Stability-Indicating Assay for Tolmetin Sodium in Solid Dosage Forms

CASIMIR A. JANICKI ˣ **and KATHERINE F. DALY** *

Received December 12, 1978, from the *Pharmacy Research Department, McNeil Laboratories, Fort Washington, PA 19034.* Accepted for publication August 22, 1979. *Present address: Instrument Products, E. I. du Pont de Nemours and Co., Wilmington, DE 19898.

Abstract □ A spectrophotometric assay for determining tolmetin sodium in pharmaceutical solid dosage forms is described. Tolmetin sodium is separated from common pharmaceutical excipients and probable degradation products. Recovery, precision, and accuracy data are provided. Two TLC methods are included which can be used to monitor qualitatively the stability of aged dosage forms.

Keyphrases □ Tolmetin sodium—stability-indicating assay, solid dosage forms □ Anti-inflammatory agents—tolmetin sodium, stability-indicating assay, solid dosage forms □ Stability—tolmetin sodium, spectrophotometric assay, solid dosage forms

Tolmetin[1] sodium [sodium 1-methyl-5-(4-methylbenzoyl)-1*H*-pyrrole-2-acetate dihydrate] was introduced commercially as an anti-inflammatory drug for the treatment of rheumatoid arthritis. Since it is a pyrrole derivative (1), it differs chemically from other anti-inflammatory agents. During the development of a suitable dosage form, a precise and accurate stability assay was needed. GLC and spectrophotometric assays were developed (2–4) to evaluate the absorption, distribution, and excretion of tolmetin sodium in human subjects. High-pressure liquid chromatographic methods were developed recently for assessing the purity of tolmetin sodium (5) and for determining tolmetin and its major metabolite in plasma (6).

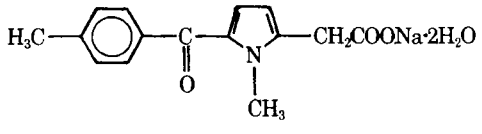
A spectrophotometric assay is described that can determine tolmetin sodium in the presence of probable degradation products and common pharmaceutical tablet and capsule excipients. Precision and accuracy data also are provided. The method is quick and simple, requires only readily available equipment, and has the specificity needed for stability studies. Two TLC methods also are included which can be used to monitor qualitatively the stability of aged dosage forms.

EXPERIMENTAL

Reagents—All reagents met ACS specifications. Reference standard tolmetin sodium[2] was used as the standard. The TLC plates were 5 × 20-cm precoated silica gel GF[3]. Commercial tolmetin sodium tablets containing the equivalent of 200 mg of tolmetin were used[2].

Standard Preparation—Tolmetin sodium reference standard, 122.6 mg (equivalent to 100.0 mg of tolmetin), was placed in a 120-ml screw-capped bottle. Water, 25 ml, was added, and the mixture was swirled. Ethylene dichloride, 50 ml, and 5 ml of 6 *N* HCl were added by pipet. The standards were analyzed as described under *Procedure*.

Procedure—At least 20 tablets were weighed and finely powdered. A portion of the powder equivalent to 100 mg of tolmetin was weighed accurately and transferred to a 120-ml screw-capped bottle. Ethylene dichloride, 50 ml, and 25 ml of 1 *N* HCl were added by pipet, and the mixture was shaken mechanically[4] for 15 min.



tolmetin sodium dihydrate

The solution was centrifuged to separate the phases and aspirated, and the aqueous layer was discarded. Five milliliters of the ethylene dichloride layer (2 mg/ml) was pipetted into a 100-ml volumetric flask, and the solution was diluted to volume with ethylene dichloride and mixed (0.1 mg/ml). Five milliliters of this solution was pipetted into a 120-ml screw-capped bottle containing exactly 50 ml of 0.1 *N* NaOH. The solution was shaken mechanically for 30 min and centrifuged. Then the sodium hydroxide layer was transferred to a clean container (0.01 mg/ml).

