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RESEARCH PAPERS

QUININE ACTINOMETRY AS A METHOD FOR CALIBRATING ULTRAVIOLET RADIATION INTENSITY IN LIGHT -STABILITY TESTING OF PHARMACEUTICALS

S. Yoshioka [1], Y. Ishihara [2], T. Terazono [2], N. Tsunakawa [3], M. Murai [4], T. Yasuda [5], Kitamura [5], Y. Kunihiro [6], K. Sakai [6], Y. Hirose [7], K. Tonooka [8], K. Takayama [8], F. Imai [9], M. Godo [9], M Matsuo [10], K. Nakamura [10], Y. Aso [1], S. Kojima [1], Y. Takeda [11] and T. Terao [1]

[1]National Institute of Health Sciences, Tokyo 158, [2]Chugai Pharmaceutical Co., LTD, Tokyo, [3]Daiichi Pharmaceutical Co., LTD, Tokyo, [4]Eisai Co., LTD, Tokyo, [5]Fujisawa Pharmaceutical Co., LTD, Osaka, [6]Mochida Pharmaceutical Co., LTD, Shizuoka, [7]Sandoz Pharmaceuticals, LTD, Ibaraki, [8]Sankyo Co., LTD, Tokyo, [9]Takeda Chemical Industries, LTD, Osaka, [10] Tanabe Seiyaku Co., LTD, Osaka, [11]National Institute of Health Sciences, Osaka.

ABSTRACT

A collaborative study was carried out by seven different laboratories to evaluate quinine actinometry as a universal, standardized method for calibrating UV radiation intensity from light sources used in light-stability testing of pharmaceutical products. Near UV fluorescent lamps, white fluorescent lamps, metal halide lamps and xenon arc lamps were employed as light sources. The increase in absorbance at 400 nm of aqueous quinine solutions was found to be proportional to the integrated UV energy emitted from the light sources. The linearity observed between absorbance and integrated UV energy indictates that quinine actinometry can be used to measure the intensity of UV radiation at wavelengths around 330 nm. The slopes of regression curves of absorbance vs integrated UV energy varied among the lamps used due to differing spectral distributions. Light degradation of nifedipine, a model photosensitive drug, was studied based on quinine actinometry.

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INTRODUCTION

Photostability of pharmaceutical products is assessed using light sources which simulate sunlight or room lighting. A universal, standardized method for calibrating the radiation intensity from these light sources is required in order to establish an internationally harmonized light-stability testing method for pharmaceuticals.

Various kinds of actinometry have been employed to measure radiation intensity (1-5). Physical actinometry using radiometers or photometers often yields differing results between meters, depending on the spectral characteristics as well as the calibration method of the meters. In contrast, chemical actinometry based on the measurement of a chemical change occurring in a photosensitive standard is considered to be an accurate and reproducible method for measuring the intensity of radiation to which phamaceutical products are exposed in light-stability testing (3, 4). Integrating chemical actinometers measure the total number of quanta absorbed by the standard. Therefore, the total amount of light-exposure measured by integrating chemical actinometers is not affected by fluctuations in the intensity or spectral distribution of the lamp emission, which may result from changes in variables such as line voltage or lamp age over the course of the study.

Ferrioxalate, which absorbs a wide spectral range (UV and visible) of light, has been used as a chemical actinometry standard in the light-stability testing of various materials such as paints and textiles (3, 5). In the present study, quinine actinometry was evaluated as a standardized method for calibrating UV radiation intensity in light-stability testing of pharmaceutical products. In contrast to ferrioxalate actinometry, quinine actinometry measures light intensity in only the UV region. Seven different laboratories jointed in this collaborative study to vailidate quinine actinometry.

MATERIALS AND METHODS

Materials

Quinine hydrochloride and nifedipine were purchased from Fujisawa-Astra (Osaka) and Sigma (St. Louis), respectively.

Quinine solutions were contained in 20-ml colorless ampoules (JIS R3512, No.5) having the transmittance curve shown in Fig.1-a. Nifedipine solutions were placed in 15-ml colorless vials (N-16, Maruemu, Osaka) having the transmittance curve shown in Fig.1-b.

