

As. 115.541

SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO



No. 11-003796- -00005-0000

Señor

**DIRECTOR DIVISIÓN DE NUEVAS CREAM**

**SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO**

Fecha: 2013-02-19 16:07:45 Dep. 2020 DIR.NUEVASCR  
Tra. 11 PATENTEIYII Eve: 379 FASENACIONALII  
Act. 523 RESPUESTA Folios: 18

E.

S.

D.

REF: Expediente No. **11 3796**– solicitud de patente de invención a favor de **FAES FARMA, S.A.** Título **DERIVADOS DE 5-(4-METANOSULFONILFENIL)-TIAZOL PARA EL TRATAMIENTO DE ENFERMEDADES INFLAMATORIAS AGUDAS Y CRÓNICAS.**

**RESPUESTA A EXAMEN DE FONDO**

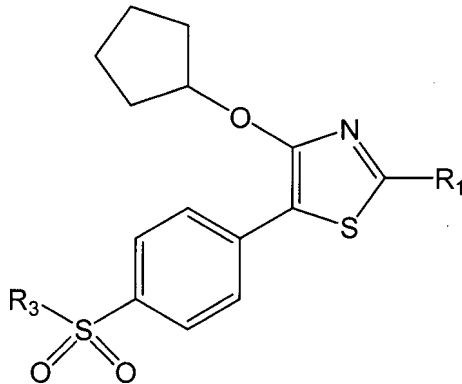
**JUAN PABLO CADENA SARMIENTO**, portador de la tarjeta profesional No. 57.492 del C.S.J., obrando como apoderado de la sociedad solicitante me permito dar respuesta al oficio número 21360 relacionado con la patentabilidad de la presente invención y notificado por fijación en lista el 27 de Noviembre de 2012.

Nos permitimos presentar a consideración del señor examinador un nuevo pliego reivindicatorio, compuesto por cinco (5) reivindicaciones, en donde no se amplía en ningún momento la materia inicialmente divulgada, y se encuentra enteramente sustentado en la descripción.

Nos permitimos informar que el nuevo pliego reivindicatorio ha sido debidamente modificado con el fin de superar las objeciones emitidas por su despacho en relación con los usos o segundo usos, mediante la eliminación de las reivindicaciones 1-11, 17 y 18. Por lo tanto, consideramos que el nuevo pliego reivindicatorio se compone de materia plenamente patentable de acuerdo con la Decisión 486.

De esta manera, la nueva reivindicación 1 establece,

*1. Un compuesto de fórmula (I') que es:*



(I')

en donde:

*R<sub>1</sub> se selecciona de alquilo C<sub>1</sub>-C<sub>6</sub> sustituido o no sustituido, cicloalquilo sustituido o no sustituido, arilo sustituido o no sustituido y heterociclilo sustituido o no sustituido;*

*R<sub>3</sub> es un radical alquilo C<sub>1</sub>-C<sub>6</sub> no sustituido;*

*o una sal, un profármaco y/o un solvato farmacéuticamente aceptable del mismo.*

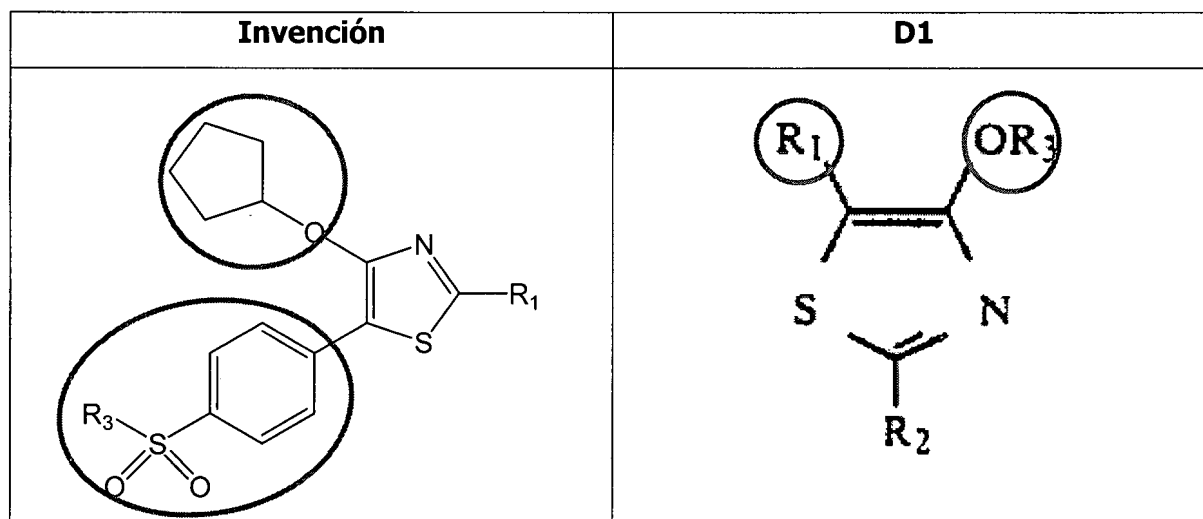
Así, el señor Examinador puede notar que las reivindicación contenidas en el nuevo pliego reivindicatorio reclaman protección para un compuesto químico de formula (I') y una composición química que lo contiene.

En este orden de ideas consideramos que el alcance de las nuevas reivindicaciones se encuentra debidamente sustentado en la totalidad de las enseñanzas de la descripción y corresponde a materia plenamente patentable, por la cual solicitamos respetuosamente aceptar el nuevo pliego reivindicatorio como claro y conciso.

### **Nivel inventivo**

Nos permitimos exponer los argumentos mediante los cuales defendemos el nivel inventivo de la presente invención a la luz de las enseñanzas de las referencias citadas a lo largo del Oficio Técnico emitido por su Despacho.

De acuerdo con la materia revelada por la nueva reivindicación 1, es posible establecer algunas diferencias estructurales entre las formulas reveladas tanto por la presente invención, como por la referencia **D1 (US5124342)**. Así, nos permitimos exponer las diferencias mencionadas:



En este orden de ideas, tal como lo reconoce el señor Examinador, las diferencias entre la presente invención y D1 consisten en:

- (i) El sustituyente sobre el átomo de oxígeno en la posición 4 del anillo de tiazol corresponde a ciclopentil, mientras que en contraste el radical equivalente  $R^3$  en D1 se selecciona de:

- Hidrógeno,
- Una sal farmacéuticamente aceptable,
- $CO R_4$ ,
- $CO CX_1 X_2 NR_6 R_7$ ,
- $CR_8 R_9 OR_{10}$ ,
- $CH_2 CR_8 (OR_{10}) CH_2 OR_{11}$ ; y
- $Si R_{12} R_{13} R_{14}$ .

De esta manera, D1 establece claramente que cuando el radical  $R^3$  corresponde a un grupo alquilo, dicho grupo alquilo debe ser necesariamente acompañado por un grupo funcional oxigenado, es decir, hidroxilo o alcoxilo.

- (ii) La fórmula (I') de la presente invención contempla específicamente la presencia de un grupo 4-alquilsulfonilfenilo en la posición 5 del anillo de tiazol. Por su parte, D1 especifica que el correspondiente radical  $R^1$  se selecciona de fenilo o fenilo sustituido con halógeno, alquilo  $C_{1-12}$ , ciano, nitro,  $COR^4$ ,  $SO_2R^4$ ,  $NR^5R^6$  y  $OR^6$ , en donde  $R^4$  es alquilo  $C_{1-12}$ , y  $R^5$  y  $R^6$  son hidrogeno o alquilo  $C_{1-12}$ .

