition infusion). Solutions were rapidly stirred by means of a magnetic stirrer, a heat insulation pad being placed between stirrer top and beaker to prevent any temperature change in the solution in the beaker. At selected time intervals, samples (0.25 ml) were removed for immediate analysis by HPLC as previously described (Allwood, 1982). Controls consisted of zero time (unexposed) samples from each experiment and from stirred samples stored in darkness. There was no evidence of vitamin A degradation in these latter samples.

In order to study the relationship between ultraviolet irradiation and vitamin A degradation, a series of experiments were conducted in the manner described. Data collected using several levels of radiant exposure resulting in up to 97% vitamin degradation were pooled; 158 data points were collected and a line of best fit was generated (Fig. 1, curve A) based on a least-squares method. The equation was of the form:

$$y = A_0 + A_1x + A_2x^2 + A_3x^3$$

[2] Data from Ultra-violet Products Ltd. Catalogue, Cambridge.

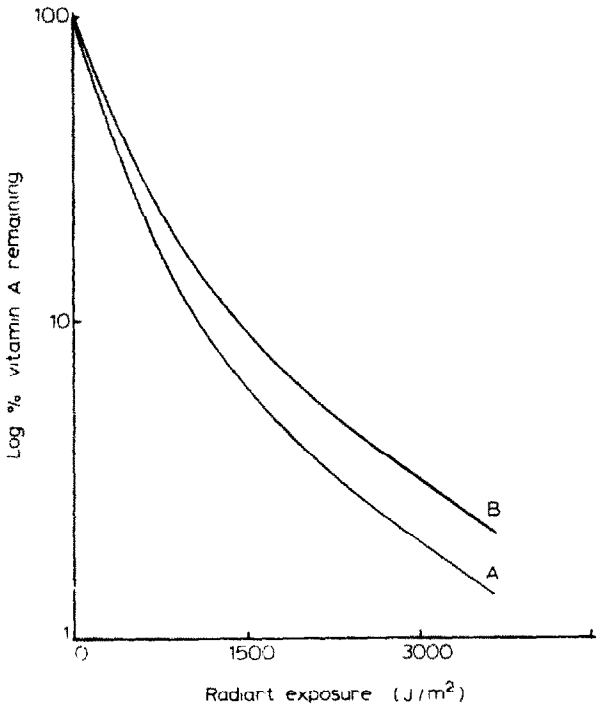

Fig. 1. The degradation of vitamin A (4 IU/ml in 5% dextrose) exposed to ultraviolet radiation. A: accumulated data from 158 points over a range of irradiances from 0.5 to 7.5 $mW \cdot cm^{-2}$. B, accumulated data from 35 points, each representing the degradation of a second addition of vitamin A to a previously exposed solution in which 1% of the first sample is not degraded at the commencement of the second exposure. Each plot is a best fit line obtained using a least-squares method of curve fitting.

where y = ln (concentration of vitamin A %); x = radiant exposure ($J \cdot m^{-2}$); $A_0 = 0.45860$ E-1; $A_1 = -0.23355$ E-2; $A_2 = 0.76334$ E-6; $A_3 = -0.10286$ E-9.

The correlation coefficient of this data was 0.993. Clearly, degradation is non-exponential. One explanation for the observation is that products of degradation of vitamin A provided protection for the residual vitamin A. Further studies were carried out in which a sample was exposed to irradiation for sufficient time to reduce the residual vitamin A content to less than 1%. A second addition of vitamin A was

TABLE 1

THE EFFECT OF AMINO ACID INFUSIONS ON DEGRADATION OF VITAMIN A EXPOSED TO ULTRAVIOLET RADIATION

Solution	Relative radiant exposure to give 50% degradation (5% dextrose = 1.00)
Vamin-glucose	1.44
Synthamin 9	2.00
Synthamin 17	1.81
Synthamin 9 + dextrose (20%)	1.53

made to restore the concentration to about 4 IU/ml and this solution re-irradiated. The results are shown in Fig. 1, curve B. Using a similar formula to the one above, the following coefficients were obtained:

$A_0 = 0.45787$ E-1

$A_1 = -0.18702$ E-2

$A_2 = 0.52721$ E-6

$A_3 = -0.65232$ E-10

The correlation coefficient of this data was 0.997. This indicates that some protection is provided from degradation products since the rate of breakdown of vitamin A in the second sample is reduced. The effect of vitamin A concentration on degradation rate was also studied. Results are illustrated in Fig. 2. Degradation rates