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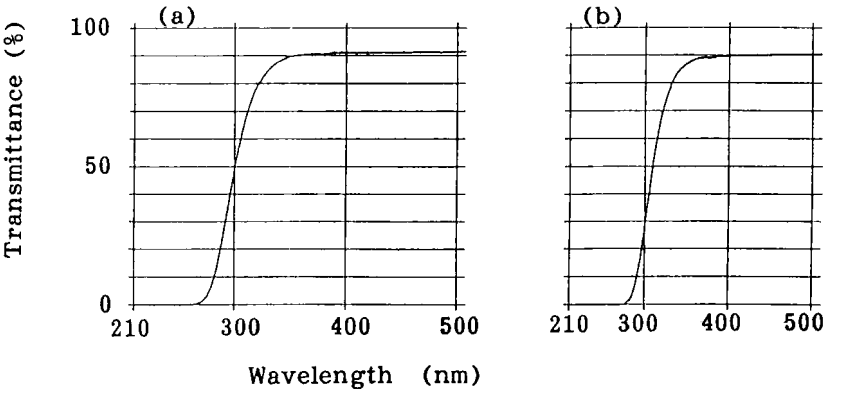

Fig. 1. Transmittance curves of ampoules (a) and vials (b).

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Apparatus

Each laboratory used different light exposure chambers equipped with one or two lamps. Three different kinds of lamps (a near UV fluorescent lamp, a white fluorescent lamp, and a xenon arc or metal halide lamp) were used in each laboratory. Near UV fluorescent lamps employed in the laboratories were FL20SBL, FL40BL and FLR20SBL/M (Toshiba). White fluorescent lamps used were FL20SSEX/18 (National), FLR20S (Toshiba), FL20SSW/18 (Toshiba), FLR20SW/M (Mitsubishi or National) and FLR40SW/M (NEC). Xenon arc lamps used were QLGXe2500 (Suga), 3.5KwXe (ATLAS Electric Device Company) and XC-100AF (Seric). Metal halide lamps used were MLBOC400C-H (Mitsubishi), DR400/TL (Toshiba) and DR125/TL (Toshiba).

The UV radiation energy of the lamps was measured by a radiometer having a detection range of 310 to 400 nm (TOPCON UVR-36). Illumination intensity was determined using a photometer (TOPCON, IM-3).

Sample Preparation and Procedures

Quinine hydrochloride (25.00 g) was dissolved in water to make a 5 % quinine solution. Ten ml of the solution were put into each of 20-ml ampoules and sealed hermetically. The ampoules were divided into three groups, each of which was exposed to either a near UV lamp, a white fluorescent lamp, or a xenon arc or metal halide lamp. For comparison, ampoules completely shielded with aluminium foil were also exposed to the lamps. Exposures were performed at three levels of integrated light intensity for each lamp; about 10, 20, and 30 W hr/m ^{2} for the near UV lamp; and about 1, 2, and 3 x10^{5} lux hr

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for the white fluorescent lamp and the xenon arc or metal halide lamp. The desired integrated light intensity was obtained by controlling the radiation period. After light exposure, the absorbance of the quinine solution at 400 nm was determined using a cell with a 1 cm path length. Similar experiments were carried out with a 2.5 % quinine solution to examine the effect of quinine concentration on photoreactivity.

The quinine solution exposed to light was subjected to high performance liquid chromatography (HPLC) to detect decomposition products. The method described in the Purity (4) Related Substances of Quinine Hydrochloride of JPXII was employed.

Nifedipine (5.00 g) was dissolved in dehydrated ethanol to make a 1 % nifedipine solution. Ten ml of the solution were put into each of 15-ml vials, and exposed to the lamps in a procedure similar to that used for quinine. Integrated light intensity was about 3, 6 and 10 W hr/m ^{2} for the near UV lamp; and about 0.7, 1.4, and 2.0 x10^4 lux hr for the white fluorescent lamp and the xenon arc or metal halide lamp. After light exposure, the absorbance of the solution was determined at 450 and 700 nm.

Decomposition products formed were determined by HPLC. The nifedipine solutions exposed to light were diluted to 0.01 % with the mobile phase, and injected into a column (YMC-Pack ODS AM-302, 4.6mm i.d., 15cm) maintained at 40°C. Mobile phase was a 1:1 mixture of 20 mM phosphate (pH 3.0) and methanol. The flow rate was 1.3 ml/min, and the eluent was monitored at 254 nm.

RESULTS AND DISCUSSION

Validation of Quinine Actinometry

Qunine solution transmittance decreased with exposure to light from the near UV lamp, the white fluorescent lamp, and the xenon arc or metal halide lamp. Figure 2 shows a typical profile of the transmittance change as a function of the integrated light intensity. Absorbance at 400 nm is plotted against integrated UV energy as determined by a radiometer in Figs. 3, 4, and 5 for the near UV lamp, the white fluorescent lamp, and the xenon arc or metal halide lamp, respectively. Absorbance appeared to increase linearly with increasing integrated UV energy for all the lamps used.