The UV absorption spectrum of the sample was recorded concomitantly with the reference standard in 1-cm cells, using 0.1 *N* NaOH saturated with ethylene dichloride as the blank. The percent tolmetin was calculated by:

$$\% \text{ tolmetin} = \frac{A_u}{A_s} \times \frac{C_s}{C_u} \times 100 \qquad \text{(Eq. 1)}$$

where A_u and A_s are the absorbances of the sample and standard, respectively, at the wavelength of maximum absorbance near 322 nm and C_s and C_u are the concentrations in milligrams per milliliter of the standard and sample, respectively.

Recovery—A stock solution of 24.6 mg of tolmetin sodium/ml of methanol was prepared. Two 5-ml samples were evaporated to dryness. One sample was assayed by the described procedure, and the second sample was diluted directly to the same final concentration with 0.1 *N* NaOH. Thirteen such experiments were run by five analysts.

Specificity—Twenty-five milligrams of I (*p*-toluic acid), III, IV, IX, and XI (Table I) were weighed directly into 120-ml screw-capped bottles and assayed as described.

Precision and Accuracy—An aqueous stock solution containing 1.23 mg of tolmetin sodium/ml was prepared, and 25-ml portions were assayed. The dilution was changed accordingly in the second assay step. Twelve assays were performed by six analysts.

In a second experiment, 40 tablets from a stability test program were ground to a fine powder and samples were assayed. Sixteen assays were performed by seven analysts.

Hydrolysis—Tolmetin sodium samples, 250 mg, were added to separate round-bottom flasks containing 50 ml of 1 *N* NaOH, water, or 1 *N* HCl. The samples were refluxed for 72, 72, or 24 hr, respectively.

Another sample of 615 mg of tolmetin sodium was dissolved in 500 ml of water and placed in a water-jacketed quartz immersion well and irradiated with a high-pressure mercury arc lamp for 24 hr. The resulting solutions and precipitants were assayed for intact tolmetin.

TLC—Tolmetin sodium and the 10 test compounds (Table I) were dissolved in methanol. The TLC chambers were 1.89-liter, wide-mouth Mason jars fitted with standard lids. The solvent systems were acetic acid–chloroform (5:95 v/v) (A) and *n*-butanol saturated with ammonia test solution (USP XIX) (B). For the *n*-butanol–ammonia solution, the plates were developed in preequilibrated paper-lined jars. Each plate was developed until the solvent front was ~2.5 cm from the top. The plates were air dried and viewed under short-wavelength UV light. The plates then were stained with iodine vapor. The plates also may be scanned with a densitometer[5].

RESULTS AND DISCUSSION

To determine the specificity of the assay for tolmetin sodium, I, III, IV, IX, and XI were assayed. The resulting UV spectra showed absorbance only from the sample derived from I. Since there was no interference

[1] Tolectin, McNeil Laboratories.
[2] McNeil Laboratories.
[3] Analtech Inc., Wilmington, Del.
[4] Wrist-action shaker, Burrell Corp., Pittsburgh, Pa.
[5] Schoeffel model SD3000 spectrodensitometer.

0022-3549/80/0200-0147$01.00/0

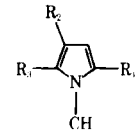


Table I—TLC R_f Values of the Test Compounds[a]

Compound	R_1	R_2	R_3	R_f Values Solvent A	R_f Values Solvent B
I (*p*-toluic acid)	—	—	—	0.69	0.78
II	$CH_2COOC_2H_5$	H	*p*-$CH_3C_6H_4CO$	0.66	0.79
III	CH_2COOCH_3	H	*p*-$CH_3C_6H_4CO$	0.66	0.80
IV	CH_2COOH	H	H	0.67	0.42
V	COOH	*p*-$CH_3C_6H_4CO$	CH_3	0.66	0.82
VI (tolmetin)	CH_2COOH	H	*p*-$CH_3C_6H_4CO$	0.54	0.50
VII	$CONH_2$	*p*-$CH_3C_6H_4CO$	CH_3	0.37	0.75
VIII	CH_2COOH	*p*-$CH_3C_6H_4CO$	H	0.29	0.44
IX	CH_2CONH_2	H	*p*-$CH_3C_6H_4CO$	0.18	0.92
X	CH_2CONH_2	*p*-$CH_3C_6H_4CO$	H	0.07	0.64
XI	CH_3	H	*p*-$CH_3C_6H_4CO$	0.68	0.79

[a] Detection limits under UV light ranged from 0.1 to 0.5 μg.

from *p*-toluic acid from 360 to 275 nm (Fig. 1), this compound does not interfere in the tolmetin sodium assay. The other compounds either did not extract into the sodium hydroxide layer or, in the case of IV, did not have any UV absorbance.