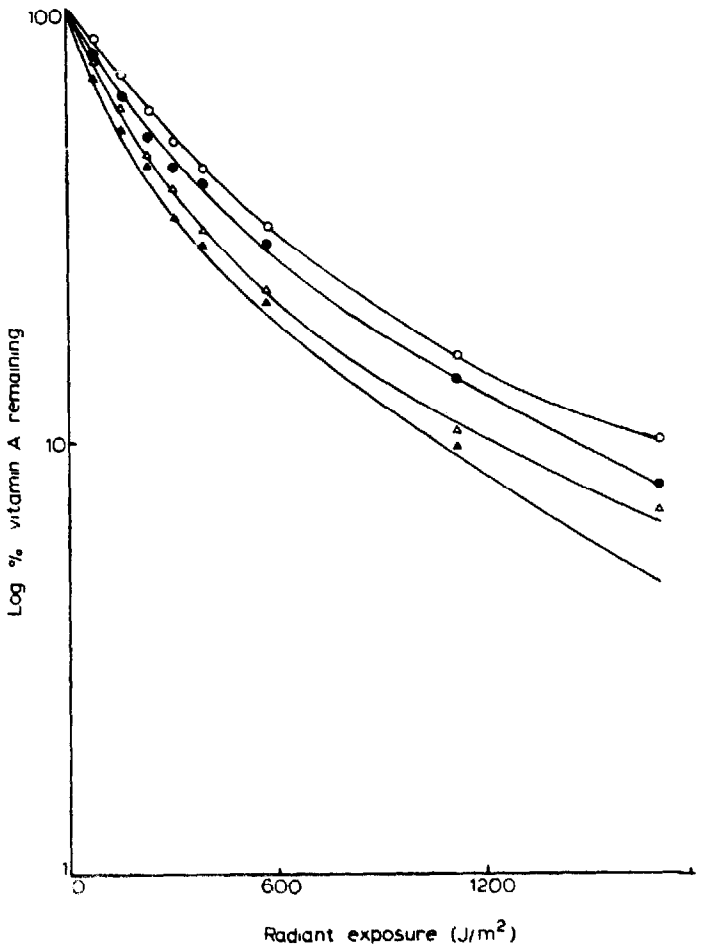

Fig. 2. The influence of vitamin A concentration on degradation during exposure to ultraviolet radiation. ○, 4 IU/ml; ●, 3 IU/ml; △, 2 IU/ml; ▲, 1 IU/ml.

are reduced at higher concentrations of vitamin A suggesting again that some protective effect is evident. However, the differences were relatively small.

Infusions are commonly packed in plastic (PVC) containers. The effect of PVC on degradation rate was examined. It was found that degradation rates of vitamin A exposed to ultraviolet radiation in PVC containers were identical to degradation of the compound in glass beakers. Therefore the results obtained in glass containers can be used to predict vitamin A losses in infusions contained in PVC bags.

Vitamin A is added to complex total parenteral nutrition infusions in big bags. The composition of the infusion was found to have a significant influence on degradation rates. The amino acid solutions examined protected vitamin A. The degradation rates were reduced as illustrated in Table 1. It is clear that degradation depends on the composition of each amino acid solution. The rate of degradation can be reduced by up to 50% in some amino acid mixtures compared with degradation in dextrose infusion.

Discussion

The degradation of vitamin A exposed to ultraviolet radiation progresses exponentially over the initial exposure period after which the rate of breakdown is progressively reduced. This reduction is apparently due to a protective effect of degradation products. The chemical reaction can be described by a polynomial, the constants varying with the composition of the solvent. This enables losses to be predicted if the irradiance is known. Using published calculations (Diffey, 1977) it has been estimated that at solar noon during summer in the U.K. the irradiance on an unshaded horizontal surface at sea level is around 40 $W \cdot m^{-2}$ in the A region (400–315 nm) of the ultraviolet spectrum. This value could fall to approximately 100 times less in midwinter. This range of values includes the maximum irradiance of 29.4 $W \cdot m^{-2}$ used in this study. This supports the experimental findings of Allwood (1982) that daylight-induced degradation can cause clinically significant losses from total parenteral nutrition infusions. The factors which affect the ultraviolet component of daylight are complex and include atmospheric ozone absorption, atmospheric Rayleigh scattering, Mie scattering, perihelion and aphelion variations, solar altitude, cloud cover, surface reflection, ground reflection and altitude. Therefore the irradiance in a particular locality cannot be readily predicted. The meter used in this study does provide an indication of irradiance.

The observation that degradation of vitamin A following prolonged exposure to ultraviolet radiation is not exponential but results in significant tailing of each curve may be of clinical significance. The recommended daily requirement by intravenous administration is 3300 IU (Anon, 1979). Since rapid losses follow exposure to daylight, an excess of vitamin A should be added to infusions and total parenteral nutrition regimes to compensate for degradation before and during infusion. The effect of the reducing rate of loss observed could account for up to 43.3% (from Fig. 1) of vitamin A delivered, compared with losses where the process was exponential. In practice it has been reported that exposure to daylight during administration can

result in < 10% of the added vitamin A being delivered (Allwood, 1982).

While degradation can be reduced or eliminated by covering the containers with a daylight-absorbing plastic film, significant degradation will also occur during passage of the infusion through the administration set.

In conclusion, this study has shown that the degradation of vitamin A is initially approximately proportional to ultraviolet radiant exposure. This can be measured using a detector which is relatively easy to operate and can be used to assess exposure levels encountered in practice. Such information can be used to predict likely approximate losses. The degradation of vitamin A is largely unaffected by PVC film compared with glass but the vitamin can be protected by degradation products and to a much greater extent by amino acids in the solution. The nature of the protective effect has not been elucidated.

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References

Allwood, M.C., The influence of light on vitamin A degradation during administration. Clin. Nutr., 1 (1982) 63–70.

Anon, Multivitamin preparation for parenteral use. Am. Med. Ass., Department of Food and Nutrition, J. Parent. Ent. Nutr., 3 (1979) 258–262.

Davies, A., Deane, G.H.W. and Diffey, B.L., Possible dosimeter for ultraviolet radiation. Nature (Lond.), 261 (1976) 169–170.

DeRitter, E., Vitamins in pharmaceutical formulations. J. Pharm. Sci., 71 (1982) 1073–1096.

Diffey, B.L., The calculation of the spectral distribution of natural ultraviolet radiation under clear day conditions. Phys. Med. Biol., 22 (1977) 309–316.

Diffey, B.L. and Challoner, A.V.J., Absolute radiation dosimetry in photochemotherapy. Phys. Med. Biol., 23 (1979) 1124–1129.

Mountford, P.J. and Pepper, M.G., A wide-band ultraviolet radiation monitor for measuring the output of monochromators used in dermatology. Phys. Med. Biol., 26 (1981) 925–930.

Pepper, M.G. and Mountford, P.J., A meter for ultraviolet dose and irradiance. Phys. Med. Biol., 28 (1983) 257–267.