En este orden de ideas, el señor Examinador consideró lo siguiente,

*"Sin embargo, el documento D1 sugiere la inclusión en la posición 5 del anillo de tiazol de un radical arilo sustituido con un grupo  $SO_2R^4$  siendo  $R^4$  un grupo alquilo y la inclusión en la posición 4 de un grupo  $OR^3$  donde  $R^3$  es  $CR^8R^9OR^{10}$  y al menos 2 grupos de  $R^8$ ,  $R^9$ ,  $R^{10}$  y  $R^{11}$  forman juntos un anillo conteniendo 5 a 10 átomos dando lugar a un anillo heterocíclico de 5 a 10 átomos comprendiendo un átomo de oxígeno o un anillo carbocíclico de 5 a 10 átomos sustituido por un grupo  $OR^{10}$  para obtener un compuesto inhibidor de 5-lipooxigenasa del cual es conocida su acción inmunomoduladora y su utilidad en el tratamiento de estados inflamatorios agudos y crónicos.*

Al respecto, nos permitimos hacer claridad en los siguientes aspectos,

- D1 contempla una amplia lista de opciones para el radical  $R^1$  (posición 5 del anillo de tiazol), sin hacer mención particular de ninguno de dichos sustituyentes. Por tanto, aunque la sustitución contemplada por la presente invención (4-alquilsulfonilfenilo) se encuentre incluida de manera general dentro de las opciones listadas para  $R^1$ , dicha referencia no contempla ningún compuesto que presente dicha sustitución específica, y por tanto tampoco describe la actividad de compuesto sustituidos con 4-alquilsulfonilfenilo.

De acuerdo a lo anterior, el normalmente versado en la materia no encuentra ninguna motivación para decidir realizar la sustitución específica contemplada por la presente invención, pues se enfrenta a una amplia gama de posibilidades dirigidas a la sustitución del mismo radical.

- D1 enseña que en el evento de que los grupos  $R^8$ ,  $R^9$ ,  $R^{10}$  y  $R^{11}$  formen un anillo de 5 a 10 miembros, dicho anillo necesariamente va a contener un grupo oxigenado, bien dentro del anillo o como sustituyente del mismo, es decir, dicho anillo no puede corresponder exactamente a un anillo de ciclopentano como en el compuesto de la presente invención.

De hecho, nos permitimos adjuntar al presente memorial el documento *Journal of Medicinal Chemistry* 1991, 34, 2158-2165, el cual corresponde a un artículo publicado por los inventores relacionados con la referencia D1, en donde, se describe la actividad antiinflamatoria de diversos compuestos contemplados por D1.

Dicho documento contempla en la tabla II un listado de compuestos numerados, que se diferencian en el patrón de sustitución, a los cuales se les asocia un valor de  $IC_{50}$  *in vitro* relacionados con la inhibición de la 5-lipooxigenasa. Dentro de dichos compuestos se resalta el compuesto número 44, el cual comprende como radical  $R^3$ , es decir, como sustituyente sobre el átomo de oxígeno, un grupo alquilo no sustituido ( $-CH_3$ ), estableciéndose que tal compuesto presenta una inhibición no significativa (NS) de 5-lipooxigenasa a concentraciones del orden  $\mu M$ . Así, en la página 2 dicho artículo establece que reemplazar la función 4-hidroxi por metoxi derivó en la obtención de **análogos inactivos** para la inhibición de 5-lipooxigenasa.

Por tanto, el normalmente versado en la materia basado en las enseñanzas de D1 comprende que el sustituyente del radical  $R^3$  necesariamente debe comprender un grupo oxigenado, ya sea lineal, como parte de un heterociclo o como sustituyente de un anillo alquilo, pues la presencia de dicho átomo de oxígeno repercute favorablemente en la actividad inhibidora de 5-lipooxigenasa.

En este orden de ideas, la siguiente afirmación del señor Examinador queda completamente desvirtuada,

*"En consecuencia, la persona del oficio normalmente versado en la materia, esperaría que al sustituir el anillo de tiazol divulgado en el documento D1 por un sustituyente 4-metilsulfonilfenil en la posición 5 y un radical cicloalquilo (tal como lo sugiere el documento D1 al contemplar el radical  $R^3$  como un anillo carbocíclico de 5 a 10 átomos) insustituido; se mantenga el efecto del compuesto en el*

*tratamiento de enfermedades inflamatorias como la artritis reumatoide, para llegar al objeto de la invención 12, por lo cual se considera obvia”*

Toda vez que se ha demostrado que la referencia D1 no brinda ninguna sugerencia al normalmente versado en la materia para realizar la sustitución específica del radical R<sup>1</sup> con 4-metilsulfonilfenil, y que explícitamente los inventores de D1 reconocen que la sustitución del radical R<sup>3</sup> debe comprender un grupo oxigenado para así conservar la actividad inhibidora de 5-lipooxigenasa.

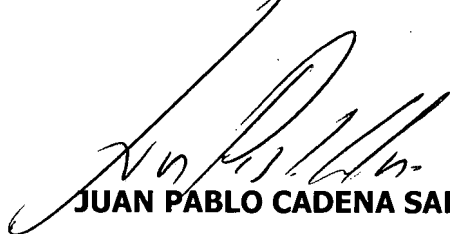
De esta manera, el normalmente versado en la materia enfrentado a la necesidad de obtener nuevos compuestos anti-inflamatorios, basándose en las enseñanzas de D1 no optaría por el desarrollo de un compuesto que comprende un sustituyente 4-metilsulfonilfenilo, pues existen múltiples opciones para tal sustitución, y aun si decidiera, por un motivo desconocido, incluir específicamente el sustituyente en mención, dicho versado no optaría por incluir un anillo de ciclopentilo como sustituyente sobre el átomo de oxígeno en posición 4 del anillo de tiazol pues sabría qué tal práctica derivaría en la obtención de un compuesto sin actividad anti-inflamatoria.

Lo anterior nos permite concluir que en el análisis llevado a cabo por el señor Examinador se determinó la obviedad de la invención basándose en un análisis en retrospectiva pues en ningún momento se demostró correctamente como las referencias citadas sugerían al versado en la materia los patrones de sustitución específicos reivindicados, más aun teniendo en cuenta que D1 aleja al versado en la materia de incluir un alquilo no sustituido como parte del radical R<sup>3</sup>, lo cual no es correcto pues para considerar obvia una invención las enseñanzas del estado de la técnica deben sugerir al normalmente versado en la materia la forma de llegar a la invención sin emplear actividad inventiva.

Por tanto, los compuestos de la presente invención contemplan una marcada diferencia estructural respecto a los compuestos de D1 la cual no es obvia a partir de las enseñanzas de dicha referencia, de manera tal que el normalmente versado en la materia no se encontraría en capacidad de llegar a la presente invención con una expectativa real de éxito. Así, la presente invención es inventiva a la luz del arte previo.

En vista de lo anteriormente expuesto, respetuosamente solicitamos el retiro de las objeciones de la presente solicitud y la concesión del privilegio solicitado, ya que en opinión del solicitante cumple con todos los requisitos establecidos en la decisión 486.

Del Señor Director



**JUAN PABLO CADENA SARMIENTO**

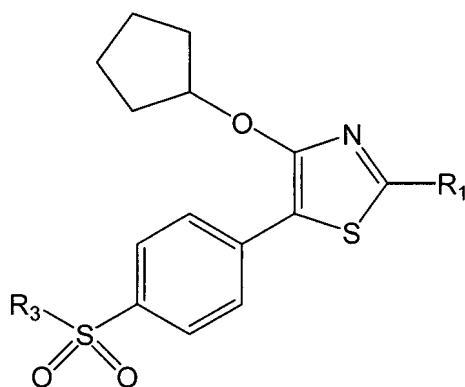
T.P. No. 57.492 del C.S.J.