Compound I is *p*-toluic acid, and IV is the pyrrole acetic acid fragment of the tolmetin molecule. Both compounds are probable degradation products of tolmetin sodium. Compound III is the methyl ester of tolmetin, IX is the amide of tolmetin and a precursor in tolmetin synthesis, and XI is the decarboxylated tolmetin product. Compound III was used as a model compound in the event that esterification became a degradation route.

Since the five model compounds could be considered only as possible degradation or reaction products, attempts were made to degrade tolmetin sodium. Tolmetin sodium solutions were hydrolyzed as discussed under *Experimental*. The alkaline hydrolysis solution assayed 88% for intact tolmetin sodium, while the water hydrolysis solution assayed 94%. The solution from the attempted UV light degradation assayed 82%. TLC data showed many minor degradation products, but their identification was not attempted.

The acid solution was orange and contained a violet precipitate. The acid hydrolysis mixture assayed 64% for intact tolmetin sodium. The major TLC spots were at the origin and at R_f 0.5 and 0.7 using acetic acid–chloroform. With UV and IR, the sample at R_f 0.5 was identified as tolmetin, and the spot at R_f 0.7 was *p*-toluic acid. The spot at the origin was believed to be a highly polar molecule or a species of relatively high molecular weight containing many polar groups. *N*-Methylpyrroles are known to form polymers in acidic media, which are highly colored when polymerized under aerobic conditions (7).

To demonstrate the efficiency of the tolmetin sodium extraction, an aqueous stock solution of tolmetin sodium was prepared. Five analysts assayed 13 portions of it as directed for the tablets, and other portions were diluted simultaneously with 0.1 *N* NaOH. The data (Table II) indicated essentially complete drug extraction.

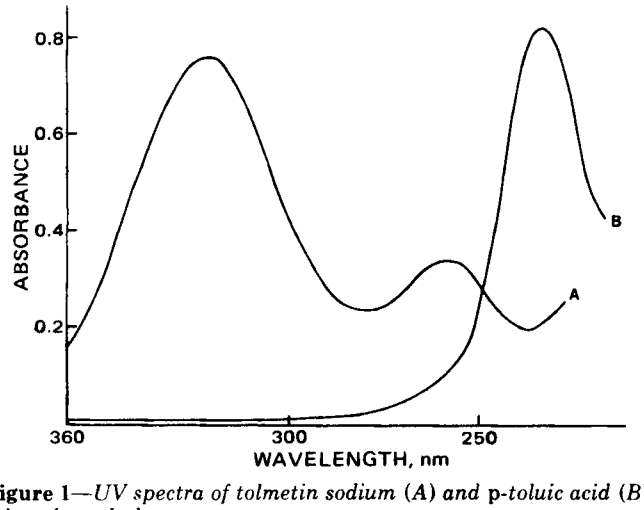


Figure 1—*UV spectra of tolmetin sodium (A) and p-toluic acid (B) taken through the assay.*

Another experiment was conducted to demonstrate the efficiency of the extraction for powdered tablet samples. Duplicate tablet powder samples were taken through the assay along with duplicate samples extracted with 25- and 75-ml samples of ethylene dichloride. The acid solution volume remained as described. The assay varied only in the dilution step with the last extraction step remaining as described. The mean of the six assays was 99.7% label with a standard deviation of 0.16.

To show that the initial extraction step in the assay did not affect tolmetin stability, reference standard tolmetin sodium was treated as described under *Experimental*, except that sufficient 12 *N* HCl was added to make the aqueous layer 3 *N* instead of 1 *N*. The solution was shaken, and samples were removed after 1 and 24 hr for TLC analysis. Using a densitometer, <0.2% degradation was determined after 1 hr and <0.8% was determined after 24 hr. Less than 0.1% degradation was detected by TLC using the method under *Procedure*.

