Pergamon

Talanta, Vol. 41, No. 2, pp. 233–236, 1994

0039-9140/94 $6.00 + 0.00

SOLVENT EFFECT ON THE DETERMINATION OF SULFAMETHAZINE BY ROOM-TEMPERATURE PHOTOCHEMICALLY INDUCED FLUORESCENCE

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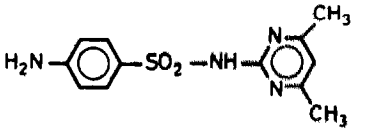
(*Received* 10 *June* 1993. *Revised* 15 *July* 1993. *Accepted* 2 *August* 1993)

Summary—Room-temperature photochemically-induced fluorescence (RTPF) was applied to the determination of sulfamethazine (SMT) in methanol, ethanol and 2-propanol. Optimal ultraviolet irradiation times ranged between 2 and 6 min. Linear calibration graphs were obtained over a concentration range of more than one order of magnitude. The relative standard deviations were within the range 1.4–2.2%. Limits of detection were between 40 and 80 ng/ml. The method was evaluated for its applicability to the analysis of SMT in pharmaceutical formulations.

Sulfamethazine (SMT) (sulfadimidine) (I) belongs to the group of pharmaceutically-important heterocyclic sulfonamides. This



(I)

compound is widely employed in medicine and veterinary practice as antibacterial drug in pharmaceutical preparations. Several analytical methods have been proposed for its determination and that of other sulfonamides.[1–14]

An interesting property of sulfonamides is their photochemical reactivity.[15,16] Recently, we utilized this photochemical behaviour for determining SMT and other heterocyclic derivatives spectrofluorimetrically.[17–19] We developed a room-temperature photochemically-induced fluorescence (RTPF) method based on ultraviolet (UV) irradiation of sulfonamides in aqueous medium and rapid formation of strongly fluorescent photoproducts.[17] Irradiation times required were between 10 and 30 min, and concentrations of 0.25–3.0 μg/ml could be quantitated. Also, we combined RTPF with flow injection analysis,[18] and applied the technique to the analysis of SMT in pharmaceutical compounds and food.[19]

In this paper, we report on the effects of alcoholic solvents on the RTPF determination of SMT. The use of these non-aqueous media produced a marked decrease of the UV irradiation time. We applied the technique to the analysis of SMT in pharmaceutical preparations.

EXPERIMENTAL

Reagents

Sulfamethazine was purchased from Sigma. Analytical-reagent grade (Aldrich) methanol, ethanol and 2-propanol were used to prepare 200 μg/ml SMT stock solutions, and to make serial dilutions. Pharmaceutical preparation of SMT (sulphadimerazine 33% Noé) was a gift from Lab. Noé-Socopharm (Château-Thierry, France).

Apparatus

A Perkin-Elmer model LS-5 spectrophotofluorometer was used for the fluorescence measurements. An Osram 200-W mercury arc lamp with an Oriel Model 8500 power supply was utilized for the photolysis of SMT. The RTPF experimental set-up was as described previously.[20]

Procedure

An aliquot of each SMT sample was placed in a 1-cm quartz cuvette and irradiated at room temperature with the UV light of the mercury

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Table 1. Experimental conditions for determination of sulfamethazine in alcohols

Solvent	λ (nm) Excitation	Emission	t_{irr}^{OPT}* (min)
Methanol	260	341	3
Ethanol	290	345	6
2-Propanol	290	345	2

*t_{irr}^{OPT} = Optimal irradiation time corresponding to the maximum fluorescence signal.

arc lamp for a fixed time. Fluorescence intensity measurements were performed at constant excitation and emission wavelengths, using optimal irradiation time values depending of the solvent (Table 1).

RESULTS AND DISCUSSION

Effect of solvent

The fluorescence excitation and emission maxima wavelengths and optimal irradiation times are reported in Table 1 for SMT in methanol, ethanol and 2-propanol. A 30-nm red-shift of the SMT excitation wavelength was observed upon going from methanol to ethanol and 2-propanol, whereas no significant shift of the emission wavelength was noted when changing of solvent. Upon UV irradiation, SMT did not exhibit any important change in the shape of the emission spectra for all solvents (Figs 1 and 2), but a 2- to 5-fold increase of the fluorescence signal occurred (Figs 3 and 4). The optimal irradiation times, corresponding to the maximum fluorescence intensity, were found to be 3, 6 and 2 min in methanol, ethanol and 2-propanol, respectively. These values, obtained

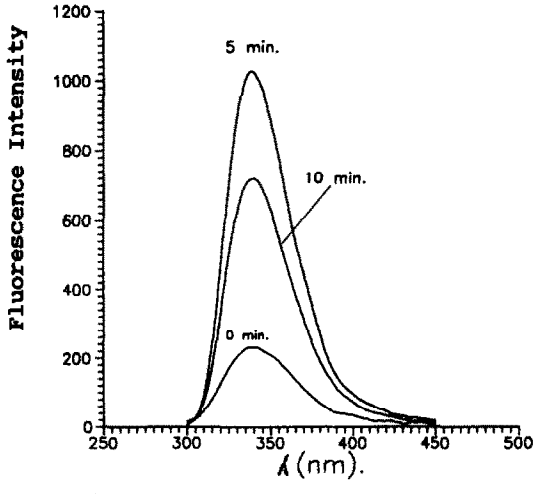

Fig. 1. Fluorescence emission spectra of non-irradiated and irradiated 1.5 μg/ml sulfamethazine in methanol. Curve 1: non-irradiated sulfamethazine; curves 2 and 3: sulfamethazine irradiated during 5 and 10 min.