C.C. 80.410.337 de Usaquén

Calle 70 A No. 4 - 41

**REIVINDICACIONES**

1. Un compuesto de fórmula (I') que es:



(I')

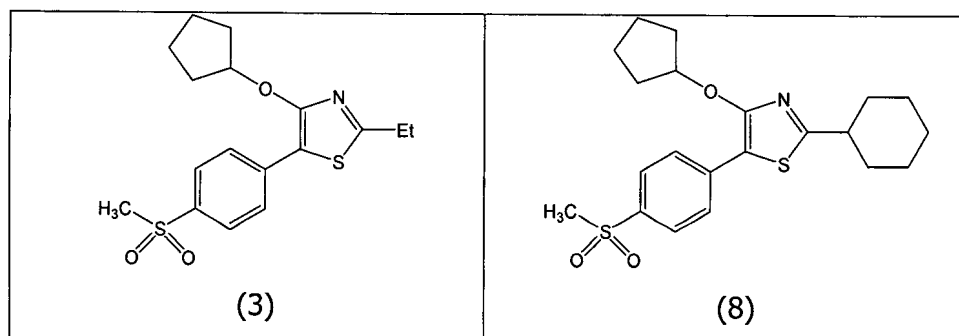
en donde

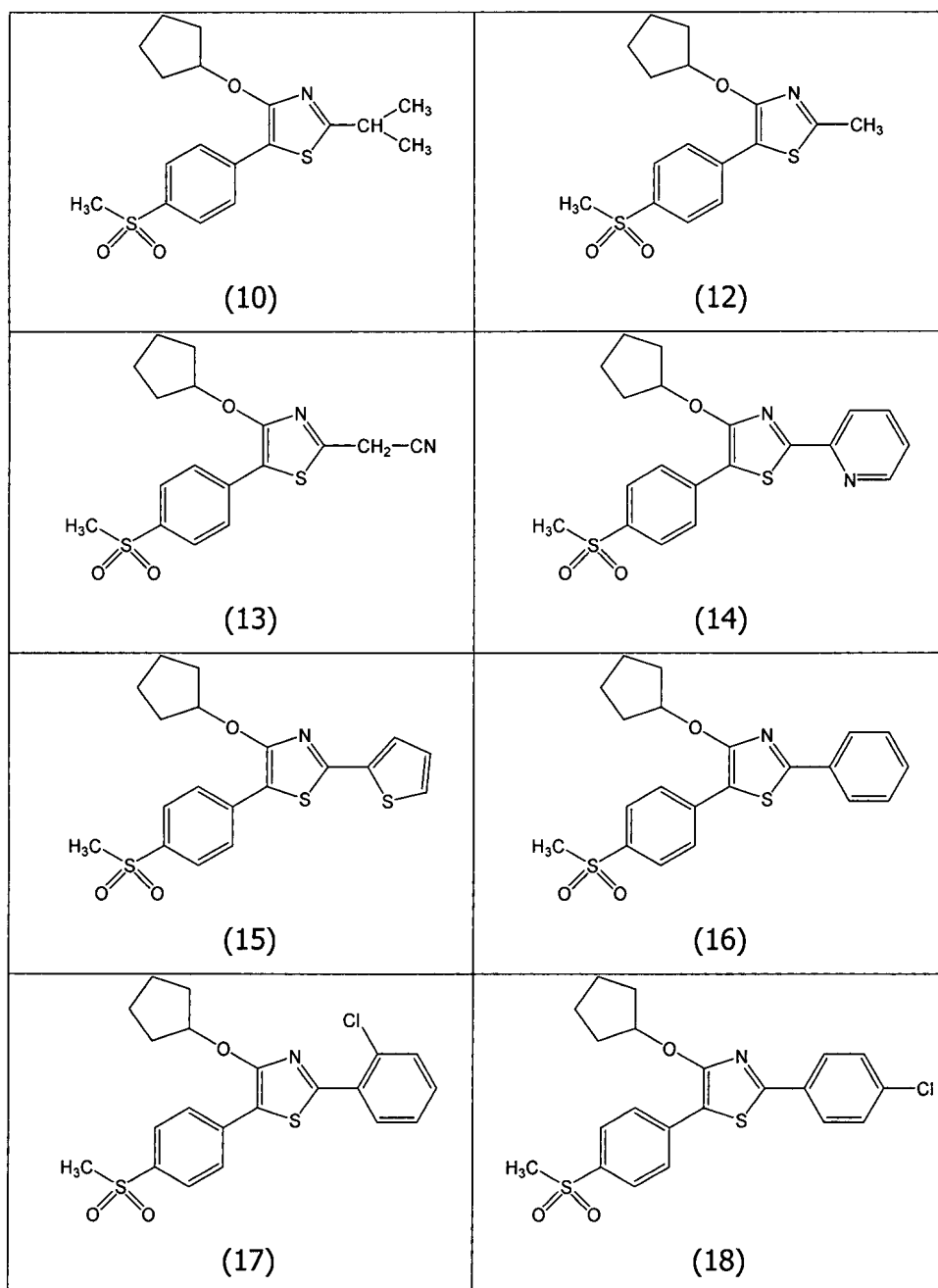
$R_1$  se selecciona de alquilo  $C_1$ - $C_6$  sustituido o no sustituido, cicloalquilo sustituido o no sustituido, arilo sustituido o no sustituido y heterociclilo sustituido o no sustituido;

$R_3$  es un radical alquilo  $C_1$ - $C_6$  no sustituido;

o una sal, un profármaco y/o un solvato farmacéuticamente aceptable del mismo.

2. El compuesto según la reivindicación 2, en el que  $R_1$  es un alquilo  $C_1$ - $C_6$  sustituido o no sustituido.
3. El compuesto según la reivindicación 3, en el que  $R_1$  es metilo.
4. El compuesto según la reivindicación 1, que se selecciona de lo siguiente:





o una sal, un profármaco y/o un solvato farmacéuticamente aceptable del mismo.

5. Una composición farmacéutica que comprende un compuesto de fórmula (I') según cualquiera de las reivindicaciones 1 a 4, o una sal, un profármaco o un solvato farmacéuticamente aceptable del mismo, y un portador, adyuvante y/o vehículo farmacéuticamente aceptable.

Research Article

4-Hydroxythiazole inhibitors of 5-lipoxygenase

Francis A. J. Kerdesky, James H. Holms, Jimmie L. Moore, Randy L. Bell, Richard D. Dyer, George W. Carter, Dee W. Brooks

J. Med. Chem., 1991, 34 (7), pp 2158-2165  
DOI: 10.1021/jm00111a035  
Publication Date: July 1991

ACS Legacy Archives

Note: In lieu of an abstract, this is the article's first page.

Click to increase image size

J. Med. Chem. 1991, 34, 2158-2165

**4-Hydroxythiazole Inhibitors of 5-Lipoxygenase**

Francis A. J. Kerdesky,\* James H. Holms, Jimmie L. Moore, Randy L. Bell, Richard D. Dyer, George W. Carter, and Dee W. Brooks<sup>†</sup>

Immunoscience Research Area, Abbott Laboratories, Abbott Park, Illinois 60064. Received October 23, 1991

4-Hydroxythiazoles have been identified as potent inhibitors of 5-lipoxygenase in vitro with  $IC_{50}$ 's of less than 1  $\mu$ M. An investigation of structure-activity relationships showed that the most potent inhibitors of this series are the 5-phenyl derivatives. The corresponding thiazole-4-one analogues were found to be relatively inactive. The 4-hydroxythiazoles were active inhibitors against 5-lipoxygenase in both intact rat polymorphonuclear leukocytes and human whole blood. The compounds were also selective inhibitors of 5-lipoxygenase, displaying only weak activity against other related enzymes, cyclooxygenase and 12- and 15-lipoxygenase.

The enzyme 5-lipoxygenase catalyzes the initial step in the metabolism of arachidonic acid leading to the leukotrienes, which have been implicated as important mediators of various human disorders.<sup>1</sup> Modulation of the activity of this enzyme provides a new therapeutic approach to treat leukotriene-mediated afflictions and should offer an important means to further elucidate the role of leukotrienes in pathophysiological events.

During investigations to develop selective inhibitors of 5-lipoxygenase,<sup>2</sup> it was discovered that 5-methyl-2-phenyl-4-hydroxythiazole (1) had potent inhibitory activity ( $IC_{50}$  = 0.96  $\mu$ M) against the RBL-1, 5-lipoxygenase enzyme (20000g supernatant), whereas the closely related 2-phenylthiazoline-4-one (ortho isomer) (2) was relatively

**Scheme 1**

Reaction of 5-methyl-2-phenyl-4-hydroxythiazole (1) with arachidonic acid to form leukotrienes.