Precision and accuracy were tested initially on an aqueous tolmetin

Table II—Recovery Data for Tolmetin Sodium

Analyst	Absorbance at Maximum near 322 nm Through Assay	Diluted Directly
1	0.756	0.765
	0.753	0.763
2	0.763	0.761
	0.756	0.761
3	0.757	0.759
	0.761	0.763
4	0.759	0.764
	0.775	0.764
	0.768	0.768
5	0.772	0.777
	0.772	0.783
	0.773	0.782
	0.766	0.778
Mean	0.764	0.769
SD	0.0075	0.0084

Table III—Precision and Accuracy Data

Analyst	Label Amount, % Solution	Tablets
1	100.8, 100.9	99.5, 99.6
2	100.1, 99.8	102.2, 101.9
3	100.7, 100.3	100.2, 99.2
4	100.1, 99.4	98.3, 98.8
5	99.9, 99.7	99.7, 99.9, 99.6
6	99.9, 100.4	100.1, 100.5
7		98.9, 99.4, 98.6
Mean	100.2	99.8
SD	0.47	1.07

sodium solution. The data had an accuracy of 100.2% with a standard deviation of 0.47% for the 12 assays (Table III). The experiment was conducted over 18 days. A precision study was conducted on a composite sample obtained by grinding 40 commercial tablets. The mean from the 16 assays was 99.8% over 2 months with a standard deviation of 1.07%. The data demonstrated that the method gives precise and accurate data.

Common tablet and capsule excipients such as starch, lactose, cellulose, and stearate lubricants were taken through the assay and did not interfere.

The assay was used to follow the stability of aged tablets and capsules containing up to an equivalent of 400 mg of tolmetin (490.1 mg of tolmetin sodium). Tablets were assayed for up to 5 years at room temperature and at various temperatures up to 80° for 3 months. The stability data demonstrated that the tablets and capsules were very stable. TLC was conducted on the aged samples using the two solvent mixtures. The data showed that tolmetin sodium remained intact in the dosage forms even after aging for 5 years at room temperature and accelerated aging at temperatures up to 80°. Less than 2% degradation was noted under the accelerated conditions.

Two solvent mixtures of different polarity were chosen for TLC to minimize the possibility of unresolved degradation products that could not be predicted. The compounds chosen to demonstrate the specificity of the TLC methods were, with one exception, previously postulated to be theoretical impurities in tolmetin sodium (5). The data (Table I) demonstrated that the solvent systems are suitable for separating tolmetin sodium from possible degradation products as well as theoretical impurities. Further confirmation was made on an aged tablet sample that was chromatographed and isolated from the TLC plate. The material was found to be identical to tolmetin sodium by UV, IR, and mass spectrometry.

The assay was used additionally to follow the stability of tolmetin sodium solutions buffered from pH 4.5 to 10.0 at 80°[6]. The data supported the reflux experiments, which showed that tolmetin sodium is relatively stable at neutral and alkaline pH values but unstable at acidic pH.

The data demonstrated that the method is capable of monitoring the stability of tolmetin sodium in solid dosage forms. The method is precise and accurate and utilizes common laboratory equipment. The hydrolysis experiments illustrated that tolmetin is stable under neutral and alkaline conditions but not under acidic conditions. The TLC methods can monitor qualitatively tolmetin sodium stability in aged dosage forms.

REFERENCES

(1) J. R. Carson, D. N. McKinstry, and S. Wong, *J. Med. Chem.*, **14**, 646 (1971).

(2) M. L. Selley, J. Thomas, and E. J. Triggs, *J. Chromatogr.*, **94**, 143 (1974).

(3) W. A. Cressman, B. Lopez, and D. Sumner, *J. Pharm. Sci.*, **64**, 1965 (1975).

(4) K. T. Ng, *J. Chromatogr.*, **166**, 527 (1979).