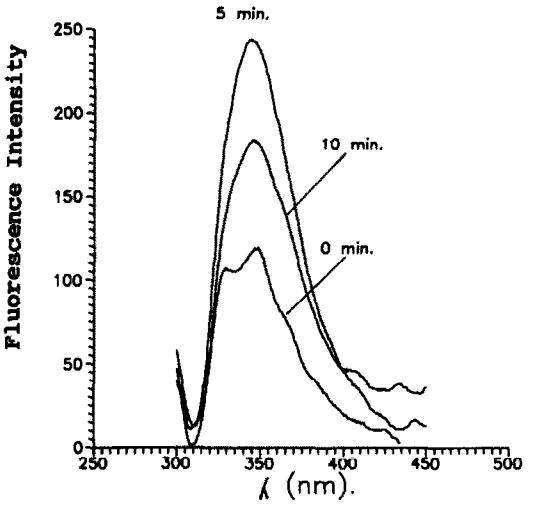

Fig. 2. Fluorescence emission spectra of non-irradiated (curve 1) and irradiated 1.0 μg/ml sulfamethazine in 2-propanol; curves 2 and 3: sulfamethazine irradiated during 5 and 10 min.

in alcoholic media, are significantly shorter than that of 10 min determined previously for SMT in water.[17] It demonstrates the advantage of utilizing an alcohol rather than an aqueous solvent for improving the speed of the RTPF determination of SMT.

Analytical figures of merit

The analytical figures of merit for the determination of SMT in methanol, ethanol and 2-propanol are given in Table 2. Linear calibration plots were established over a concentration range of more than one order of magnitude in all solvents. The correlation coefficients were close to unity, indicating that

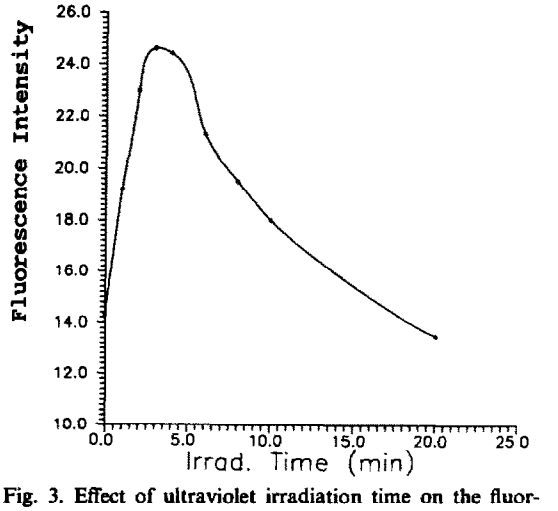

Fig. 3. Effect of ultraviolet irradiation time on the fluorescence intensity of 1.5 μg/ml sulfamethazine in methanol.

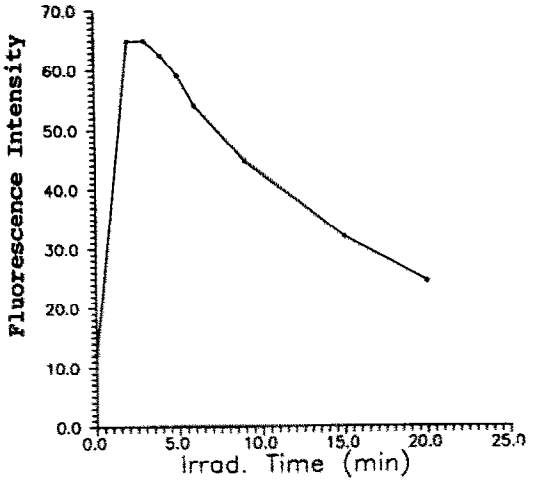

Fig. 4. Effect of ultraviolet irradiation time on the fluorescence intensity of 1.0 μg/ml sulfamethazine in 2-propanol.

Table 3. Determination of sulfamethazine in a pharmaceutical preparation*

Solvent	SMT (*ppm*) Added	Found	Recovery (%)
Methanol	—	3.32	—
	0.80	4.08	99
	1.92	5.12	98
	2.40	5.55	97
Ethanol	—	0.61	—
	0.60	1.17	97
	1.00	1.50	93
	2.00	2.56	98
2-Propanol	—	0.47	—
	0.60	1.07	100
	1.00	1.47	100
	1.40	1.87	100

*Sulphadimerazine 33% Noé.

the precision of analytical curves is excellent. The RSD values ranged between 1.4 and 2.2%. The limits of detection (LODs) were very low, between 40 and 80 ng/ml, according to the alcohols used. These values are markedly smaller than the LODs of 74 ng/ml and 120 ng/ml, obtained, respectively for the RTPF and RTPF–FIA determination of SMT in water.[18,19] These data indicate that an alcoholic media results in an increase in the sensitivity and precision of the RTPF determination of SMT.