Arachidonic acid + 1 → Leukotrienes

Arachidonic acid + 2 → Leukotrienes

- Tools
- Add to Favorites
  - Download Citation
  - Email a Colleague
  - Permalink
  - Order Reprints
  - Rights & Permissions
  - Citation Alerts

- SciFinderLinks
- SciFinder
- Get Reference Detail
  - Get Substances
  - Get Citing
  - Explore by:
    - Author of this Article
    - Any Author
    - Research Topic (Now with patent search)
  - Kerdesky, Francis A. J.
  - Search

## 4-Hydroxythiazole Inhibitors of 5-Lipoxygenase

Francis A. J. Kerdesky,\* James H. Holms, Jimmie L. Moore, Randy L. Bell, Richard D. Dyer, George W. Carter, and Dee W. Brooks\*

Immunosciences Research Area, Abbott Laboratories, Abbott Park, Illinois 60064. Received October 23, 1990

4-Hydroxythiazoles have been identified as potent inhibitors of 5-lipoxygenase in vitro exhibiting  $IC_{50}$ 's of less than  $1 \mu M$ . An investigation of structure-activity relationships showed that the most potent inhibitors of this series are the 5-phenyl derivatives. The corresponding thiazolidin-4-one analogues were found to be relatively inactive. The 4-hydroxythiazoles were active inhibitors against 5-lipoxygenase in both intact rat polymorphonuclear leukocytes and human whole blood. The compounds were also selective inhibitors of 5-lipoxygenase, displaying only weak activity against other related enzymes, cyclooxygenase and 12- and 15-lipoxygenase.

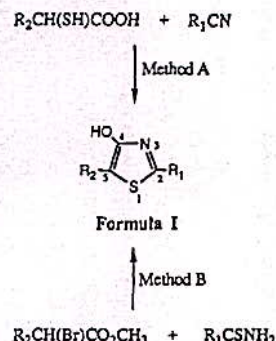
The enzyme 5-lipoxygenase catalyzes the initial step in the metabolism of arachidonic acid leading to the leukotrienes, which have been implicated as important mediators of various human disorders.<sup>1</sup> Modulation of the activity of this enzyme provides a new therapeutic approach to treat leukotriene-mediated afflictions and should offer an important means to further elucidate the role of leukotrienes in pathophysiological events.

During investigations to develop selective inhibitors of 5-lipoxygenase,<sup>2</sup> it was discovered that 5-methyl-2-phenyl-4-hydroxythiazole (1) had potent inhibitory activity ( $IC_{50} = 0.96 \mu M$ ) against the RBL-1, 5-lipoxygenase enzyme (20000g supernatant), whereas, the closely related 2-phenylthiazolidin-4-one (keto tautomer) (2) was relatively inactive (26% inhibition at  $30 \mu M$ ). Nuclear magnetic resonance (in  $DMSO-d_6$  and  $CDCl_3$ ) and infrared (KBr) spectrometric studies indicated that 1 existed entirely in the hydroxythiazole or enol form, while 2 existed as a mixture of keto and enol tautomers with the keto form predominant.<sup>3</sup> Several other related compounds were prepared and evaluated (Figure 1) to address the hypothesis that the 4-hydroxythiazole or enol form was necessary for potent inhibitory activity. Thiazolidinone 3 constrained to the keto form by  $\alpha$ -disubstitution was also inactive. Removal of the 4-hydroxy group as in thiazole 4 resulted in a compound with no inhibitory activity. Another pair of compounds 5 and 6 showed a similar phenomenon; that is, hydroxythiazole 5 was an inhibitor ( $IC_{50} = 3.9 \mu M$ ) while thiazolidinone 6 was only marginally active (8% inhibition at  $30 \mu M$ ). These initial observations led us to propose that the 4-hydroxythiazole unit was a promising lead for the development of potent 5-lipoxygenase inhibitors.

## Chemistry

The 4-hydroxythiazoles were prepared via two general synthetic routes<sup>4</sup> outlined in Scheme I. The first approach, Method A, involved the reaction of a nitrile with an  $\alpha$ -mercaptoacetic acid derivative at  $100^\circ C$  for several hours. The second approach, Method B, utilized the condensation of an  $\alpha$ -halo ester with an appropriately substituted thioamide in toluene at  $80^\circ C$  for several hours. Conventional methods of hydroxyl derivitization were used to provide analogues of the parent hydroxythiazoles.

## Scheme I



## Evaluation of Compounds

The structure-activity relationships for the 4-hydroxythiazole system were investigated at the S, C<sub>2</sub>, OH, and C<sub>5</sub> sites. The compounds were initially evaluated for inhibitory activity in a 5-lipoxygenase assay<sup>5</sup> utilizing the 20000g supernatant of sonicated homogenized rat basophilic leukemia cells.

Compounds with interesting activity in the broken cell 5-lipoxygenase assay were further evaluated against washed rat polymorphonuclear leukocytes stimulated to make LTB<sub>4</sub> by addition of the calcium ionophore A 23187 and against ionophore-stimulated human whole blood.

**Heteroatom Replacements at S.** Replacement of the sulfur atom with more electronegative atoms such as oxygen or nitrogen reduced inhibitory activity (Figure 2). For example, oxazole 8, which exists primarily in the keto tautomeric form according to NMR and IR studies, had an  $IC_{50}$  of  $8.1 \mu M$ , being about 16 times less potent than 4-hydroxythiazole 7 ( $IC_{50} = 0.53 \mu M$ ). 4-Hydroxyimidazole 9 was essentially inactive at  $32 \mu M$ . From this brief analysis the thiazole group was chosen as the template for further refinement of inhibitory activity.

**Substituents at C<sub>2</sub>.** The effect of various substituents at C<sub>2</sub> of the 4-hydroxythiazole system represented by Formula I with respect to 5-lipoxygenase inhibitory activity was evaluated (Table I). The phenyl derivative 1 was the prototype and had an  $IC_{50}$  of  $0.96 \mu M$ . Other heterocyclic groups at C<sub>2</sub> were examined and analogues such as 3-pyridyl (5), 2-thienyl (10), 2-furyl (11), 2-pyridyl (12), and 2-(6-methoxybenzothiazolyl) (14) exhibited  $IC_{50}$ 's in the  $0.89$ – $3.9 \mu M$  range. The 4-pyridyl (13), 3-quinolinyl (15), and 3-pyrazolyl (16) analogues were substantially less potent.

Compounds with polar substituents at C<sub>2</sub> as in the esters (18 and 19), carboxyl (20), amino (21), and hydrazino (22) analogues or a simple methyl derivative (17) were all inactive at  $32 \mu M$ .

- (1) Schewe, T.; Rapoport, S. M.; Kuhn, H. In *Advances in Enzymology*; Meister, E., Ed.; J. Wiley: New York, 1986; Vol. 58, pp 191-272.
- (2) Summers, J. B.; Kim, K. H.; Mazdiyasni, H.; Holms, J. H.; Ratajczyk, J. D.; Stewart, A. O.; Dyer, R. D.; Carter, G. W. *J. Med. Chem.* 1990, 33, 992.
- (3) The keto-enol equilibrium of substituted thiazoles has been studied: *Thiazole And Its Derivatives*, Vol. 34, Part 2 of *The Chemistry of Heterocyclic Compounds*; Metzger, J. V., Ed.; J. Wiley: New York, 1979; pp 419-446 and references contained therein.
- (4) *Comprehensive Heterocyclic Chemistry*; Potts, K. T. Ed.; Pergamon Press: Oxford, 1984; Vol. 6, p 235.

- (5) Dyer, R. D.; Bornemier, D. A.; Haviv, F.; Carter, G. W. *Fed. Proc., Fed. Am. Soc. Exp. Biol.* 1985, 44, 904.

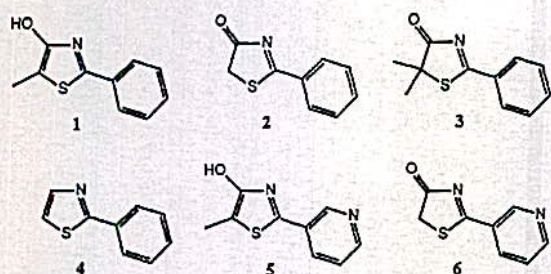


Figure 1.

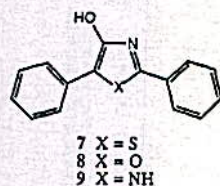


Figure 2.