(5) R. K. Gilpin and C. A. Janicki, *ibid.*, **147**, 501 (1978).

(6) J. W. Ayres, E. Sakmar, M. R. Hallmark, and J. G. Wagner, *Res. Commun. Chem. Pathol. Pharmacol.*, **16**, 475 (1977).

(7) G. F. Smith, in "Advances in Heterocyclic Chemistry," vol. 2, A. R. Katritzky, Ed., Academic, New York, N.Y., 1963, pp. 287–309.

ACKNOWLEDGMENTS

The authors acknowledge the technical assistance of Mrs. R. Rocco, Dr. J. S. Ranweiler, Dr. C. Y. Ko, Mrs. C. M. Moral, Mr. L. T. Olszewski, and Dr. R. H. Erlich.

[6] W. A. Cressman, McNeil Laboratories, Fort Washington, PA 19034, personal communication.

Preparation and Biological Actions of *N*-2,2,2-Trifluoroethyl-2-(3,4-dihydroxyphenyl)ethylamine

JOHN P. O'DONNELL *x, A. J. AZZARO ‡, and P. R. URQUILLA ‡

Received April 9, 1979, from the **School of Pharmacy and the* ‡*Departments of Neurology and Pharmacology, School of Medicine, West Virginia University Medical Center, Morgantown, WV 26506.* Accepted for publication August 22, 1979.

Abstract □ *N*-2,2,2-Trifluoroethyl-2-(3,4-dihydroxyphenyl)ethylamine was synthesized and compared to *N*-ethyl-2-(3,4-dihydroxyphenyl)-ethylamine and dopamine for activity on adenylate cyclase in the rat striatum. Both dopamine and *N*-ethyl-2-(3,4-dihydroxyphenyl)ethylamine stimulated adenylate cyclase activity in a dose-dependent fashion. The *N*-trifluoroethyldopamine analog at 1×10^{-4} *M* induced a weak effect. The compounds were evaluated further by studying their relaxant effects in isolated rabbit renal and ear arteries. Both the *N*-ethyl- and *N*-trifluoroethyldopamine analogs produced a relaxant effect but demonstrated no selectivity for dopamine receptors.

Keyphrases □ Adenylate cyclase activity—dopamine, *N*- and *N,N*-substituted dopamine analogs, peripheral and central dopamine receptors, isolated striata and blood vessels □ Dopamine—antiparkinson drug, adenylate cyclase activity, dopamine analogs □ *N*-Ethyl-2-(3,4-dihydroxyphenyl)ethylamine—dopamine analog, adenylate cyclase activity □ *N*-2,2,2-Trifluoroethyl-2-(3,4-dihydroxyphenyl)ethylamine—dopamine analog, adenylate cyclase activity

The treatment of parkinsonism with levodopa has beneficial effects; however, in some patients with severely impaired dopaminergic neurons, the use of levodopa may be ineffective because the tissue cannot perform the necessary enzymatic decarboxylation to form dopamine (1, 2). An effort to generate new dopaminergic drugs for parkinsonism led to the synthesis of *N*- and *N,N*-substituted dopamine analogs (3–9). The binding of simple dopaminergic ligands to the receptor appears to be independent of substitution on the nitrogen (5). Unlike dopamine, the *N*- and *N,N*-substituted dopamine analogs cross the blood–brain barrier (3, 4). In addition, these agents are resistant to deamination by monoamine oxidase since they are secondary and tertiary amines. Moreover, these *N*- and *N,N*-substituted dopamine analogs act in the central nervous system as direct acting dopamine receptor agonists and, therefore, are independent of prejunctional dopaminergic mechanisms (4, 8).

To improve absorption and distribution of the *N*-alkyl-substituted dopamine analogs, *N*-2,2,2-trifluoroethyl-2-(3,4-dihydroxyphenyl)ethylamine (I) was prepared. The 2,2,2-trifluoroethyl group is well suited to replace the ethyl group because of similar steric properties and because the fluorine atom increases both lipid solubility and membrane penetration.

The agonistic properties of I, *N*-ethyl-2-(3,4-dihy-

0022-3549/80/0200-0149$01.00/0