Analytical applications

In order to confirm the analytical applicability of the RTPF method in alcoholic solvents, SMT was determined in the Noé 33 pharmaceutical formulation, using the standard addition procedure. The pharmaceutical formulation was dissolved and diluted in the different alcohols used. Satisfactory results were obtained in methanol, ethanol and 2-propanol, with recoveries ranging from 93 to 100% according to the solvent. These recoveries values are comparable to those obtained previously in water.[19] It demonstrates that the method does not suffer significantly of interferences from the pharmaceutical matrix used.

RTPF method can be utilized for the quality control of pharmaceutical formulations containing SMT. In addition, RTPF could be applied to the analysis of SMT in food, without noticeable interference, as found previously.[19]

CONCLUSION

We have shown that the analytical usefulness of room-temperature photochemically-induced fluorescence is significantly improved in alcoholic solvents for the determination of SMT. The use of alcoholic media results in a more rapid RTPF method with improved sensitivity and precision for analysing SMT relative to that obtained in an aqueous solvent. Typically, an irradiation time of 2 min with a lower limit of detection of 40 ng/ml was possible in 2-propanol. Thus, this solvent is recommended for the RTPF determination of sulfamethazine, especially in pharmaceutical formulations.

Table 2. Analytical figures of merit for the photochemical-fluorimetric determination of sulfamethazine in alcohols solvents

Solvent	Concentration range (μg/ml)	Regression equation*	Correlation coefficient	Limit of detection (ng/ml)†	RSD‡ (%)
Methanol	0.5–6.4	$I_F = 642.8c + 2.5$	0.999	80	1.4
Ethanol	0.3–3.0	$I_F = 777.6c + 45.0$	0.996	40	2.2
2-Propanol	0.6–2.6	$I_F = 731.4c + 6.3$	0.992	70	2.0

*I_F = Relative fluorescence signal; c = analyte concentration.

†Limit of detection defined as the concentration of solution giving a signal-to-noise (S/N) ratio of 3.

‡RSD = Mid-range relative standard deviation (n = 4–6).

REFERENCES

1. A. C. Bratton and E. K. Marshall, *J. Biol. Chem.*, 1939, **128**, 537.
2. F. Salinas, A. Espinosa Mansilla and J. J. Berzas Nevado, *Anal. Chim. Acta*, 1990, **233**, 289.
3. J. Rieder, *Chemotherapy (Basel)*, 1972, **17**, 1.
4. A. G. Xanakis and M. I. Karayannis, *Anal. Chim. Acta*, 1984, **159**, 343.
5. H. B. Falk and R. G. Kelly, *Clin. Chem.*, 1965, **11**, 1045.
6. A. Bye and A. F. J. Fox, *Clin. Chem.*, 1974, **20**, 288.
7. M. E. Abdellatef, M. N. Elbakiny and A. A. Boulkeir, *J. Pharm. Biomed. Anal.*, 1989, **7**, 571.
8. S. Othman, *Int. J. Pharm.*, 1990, **63**, 173.
9. K. K. Verma and K. K. Stewart, *Anal. Chim. Acta*, 1988, **214**, 207.
10. A. Al-Wehaid and A. A. Townshend, *Anal. Chim. Acta*, 1986, **186**, 289.
11. J. W. Bridges, L. A. Gifford, W. P. Hayes, J. N. Miller and D. Thornburn Burns, *Anal. Chem.*, 1974, **46**, 1010.
12. L. Van Poucke, D. Rousseau, B. De Spiegeleer and C. Van Peteghem, *Agric. Food Chem., Consum. Proc. Eur. Conf. Food Chem.*, 5th Ed., 1989, **2**, 438, INRA, Paris, France.
13. M. D. Smedley and J. D. Weber, *J. Assoc. Off. Anal. Chem.*, 1990, **73**, 875.
14. S. Horii, C. Momma, K. Miyahara, T. Maruyama and M. Matsumoto, *J. Assoc. Off. Anal. Chem.*, 1990, **73**, 990.
15. T. Ahmad, *Pharmazie*, 1982, **37**, 559.
16. S. Tammilehto and M. Lehtonen, *2nd Int. Symp. on Drug Analysis, Abstracts*, 1986, Brussels.
17. M. C. Mahedero and J. J. Aaron, *Analusis*, 1992, **20**, 53.
18. M. C. Mahedero and J. J. Aaron, *Anal. Chim. Acta*, 1992, **269**, 193.
19. M. Sanchez Pena, F. Salinas, M. C. Mahedero and J. J. Aaron, *J. Pharm. Biomed. Anal.*, 1992, **10**, 805.
20. J. J. Aaron, in *Molecular Luminescence Spectroscopy, Methods and Applications*, Part 3, S. G. Schulman (ed.). Wiley, New York, 1993, 85.