**Substituted 2-Aryl Derivatives.** Since compounds with a phenyl group at C<sub>2</sub>, as in compound 1, exhibited promising inhibitory potency, further examination of substituted phenyl derivatives was conducted. No apparent correlation was found to exist between biological activity [ $\log(1/IC_{50})$ ] and electronic ( $\sigma$ ) or hydrophobic ( $\pi$ ) constants. For example, the electron-donating methyl (26) and methoxy (31) analogues with  $IC_{50}$ 's of 0.63 and 1.1  $\mu$ M had similar inhibitory activity as the electron-withdrawing nitrile (27) and nitro (33) analogues with  $IC_{50}$ 's of 0.70 and 1.6  $\mu$ M. A variety of other substituents such as halogens (23–25), esters (28–30), keto (32), sulfonamide (34), trifluoromethyl (35), and trifluoromethyl thiol (36) led to only minor differences in inhibitory activity within a 0.35–2.7  $\mu$ M range. A substantial reduction in inhibitory activity was observed in analogues with ionizable groups such as carboxamide (37), amino (38), and carboxyl (39). Incorporating another phenyl ring at the 2-aryl site as in 40 resulted in reduced activity. The position of the substituent on the phenyl ring was also studied. For example, *p*-fluorophenyl compound 23 exhibited activity similar to that of the corresponding *o*-fluoro (41) and the *m*-fluoro (42). With the exception of highly polar groups and biphenyl (40), the effect of substituents as well as their position on the phenyl ring generally did not greatly alter the inhibitory activity.

**Modification of the 4-OH Site.** The importance of the enolic form of 4-hydroxythiazoles for inhibitory activity was previously supported. To further evaluate the role of the hydroxy group the following modifications were tested (Table II). Since the initially prepared 4-hydroxythiazoles had rather limited solubility both in aqueous and organic solvents, modification of the hydroxy group was also hoped to enhance solubility. Replacement of the 4-hydroxy function with an amino group provided compound 43, which was inactive. The methoxy (44) and phosphate (45) analogues were also relatively inactive.

Acyl derivatives retained 5-lipoxygenase inhibitory activity and some acetates were more potent than the corresponding hydroxy compounds. For example, acetates 61, 62, and 64 exhibited greater potency than the corresponding 4-hydroxy derivatives 60, 39, and 63, respectively. Other esters such as succinate 47, hexanoate 48, and pivalate 49 provided potent inhibition. Carbamate derivatives 53–55 and carbonates 56–58 also showed good inhibitory activity; however, sulfonate 59 was inactive.

A structural modification where the hydroxy group was moved from C<sub>4</sub> to C<sub>5</sub> on the thiazole ring was also exam-

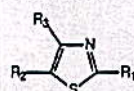
Table I. Inhibitory Activities of 4-Hydroxythiazoles of Formula I

| Formula I |                                                                                  |                                                                  |                     |                                                                                             |
|-----------|----------------------------------------------------------------------------------|------------------------------------------------------------------|---------------------|---------------------------------------------------------------------------------------------|
| no.       | R <sub>1</sub>                                                                   | R <sub>2</sub>                                                   | method <sup>a</sup> | in vitro 5-lo inhibn: IC <sub>50</sub> , $\mu$ M, <sup>b</sup> or % inhibn at $\mu$ M concn |
| 1         | C <sub>6</sub> H <sub>5</sub>                                                    | CH <sub>3</sub>                                                  | B                   | 0.96 (0.8–1.1)                                                                              |
| 5         | 3-pyridyl                                                                        | CH <sub>3</sub>                                                  | A                   | 3.9 (3.6–4.3)                                                                               |
| 7         | C <sub>6</sub> H <sub>5</sub>                                                    | C <sub>6</sub> H <sub>5</sub>                                    | B                   | 0.53 (0.52–0.55)                                                                            |
| 10        | 2-thienyl                                                                        | CH <sub>3</sub>                                                  | A                   | 1.4 (1.2–1.6)                                                                               |
| 11        | 2-furyl                                                                          | CH <sub>3</sub>                                                  | A                   | 2.4 (2.2–2.6)                                                                               |
| 12        | 2-pyridyl                                                                        | CH <sub>3</sub>                                                  | A                   | 2.1 (1.9–2.3)                                                                               |
| 13        | 4-pyridyl                                                                        | CH <sub>3</sub>                                                  | A                   | 14 (13–14)                                                                                  |
| 14        | 6-CH <sub>3</sub> O-2-benzothiazolyl                                             | CH <sub>3</sub>                                                  | A                   | 0.89 (0.71–1.1)                                                                             |
| 15        | 3-quinolyl                                                                       | CH <sub>3</sub>                                                  | A                   | 16 (14–18)                                                                                  |
| 16        | 3-pyrazolyl                                                                      | CH <sub>3</sub>                                                  | A                   | 31 (29–33)                                                                                  |
| 17        | CH <sub>3</sub>                                                                  | CH <sub>3</sub>                                                  | B                   | NS at 32 <sup>c</sup>                                                                       |
| 18        | CH <sub>2</sub> CO <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>                  | CH <sub>3</sub>                                                  | A                   | NS at 32 <sup>c</sup>                                                                       |
| 19        | CO <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>                                  | CH <sub>3</sub>                                                  | A                   | NS at 32 <sup>c</sup>                                                                       |
| 20        | COOH                                                                             | CH <sub>3</sub>                                                  | A                   | NS at 32 <sup>c</sup>                                                                       |
| 21        | NH <sub>2</sub>                                                                  | CH <sub>3</sub>                                                  | B                   | NS at 32 <sup>c</sup>                                                                       |
| 22        | NHNH <sub>2</sub>                                                                | CH <sub>3</sub>                                                  | B                   | NS at 32 <sup>c</sup>                                                                       |
| 23        | 4-F-C <sub>6</sub> H <sub>4</sub>                                                | CH <sub>3</sub>                                                  | A                   | 0.35 (0.28–0.43)                                                                            |
| 24        | 4-Br-C <sub>6</sub> H <sub>4</sub>                                               | CH <sub>3</sub>                                                  | A                   | 0.51 (0.50–0.51)                                                                            |
| 25        | 4-Cl-C <sub>6</sub> H <sub>4</sub>                                               | CH <sub>3</sub>                                                  | A                   | 0.57 (0.52–0.63)                                                                            |
| 26        | 4-CH <sub>3</sub> -C <sub>6</sub> H <sub>4</sub>                                 | CH <sub>3</sub>                                                  | B                   | 0.63 (0.62–0.64)                                                                            |
| 27        | 4-CN-C <sub>6</sub> H <sub>4</sub>                                               | CH <sub>3</sub>                                                  | A                   | 0.70 (0.58–0.83)                                                                            |
| 28        | 4-CO <sub>2</sub> C <sub>2</sub> H <sub>5</sub> -C <sub>6</sub> H <sub>4</sub>   | CH <sub>3</sub>                                                  | A                   | 0.71 (0.60–0.85)                                                                            |
| 29        | 4-CO <sub>2</sub> CH <sub>2</sub> -C <sub>6</sub> H <sub>4</sub>                 | CH <sub>3</sub>                                                  | A                   | 0.88 (0.65–1.1)                                                                             |
| 30        | 4-CO <sub>2</sub> CH <sub>2</sub> CH <sub>2</sub> -C <sub>6</sub> H <sub>4</sub> | CH <sub>3</sub>                                                  | A                   | 0.98 (0.78–1.30)                                                                            |
| 31        | 4-OCH <sub>3</sub> -C <sub>6</sub> H <sub>4</sub>                                | CH <sub>3</sub>                                                  | B                   | 1.10 (0.97–1.20)                                                                            |
| 32        | 4-COCH <sub>3</sub> -C <sub>6</sub> H <sub>4</sub>                               | CH <sub>3</sub>                                                  | A                   | 1.40 (1.2–1.6)                                                                              |
| 33        | 4-NO <sub>2</sub> -C <sub>6</sub> H <sub>4</sub>                                 | CH <sub>3</sub>                                                  | A                   | 1.6 (1.4–1.8)                                                                               |
| 34        | 4-SO <sub>2</sub> NH <sub>2</sub> -C <sub>6</sub> H <sub>4</sub>                 | CH <sub>3</sub>                                                  | A                   | 2.2 (2.0–2.4)                                                                               |
| 35        | 4-CF <sub>3</sub> -C <sub>6</sub> H <sub>4</sub>                                 | CH <sub>3</sub>                                                  | A                   | 2.5 (2.2–2.7)                                                                               |
| 36        | 4-SCF <sub>3</sub> -C <sub>6</sub> H <sub>4</sub>                                | CH <sub>3</sub>                                                  | A                   | 2.7 (2.5–2.9)                                                                               |
| 37        | 4-CONH <sub>2</sub> -C <sub>6</sub> H <sub>4</sub>                               | CH <sub>3</sub>                                                  | A                   | 4.8 (3.9–6.1)                                                                               |
| 38        | 4-NH <sub>2</sub> -C <sub>6</sub> H <sub>4</sub>                                 | CH <sub>3</sub>                                                  | A                   | 15 (13–17)                                                                                  |
| 39        | 4-COOH-C <sub>6</sub> H <sub>4</sub>                                             | CH <sub>3</sub>                                                  | A                   | 30 (28–32)                                                                                  |
| 40        | 4-C <sub>6</sub> H <sub>5</sub> -C <sub>6</sub> H <sub>4</sub>                   | CH <sub>3</sub>                                                  | A                   | 40% at 30                                                                                   |
| 41        | 2-F-C <sub>6</sub> H <sub>4</sub>                                                | CH <sub>3</sub>                                                  | A                   | 0.50 (0.48–0.52)                                                                            |
| 42        | 3-F-C <sub>6</sub> H <sub>4</sub>                                                | CH <sub>3</sub>                                                  | A                   | 0.66 (0.56–0.76)                                                                            |
| 51        | C <sub>6</sub> H <sub>5</sub>                                                    | CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>                  | B                   | 0.58 (0.55–0.62)                                                                            |
| 60        | C <sub>6</sub> H <sub>5</sub>                                                    | CH <sub>2</sub> CH <sub>2</sub> -CH <sub>2</sub> CH <sub>3</sub> | B                   | 18 (16–22)                                                                                  |
| 63        | 4-C <sub>6</sub> H <sub>5</sub> -C <sub>6</sub> H <sub>4</sub>                   | C <sub>6</sub> H <sub>5</sub>                                    | B                   | 2.2 (2.0–2.4)                                                                               |
| 65        | C <sub>6</sub> H <sub>5</sub>                                                    | CH <sub>2</sub> CH <sub>2</sub> -CH <sub>2</sub> CH <sub>3</sub> | B                   | 0.83 (0.73–0.92)                                                                            |
| 66        | C <sub>6</sub> H <sub>5</sub>                                                    | CH <sub>2</sub> CH <sub>2</sub> -C <sub>6</sub> H <sub>5</sub>   | B                   | 0.69 (0.62–0.78)                                                                            |
| 67        | C <sub>6</sub> H <sub>5</sub>                                                    | CH <sub>2</sub> COOH                                             | B                   | 24% at 30                                                                                   |
| 68        | C <sub>6</sub> H <sub>5</sub>                                                    | CH <sub>2</sub> CO <sub>2</sub> CH <sub>3</sub>                  | B                   | 3.5 (2.7–4.6)                                                                               |
| 69        | C <sub>6</sub> H <sub>5</sub>                                                    | CH <sub>2</sub> CON-(OH)CH <sub>3</sub>                          | B                   | 4.4 (3.9–5.0)                                                                               |
| 70        | C <sub>6</sub> H <sub>5</sub>                                                    | CO <sub>2</sub> C <sub>2</sub> H <sub>5</sub>                    | B                   | 17% at 30                                                                                   |
| 72        | CH <sub>3</sub>                                                                  | C <sub>6</sub> H <sub>5</sub>                                    | B                   | 0.7 (0.5–0.92)                                                                              |
| 73        | 4-F-C <sub>6</sub> H <sub>4</sub>                                                | C <sub>6</sub> H <sub>5</sub>                                    | B                   | 0.5 (0.3–0.7)                                                                               |
| 74        | 4-CH <sub>3</sub> -C <sub>6</sub> H <sub>4</sub>                                 | C <sub>6</sub> H <sub>5</sub>                                    | B                   | 0.8 (0.6–1.3)                                                                               |
| 75        | 4-COOH-C <sub>6</sub> H <sub>4</sub>                                             | C <sub>6</sub> H <sub>5</sub>                                    | A                   | 1.2 (0.92–1.5)                                                                              |
| 76        | 4-CH <sub>3</sub> O-C <sub>6</sub> H <sub>4</sub>                                | C <sub>6</sub> H <sub>5</sub>                                    | B                   | 0.37 (0.38–0.38)                                                                            |
| 77        | 4-pyridyl                                                                        | C <sub>6</sub> H <sub>5</sub>                                    | B                   | 0.54 (0.46–0.61)                                                                            |
| 77        | 3-pyridyl                                                                        | C <sub>6</sub> H <sub>5</sub>                                    | B                   | 1.1 (0.8–1.4)                                                                               |