A regression line of absorbance vs integrated UV energy was calculated from the data obtained for each lamp. Tables I, II and III show the slopes and intercepts of the regression lines, as well as the correlation coefficients obtained, for the near UV lamp, the white fluorescent lamp, and the xenon arc or metal halide lamp, respectively. All the data obtained using 7 different near

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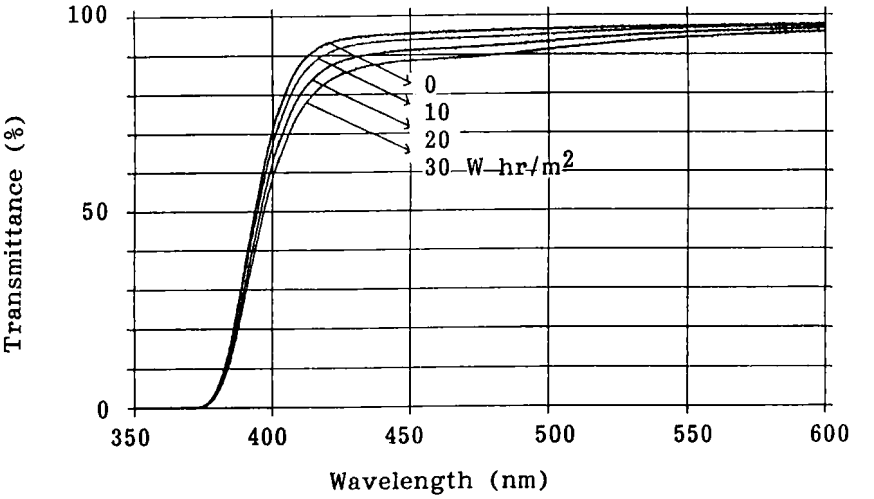

Fig. 2. Change in transmittance of quinine solutions exposed to a near UV lamp (FL40BL), as a function of integrated UV energy.

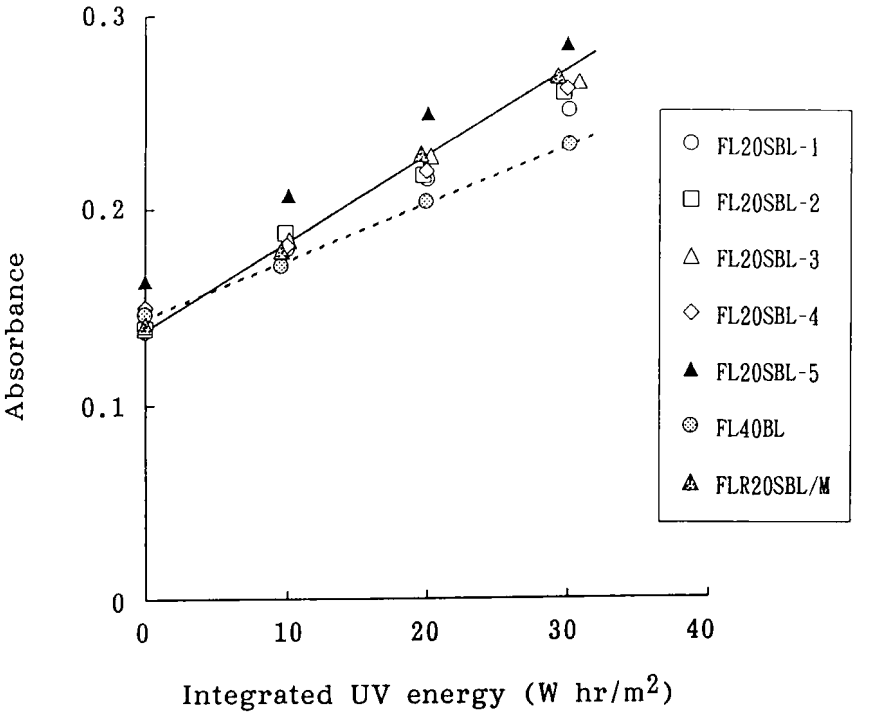

Fig. 3. Absorbance at 400 nm of quinine solutions exposed to near UV fluorescent lamps as a function of integrated UV energy.

Lines (——) and (----) indicate the regression lines with the largest and the smallest slopes, respectively.