<sup>a</sup> See Experimental Section for methods. <sup>b</sup>  $IC_{50}$  with 95% confidence limits in parentheses or mean percent inhibition values for the in vitro inhibition of 5-lipoxygenase (5-lo) from the 20000g supernatant of RBL-1 cells. <sup>c</sup> Nonsignificant (NS) inhibition at concentration in  $\mu$ M.

ined. As illustrated by 5-hydroxy analogue 50 compared to 4-hydroxy analogue 7, this modification resulted in a

Table II. Inhibitory Activities of 4-Substituted Thiazoles of Formula II



Formula II

| no. | R <sub>1</sub>                                                 | R <sub>2</sub>                                                  | R <sub>3</sub>                                                                 | method <sup>a</sup> | in vitro 5-lo inhibn:<br>IC <sub>50</sub> , μM, <sup>b</sup> or<br>% inhibn at μM concn |
|-----|----------------------------------------------------------------|-----------------------------------------------------------------|--------------------------------------------------------------------------------|---------------------|-----------------------------------------------------------------------------------------|
| 7   | C <sub>6</sub> H <sub>5</sub>                                  | C <sub>6</sub> H <sub>5</sub>                                   | OH                                                                             | B                   | 0.53 (0.52–0.55)                                                                        |
| 43  | C <sub>6</sub> H <sub>5</sub>                                  | C <sub>6</sub> H <sub>5</sub>                                   | NH <sub>2</sub>                                                                |                     | NS at 32 <sup>c</sup>                                                                   |
| 44  | C <sub>6</sub> H <sub>5</sub>                                  | C <sub>6</sub> H <sub>5</sub>                                   | OCH <sub>3</sub>                                                               |                     | NS at 32 <sup>c</sup>                                                                   |
| 45  | C <sub>6</sub> H <sub>5</sub>                                  | C <sub>6</sub> H <sub>5</sub>                                   | (CH <sub>3</sub> O) <sub>2</sub> PO <sub>2</sub>                               |                     | 14% at 32                                                                               |
| 46  | C <sub>6</sub> H <sub>5</sub>                                  | C <sub>6</sub> H <sub>5</sub>                                   | OCOCH <sub>3</sub>                                                             | C                   | 0.4 (0.3–0.54)                                                                          |
| 47  | C <sub>6</sub> H <sub>5</sub>                                  | C <sub>6</sub> H <sub>5</sub>                                   | OCOC <sub>2</sub> H <sub>4</sub> CO <sub>2</sub> C <sub>2</sub> H <sub>5</sub> | D                   | 0.75 (0.69–0.80)                                                                        |
| 48  | C <sub>6</sub> H <sub>5</sub>                                  | C <sub>6</sub> H <sub>5</sub>                                   | OCOC <sub>2</sub> H <sub>11</sub>                                              | D                   | 1.3 (1.2–1.4)                                                                           |
| 49  | C <sub>6</sub> H <sub>5</sub>                                  | C <sub>6</sub> H <sub>5</sub>                                   | OCOC <sub>2</sub> H <sub>9</sub>                                               | D                   | 9.1 (8.5–1.0)                                                                           |
| 50  | C <sub>6</sub> H <sub>5</sub>                                  | OH                                                              | C <sub>6</sub> H <sub>5</sub>                                                  |                     | 6.9 (6.0–7.9)                                                                           |
| 51  | C <sub>6</sub> H <sub>5</sub>                                  | CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>                 | OH                                                                             | B                   | 0.58 (0.55–0.62)                                                                        |
| 52  | C <sub>6</sub> H <sub>5</sub>                                  | CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>                 | OCOCH <sub>3</sub>                                                             | C                   | 0.53 (0.48–0.59)                                                                        |
| 53  | C <sub>6</sub> H <sub>5</sub>                                  | CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>                 | OCONHCH <sub>3</sub>                                                           | E                   | 8.8 (7.4–10.3)                                                                          |
| 54  | C <sub>6</sub> H <sub>5</sub>                                  | CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>                 | OCONHC <sub>4</sub> H <sub>9</sub>                                             | E                   | 24 (22–26)                                                                              |
| 55  | C <sub>6</sub> H <sub>5</sub>                                  | CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>                 | OCONHC <sub>6</sub> H <sub>5</sub>                                             | E                   | 0.46 (0.42–0.51)                                                                        |
| 56  | C <sub>6</sub> H <sub>5</sub>                                  | CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>                 | OCO <sub>2</sub> C <sub>2</sub> H <sub>5</sub>                                 | F                   | 0.91 (0.83–0.99)                                                                        |
| 57  | C <sub>6</sub> H <sub>5</sub>                                  | CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>                 | OCO <sub>2</sub> CH <sub>2</sub> C <sub>6</sub> H <sub>5</sub>                 | F                   | 2.8 (2.6–3.1)                                                                           |
| 58  | C <sub>6</sub> H <sub>5</sub>                                  | CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>                 | OCO <sub>2</sub> C <sub>3</sub> H <sub>7</sub>                                 | F                   | 3.3 (2.5–4.1)                                                                           |
| 59  | C <sub>6</sub> H <sub>5</sub>                                  | CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>                 | OSO <sub>2</sub> CH <sub>3</sub>                                               |                     | NS at 32 <sup>c</sup>                                                                   |
| 61  | C <sub>6</sub> H <sub>5</sub>                                  | CH <sub>2</sub> CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub> | OCOCH <sub>3</sub>                                                             | C                   | 0.95 (0.81–1.1)                                                                         |
| 62  | 4-COOH-C <sub>6</sub> H <sub>4</sub>                           | CH <sub>3</sub>                                                 | OCOCH <sub>3</sub>                                                             | C                   | 14 (12–16)                                                                              |
| 64  | 4-C <sub>6</sub> H <sub>5</sub> -C <sub>6</sub> H <sub>4</sub> | C <sub>6</sub> H <sub>5</sub>                                   | OCOCH <sub>3</sub>                                                             | C                   | 1.4 (0.8–2.7)                                                                           |