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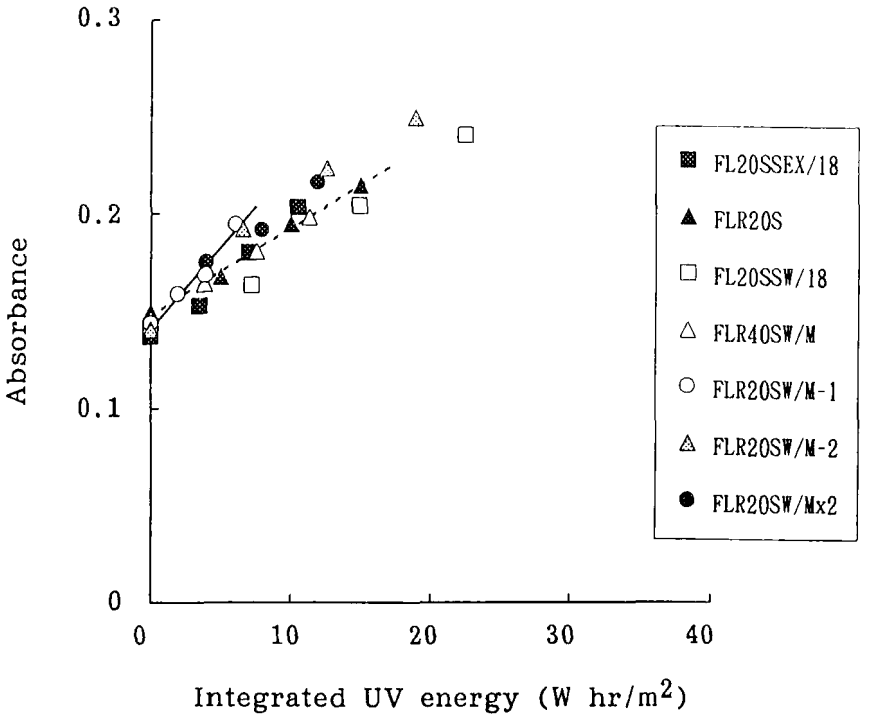

Fig. 4. Absorbance at 400 nm of quinine solutions exposed to white fluorescent lamps as a function of integrated UV energy.

Lines (——) and (- - - -) indicate the regression lines with the largest and the smallest slopes, respectively.

UV lamps (at 7 different laboratories) yielded fairly high correlation coefficients ($r>0.99$). Similarly high correlation was observed with the 3 xenon arc lamps. Correlation coefficients for white fluorescent lamps and metal halide lamps were slightly smaller, but still indicated good linearity between absorbance and integrated UV energy. These results indicated that the increase in absorbance of the quinine solutions was proportional to the integrated UV energy, and could therefore be used as a measure of the UV radiation intensity of the lamps.

Though good linearity between absorbance and integrated UV energy was indicated by the high correlation coefficients, the slopes of the regression curves varied considerably between lamps. Even slopes calculated for the same kind of lamp varied between individual lamps. This was true especially

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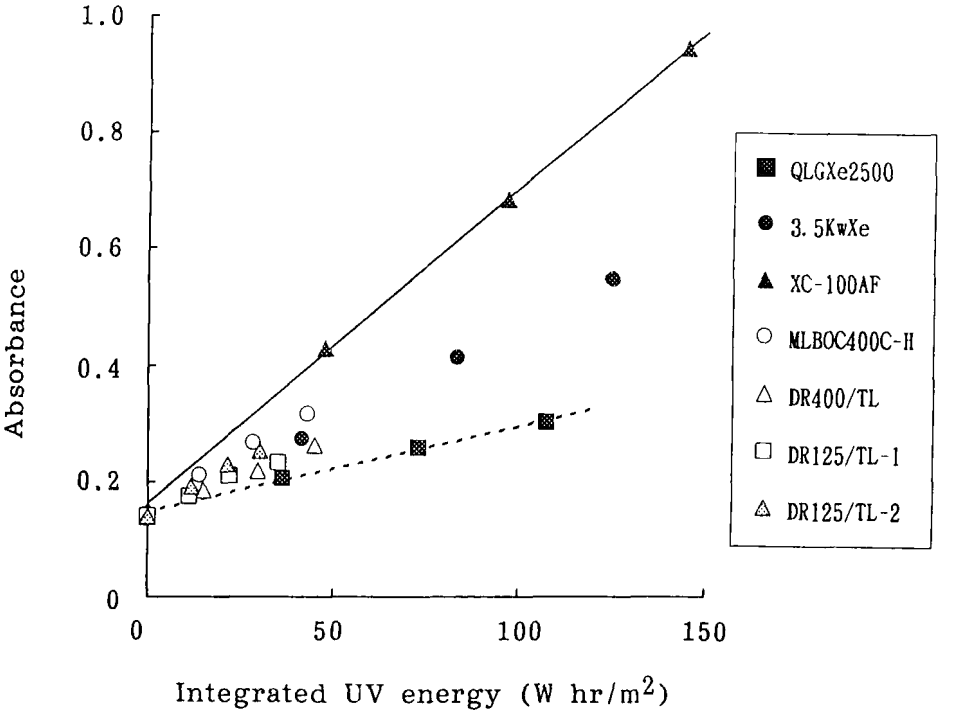

Fig. 5. Absorbance at 400 nm of quinine solutions exposed to xenon arc (■●▲) and metal halide (○△□▲) lamps as a function of integrated UV energy. Lines (——) and (----) indicate the regression lines with the largest and the smallest slopes, respectively.

for the xenon arc lamps. This variation suggests that the lamps had different spectral distributions, and that the relative proportions of radiation around 330 nm, which quinine absorbs most efficiently, varied among the lamps. If the spectral distribution of the lamps used were to coincide with that of the radiometer used to measure integrated UV energy, the slopes of regression curves for any of the lamps should be the same. The radiometer used, however, was most sensitive to radiation at approximately 370 nm, and it also detected radiation around 390 nm, a wavelength not absorbed by quinine. Consequently, different spectral distributions of the lamps may result in different slopes of regression curves.

Xenon arc lamps showed larger variation in the slopes of regression curves than did near UV fluorescent lamps, white fluorescent lamps and metal halide

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Table I.

Regression curves of absorbance vs integrated UV energy from near UV fluorescent lamps

Lamp	slope x10^3 (W hr/m ^{2})$^{-1}$	intercept	correlation coefficient (r)
FL20SBL-1	3.63	0.141	0.9942
FL20SBL-2	3.81	0.140	0.9903
FL20SBL-3	3.98	0.145	0.9990
FL20SBL-4	3.82	0.144	0.9935
FL20SBL-5	4.05	0.164	0.9994
FL40BL	2.90	0.144	0.9952
FLR20S BL/M	4.45	0.138	0.9975
average	3.81	0.145	
sd	0.47	0.009	

Table II.

Regression curves of absorbance vs integrated UV energy from white fluorescent lamps

lamp	slope x10^3 (W hr/m ^{2})$^{-1}$	intercept	correlation coefficient (r)
FL20SSEX/18	6.72	0.132	0.9924
FLR20S	4.48	0.148	0.9905
FLR20SW/Mx2	5.83	0.147	0.9840
FL20SSW/18	4.72	0.133	0.9954
FLR40SW/M	4.54	0.147	0.9851
FLR20SW/M-1	8.31	0.141	0.9615
FLR20SW/M-2	5.33	0.153	0.9848
average	5.70	0.143	
sd	0.14	0.008	

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Table III.

Regression curves of absorbance vs integrated UV energy from xenon and metal halide lamps

lamp	slope x10^3 (W hr/m ^{2})$^{-1}$	intercept	correlation coefficient (r)
QLGXe2500	1.47	0.148	0.9951
3.5kWXe	3.24	0.139	0.9988
XC-100AF	5.38	0.163	0.9988
MLBOC400C-H	3.82	0.155	0.9938
DR400/TL	2.58	0.145	0.9840
DR125/TL-1	2.57	0.146	0.9848
DR125/TL-2	3.62	0.147	0.9942
average	3.24	0.149	
sd	1.23	0.008	

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lamps. This may be due to variations in their filters which eliminate shorter wavelengths, since the original xenon arc spectral distribution should be identical among lamps.

The mechanism controlling the increase in absorbance of quinine solution upon light exposure remains unclear. HPLC of the quinine solutions exposed to light showed no peaks due to decomposition products, though a small peak at a void volume was detected for the samples exposed to UV energy greater than 100 W hr/m ^{2} (3.6x10^5 J/m ^{2}). This result indicates that a very small amount of quinine degradation brings about a significant increase in absorbance.

The five % quinine solution used in the present study may be too concentrated for use at lower temperatures because of the lower solubility of quinine hydrochloride. The increase in absorbance of a 2.5 % quinine solution exposed to a near UV fluorescent lamp was therefore compared with that of a 5.0 % solution. The slope of the regression curve obtained for the 2.5 % quinine solution was 3.47x10^{-3} (W hr/m ^{2})$^{-1}$, and was similar to that obtained for the 5.0 % solution, 3.81x10^{-3} (W hr/m ^{2})$^{-1}$. This indicated that the 2.5 % solution was concentrated enough to respond to virtually all quanta from the light source, and can be used effectively as an actinometer.