<sup>a</sup> See Experimental Section for methods. <sup>b</sup> IC<sub>50</sub> with 95% confidence limits in parentheses or mean percent inhibition values for the in vitro inhibition of 5-lipoxygenase (5-lo) from the 20000g supernatant of RBL-1 cells. <sup>c</sup> Nonsignificant (NS) inhibition at concentration in μM.

substantial loss in inhibitory potency.

Among the variations evaluated at the 4-OH site, the acetyl derivatives provided the best activity.<sup>6</sup>

**Substitution at C<sub>5</sub>.** A similar pattern of substituent effects as observed for groups at C<sub>2</sub> was found for substitution at C<sub>5</sub>. With respect to hydrocarbon chain length, propyl (51) was found to be superior to the methyl (1), ethyl (65), and butyl (60). Propyl (51) gave essentially the same activity as the more chemically stable phenyl (7). Attaching polar substituents to a hydrocarbon chain as in carboxy (67), carbomethoxy (68), and hydroxamyl (69) analogues resulted in substantial decreases in inhibitory activity. Activity, however, could be maintained by attaching a phenyl group to the hydrocarbon chain as in phenethyl analogue 66. Direct attachment of an ester function as C<sub>6</sub>, for example, compound 70, was particularly detrimental to inhibitory potency. Substitution at C<sub>5</sub> with phenyl derivatives generally provided more potent inhibitors than their corresponding methyl analogues. In some cases, the difference in activity was quite dramatic. For instance, 2,5-dimethyl compound 17, which was inactive (5% inhibition at 32 μM), was transformed into a potent inhibitor as 2-methyl-5-phenyl analogue 71 (IC<sub>50</sub> = 0.7 μM). A similar phenomenon was observed for biphenyl compound 40 (40% inhibition at 30 μM); namely, the corresponding 5-phenyl analogue 63 provided potent activity (IC<sub>50</sub> = 2.2 μM). 5-Phenyl compounds 75 and 77 were more potent inhibitors than the corresponding 5-methyl analogues 31 and 5, respectively. 5-Phenyl acid 74 and pyridine 76 were ca. 30 times more active than their 5-methyl counterparts, compounds 39 and 13. In this assay, further substitution on the 5-phenyl ring (comparing compounds

Table III. Inhibitory Activities of 4-Hydroxythiazoles in Intact Rat Polymorphonuclear Leukocytes

| no. | 5-LO IC <sub>50</sub> , <sup>a</sup> μM |                  |
|-----|-----------------------------------------|------------------|
|     | RBL-1                                   | RPMNL            |
| 26  | 0.63 (0.62–0.64)                        | 1.2 (1.1–1.3)    |
| 48  | 1.3 (1.2–1.4)                           | 0.5 (0.4–0.6)    |
| 52  | 0.53 (0.48–0.59)                        | 0.66 (0.53–0.82) |
| 53  | 8.8 (7.4–10.3)                          | 0.4 (0.2–0.6)    |
| 54  | 24 (22–26)                              | 0.80 (0.76–0.85) |
| 55  | 0.46 (0.42–0.50)                        | 1.0 (0.9–1.1)    |
| 56  | 0.91 (0.83–0.99)                        | 0.7 (0.6–1.1)    |
| 57  | 2.8 (2.6–3.1)                           | 1.2 (1.2–1.3)    |
| 61  | 0.95 (0.81–1.1)                         | 0.5 (0.5–0.6)    |
| 64  | 1.4 (0.8–2.7)                           | 0.9 (0.8–1.0)    |
| 68  | 3.5 (2.7–4.6)                           | 2.1 (1.6–2.9)    |
| 75  | 0.37 (0.36–0.38)                        | 0.45 (0.35–0.55) |

<sup>a</sup> IC<sub>50</sub> with 95% confidence limits in parentheses for the inhibition of 5-lipoxygenase.

72, 73, 74, or 75 with compound 7) gave no substantial increase or decrease in activity. As a subgroup, the 5-phenyl derivatives were the most potent inhibitors with IC<sub>50</sub>'s of 0.37–1.2 μM. Therefore 5-phenyl derivatives were considered as the preferred substitution at C<sub>5</sub> to provide potent 5-lipoxygenase inhibitory activity.

#### Reversibility of 5-Lipoxygenase Inhibition

The reversibility of the inhibition of the 5-lipoxygenase enzyme by the hydroxythiazoles was examined by incubating the enzyme with compound 75 followed by diluting the enzyme before the addition of substrate. A 6-fold dilution of a 0.3 μM initial mixture gave only 7% inhibition compared to 95% by the undiluted enzyme.

#### Activity in Other Enzyme Systems

Not only were the 4-hydroxythiazoles potent inhibitors in the broken-cell preparations but they were also active against 5-lipoxygenase in intact rat polymorphonuclear

(6) All of the hydroxyl protected analogues were stable to the conditions (pH, medium) of the assay; however, hydrolysis of the compounds may occur due to the complex nature of the contaminating proteins in the assay.

**Table IV.** Inhibitory Activities of 4-Hydroxythiazoles in Human Whole Blood

| no. | 5-LO IC <sub>50</sub> , <sup>a</sup> $\mu$ M |                  |
|-----|----------------------------------------------|------------------|
|     | RBL-1                                        | HWBL             |
| 46  | 0.4 (0.3–0.59)                               | 0.45 (0.38–0.55) |
| 53  | 8.8 (7.4–10.3)                               | 5.1 (4.4–6.0)    |
| 54  | 24 (22–26)                                   | 5.5 (4.9–6.2)    |
| 56  | 0.91 (0.83–0.99)                             | 4.5 (3.1–6.1)    |
| 58  | 1.4 (0.8–2.7)                                | 2.2 (2.0–2.5)    |
| 71  | 0.7 (0.5–0.92)                               | 5.1 (4.6–5.8)    |
| 72  | 0.5 (0.3–0.96)                               | 0.47 (0.43–0.50) |
| 73  | 0.8 (0.6–1.3)                                | 0.74 (0.65–0.85) |
| 75  | 0.37 (0.36–0.38)                             | 2.4 (1.9–2.9)    |
| 77  | 1.1 (0.8–1.4)                                | 2.9 (2.7–3.1)    |

<sup>a</sup>IC<sub>50</sub> with 95% confidence limits in parentheses for the inhibition of 5-lipoxygenase.