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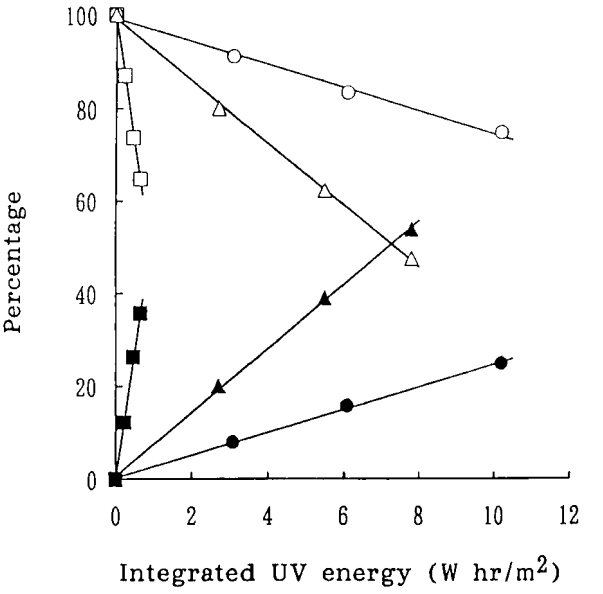

Fig. 6. Nifedipine degradation (○ △ □) and degradation product formation (● ▲ ■) as determined by HPLC after exposure to a near UV fluorescent lamp (○●), a white fluorescent lamp (□■) and a xenon arc lamp (△▲).

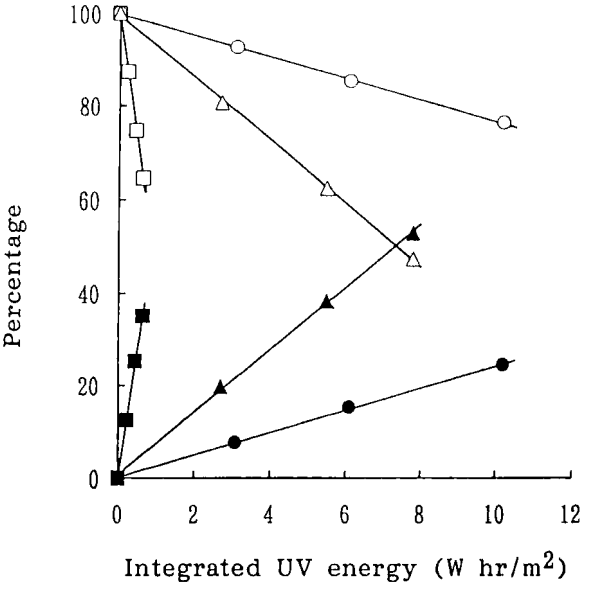

Fig. 7. Nifedipine degradation (○ △ □) and degradation product formation (● ▲ ■) as determined by absorbance changes at 450 nm and 750 nm, respectively, after exposure to a near UV fluorescent lamp (○●), a white fluorescent lamp (□■) and a xenon arc lamp (△▲).

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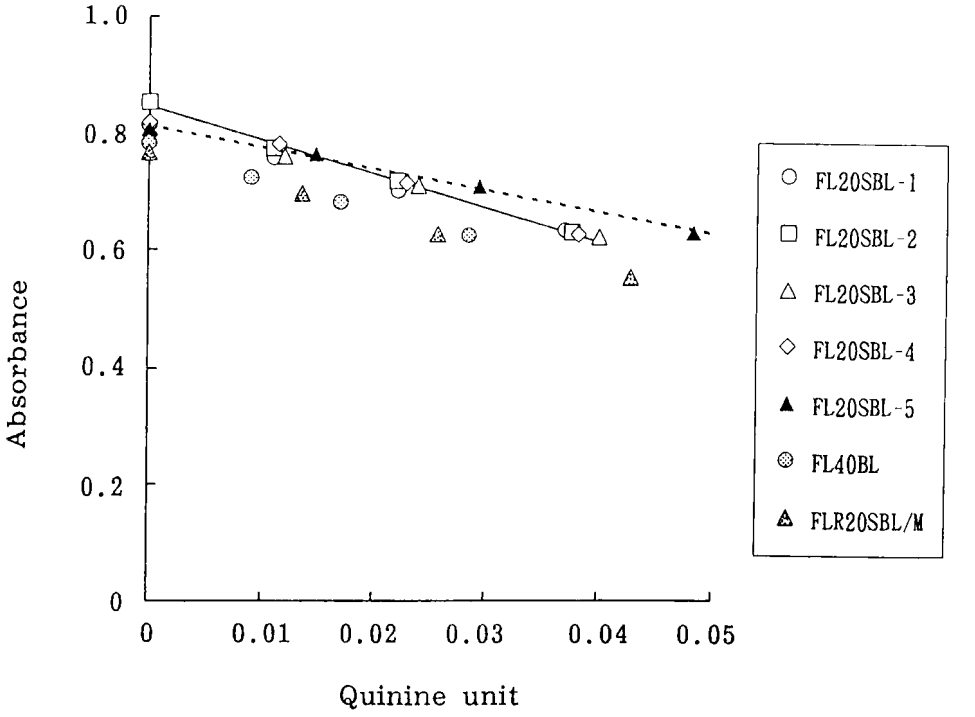

Fig. 8. Absorbance at 450 nm of nifedipine solutions exposed to near UV fluorescent lamps as a function of integrated UV energy expressed in quinine actinometric units.

Lines (——) and (----) indicate the regression lines with the largest and the smallest slopes, respectively.

Application of Quinine Actinometry to Light Stability Testing of Nifedipine

The results obtained with quinine indicate that the absorbance increase of a quinine solution is proportional to the integrated UV energy, and that quinine actinometry is useful for measuring the intensity of UV radiation at around 330 nm. To define the quinine actinometric unit, light degradation of nifedipine was studied using quinine actinometry.

Nifedipine is known to degrade upon UV and visible light exposure (3, 6). Ethanol solutions of nifedipine placed in vials were exposed to either a near UV fluorescent lamp, a white fluorescent lamp, or a xenon arc or metal halide lamp. Figure 6 shows both the amount of nifedipine remaining after light exposure, as well as the amount of degradation product determined by HPLC,

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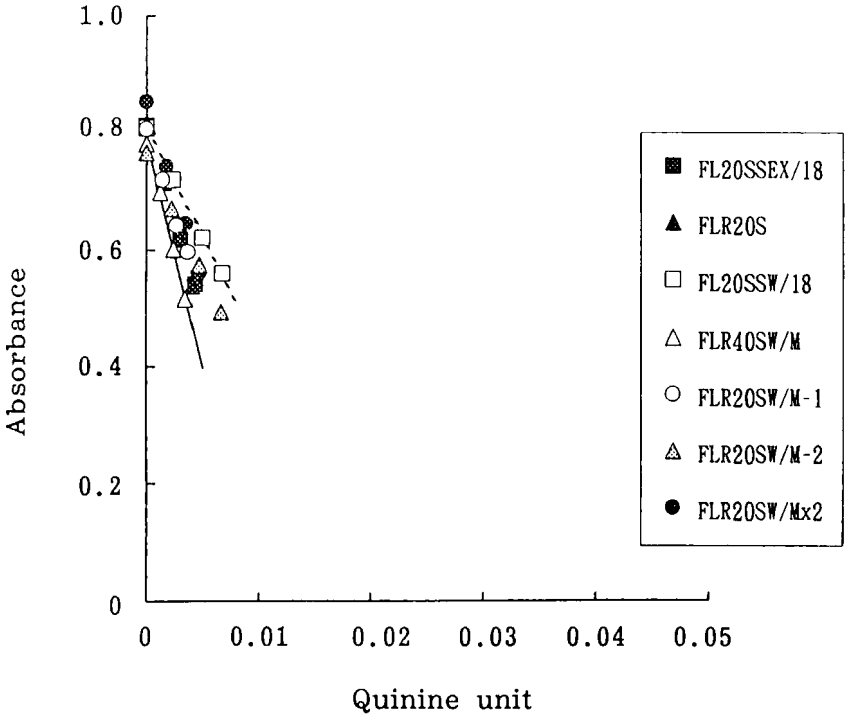

Fig. 9. Absorbance at 450 nm of nifedipine solutions exposed to white fluorescent lamps as a function of integrated UV energy expressed in quinine actinometric units.

Lines (——) and (- - - -) indicate the regression lines with the largest and the smallest slopes, respectively.

plotted agaist the integrated UV energy measured by the radiometer . The decrease in nifedipine remaining after light exposure coincided with the increase in decomposition product. As shown in Fig. 7, the absorbance at 450 nm decreased and that at 750 nm increased with an increase in integrated light intensity. The profiles of absorbance changes at 450 nm and 750 nm vs integrated light intensity agreed with the profiles of the amounts of nifedipine remaining and degradation product formed, respectively. These results indicated that the decrease in absorbance at 450 nm was due to nifedipine degradation, and thus, can be used as a measure of decomposition.

Figure 8 shows the decrease in absorbance at 450 nm of nifedipine solution plotted against the integrated intensity of radiation from a near UV fluorescent

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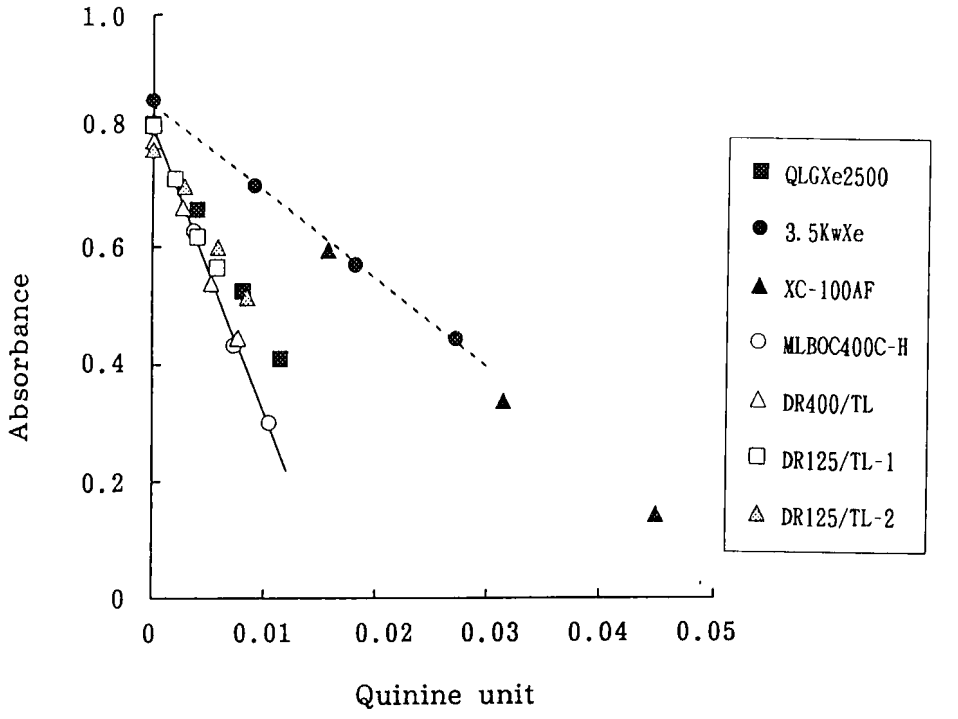

Fig. 10. Absorbance at 450 nm of nifedipine solutions exposed to xenon arc or metal halide lamps as a function of integrated UV energy expressed in quinine actinometric units.

Lines (——) and (-----) indicate the regression lines with the largest and the smallest slopes, respectively.

lamp. The increase in absorbance at 400 nm of the quinine solution after exposure (quinine actinometric unit) was used to express the integrated instensity of the radiation. Similar profiles are shown in Figs. 9 and 10 for a white fluorescent lamp, and a xenon arc or metal halide lamp, respectively.

The slopes of regression curves calculated for each lamp varied among lamps of a single type, as well as among different kinds of lamps. This indicated that the extent of nifedipine degradation was not proportional to the integrated light intensity as determined by quinine actinometry. Near UV fluorescent lamps, which emit limited radiation of visible light, caused nifedipine degradation to lesser extent than they did quinine degradation. The effect of near UV fluorescent lamps on nifedipine degradation was smaller than

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that of white fluorescent lamps, xenon arc lamps and metal halide lamps, which all have strong visible radiation. These results can be ascribed to the difference in the absorption spectra between quinine and nifedipine. Nifedipine absorbs visible, as well as UV light, while quinine absorbs only UV light.

In conclusion, quinine actinometry can be used to measure the intensity of UV radiation at around 330 nm, but not to determine the total intensity of radiation responsible for the degradation of drugs which have absorption spectra different from that of quinine. It is concluded that quinine actinometry can be a simple and reproducible standard method for calibrating the intensity of UV radiation at around 330 nm from the light sources used in light-stability testing of pharmaceutical products.

References

1. J. N. Pitts, Jr., J. K. S. Wan and E. A. Schuck, J. Am. Chem. Soc., 86, 3606 (1964)
2. N. Numao, T. Hamada and O. Yonemitsu, Tetrahedron Lett., 19, 1661 (1977)
3. K. Akimoto, K. Kurosaka, H. Nakagawa and I. Sugimoto, Chem. Pharm. Bull., 36, 1483 (1988)
4. J. T. Piechocki and R. J. Wolters, Pharmaceutical Tech., 47 (1993)
5. C. G. Hatchard and C. A. Parker, Proc. Royal Soc. London, A235, 518 (1956)
6. I. A. Majeed, W. J. Murray, D. W. Newton, S. Othman and W. A. Al-turk, J. Pharm. Pharmcol., 39, 1044 (1987)

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