**Table V.** 4-Hydroxythiazoles and Inhibition of Related Enzymes

| no. | 5-lipoxy-<br>genase <sup>a</sup><br>IC <sub>50</sub> , $\mu$ M | % inhibn at $\mu$ M              |                                   |                                   |
|-----|----------------------------------------------------------------|----------------------------------|-----------------------------------|-----------------------------------|
|     |                                                                | cyclo-<br>oxygenase <sup>b</sup> | 15-lipoxy-<br>genase <sup>c</sup> | 12-lipoxy-<br>genase <sup>d</sup> |
| 1   | 0.96                                                           | 32 at 30                         | 44 at 10                          | 51 at 100                         |
| 7   | 0.53                                                           | 47 at 30                         | 50 at 100                         | 12 at 30                          |
| 29  | 0.88                                                           | 6 at 30                          | 52 at 10                          | 0 at 100                          |
| 66  | 0.69                                                           | 11 at 100                        | 34 at 100                         | 3 at 100                          |
| 72  | 0.50                                                           | 20 at 100                        | 16 at 100                         | 0 at 100                          |
| 75  | 0.37                                                           | 25 at 100                        | 22 at 100                         | 6 at 100                          |

<sup>a</sup>IC<sub>50</sub> for the in vitro inhibition of 5-lipoxygenase from the 20000g supernatant of RBL-1 cells. <sup>b</sup>Mean percent inhibition values  $\pm$  2% SEM for the in vitro inhibition of sheep seminal vesicle cyclooxygenase. <sup>c</sup>Mean percent inhibition values  $\pm$  2% SEM for the in vitro inhibition of soybean 15-lipoxygenase. <sup>d</sup>Mean percent inhibition values  $\pm$  2% SEM for the in vitro inhibition of platelet 12-lipoxygenase.

leukocytes and human whole blood. In the intact rat polymorphonuclear leukocyte assay (Table III), a striking difference in potency was observed for compounds 53 and 54. Their activity in the intact cell was more than 20 times that found in the broken cell, exhibiting IC<sub>50</sub>'s of 0.4 and 0.8  $\mu$ M, respectively. Regarding the human whole blood (Table IV), 4-hydroxythiazoles 46 and 72 were particularly active in this assay with IC<sub>50</sub>'s of 0.45 and 0.47  $\mu$ M, respectively. The 4-hydroxythiazoles were generally several fold less potent in the human whole blood compared to the activity obtained in the intact rat polymorphonuclear leukocyte assay. This may be the result of the compounds binding to plasma protein in the human whole blood assay. With respect to other lipoxygenases, the 4-hydroxythiazoles studied were found to be only weakly effective against sheep seminal vesicle cyclooxygenase, soybean 15-lipoxygenase, and platelet 12-lipoxygenase (Table V).

#### Activity Relative to Other Common Lipoxygenase Inhibitors

A comparison of the activity of standard inhibitors with a representative 4-hydroxythiazole assayed under the same conditions is shown in Table VI. For example, 4-hydroxythiazole 75 has a potency similar to that of the reference inhibitors quercetin,<sup>7</sup> NDGA,<sup>8</sup> and A-64077,<sup>9</sup> but

**Table VI.** Comparison of 5-Lipoxygenase Inhibitory Activity with Common Reference Inhibitors

| compound  | IC <sub>50</sub> , <sup>a</sup> $\mu$ M | compound                        | IC <sub>50</sub> , <sup>a</sup> $\mu$ M |
|-----------|-----------------------------------------|---------------------------------|-----------------------------------------|
| 75        | 0.37 (0.36–0.38)                        | arachidonyl-<br>hydroxamic acid | 2.2 (2.0–2.4)                           |
| quercetin | 0.3 (0.27–0.31)                         | 15-HETE                         | 7.3 (6.8–7.8)                           |
| NDGA      | 0.4 (0.37–0.43)                         | Rev-5901                        | >45                                     |
| A-64077   | 0.6 (0.5–0.6)                           | 5,6-DHA                         | 55 (46–64)                              |
| BW-755C   | 1.3 (1.2–1.5)                           |                                 |                                         |

<sup>a</sup>IC<sub>50</sub> with 95% confidence limits in parentheses for the in vitro inhibition of 5-lipoxygenase from the 20000g supernatant of RBL-1 cells.

is more active than BW-755C,<sup>10</sup> arachidonohydroxamic acid, 15-HETE,<sup>11</sup> Rev-5901,<sup>12</sup> and 5,6-DHA.<sup>13</sup>

#### Conclusion

Substituted 4-hydroxythiazoles have been identified as a novel series of 5-lipoxygenase inhibitors. Evaluation of a structure-activity relationship study has shown that 5-phenyl-4-hydroxythiazole analogues are especially potent. This new class of inhibitors appears to be selective for 5-lipoxygenase, being only weakly active against cyclooxygenase and 12- and 15-lipoxygenase.

Initial studies in vivo using a rat peritoneal anaphylaxis model have shown the hydroxythiazoles to be inactive when they are administered orally (100 mg/kg), probably due to a lack of absorption or solubility. Compound 7, however, did exhibit significant activity when administered ip (85% inhibition of leukotriene biosynthesis at 10 mg/kg). The potential utility of these inhibitors appears to be as topical agents.

#### Experimental Section

**General Methods.** Unless otherwise noted, materials were obtained from commercial suppliers and were used without further purification. <sup>1</sup>H NMR spectra were obtained on a General Electric QE-300 NMR instrument at 300 MHz and on a Varian T-60 NMR instrument at 60 MHz. Chemical shifts are expressed in ppm downfield from internal tetramethylsilane. <sup>1</sup>H NMR data are tabulated in the following order: chemical shift, multiplicity (s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet), number of protons, coupling constant(s) in hertz. Mass spectra were recorded with an HP5985A spectrometer. Merck TLC plates were used for analytical TLC, and Merck Kieselgel 60 was used for column chromatography. Microanalyses were performed by the Abbott Analytical Department.

**Method A.** 2-(3-Pyridyl)-4-hydroxy-5-methylthiazole (5). Pyridine (2 g, 0.025 mol) was added to a mixture of thiolactic acid (10.6 g, 0.1 mol) and 2-cyanopyridine (10.4 g, 0.1 mol) at 23 °C under argon. The reaction mixture was then heated at 100 °C for 2 h. After cooling, the precipitate was collected and washed with absolute ethanol. Recrystallization from methanol afforded the product (14 g, 73%): mp 230 °C (MeOH); <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>)  $\delta$  2.25 (s, 3 H) 7.32–7.67 (m, 1 H), 8.00–8.30 (m, 1 H), 8.54–8.70 (m, 1 H), 8.98–9.10 (m, 1 H), 10.55 (s, 1 H); MS *m/e* 192 (M<sup>+</sup>). Anal. (C<sub>9</sub>H<sub>8</sub>N<sub>2</sub>OS) C, H, N.

The following 4-hydroxythiazoles were prepared in a manner similar to that of 5 with the appropriately substituted nitrile or thiolactic acid derivative.

2-(2-Thienyl)-4-hydroxy-5-methylthiazole (10): 62% yield; mp 152–163 °C (EtOH); <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>)  $\delta$  2.16 (s, 3 H), 7.08–7.28 (m, 1 H), 7.50–7.75 (m, 2 H), 10.32 (s, 1 H); MS *m/e* 197 (M<sup>+</sup>). Anal. (C<sub>9</sub>H<sub>7</sub>NOS<sub>2</sub>) C, H, N.

- (7) Hope, W. C.; Welton, A. F.; Fiedler-Nagy, C.; Batula-Bernardo, C.; Coffey, J. W. *Biochem. Pharmacol.* 1983, 32, 357.
- (8) Panganamala, R. V.; Miller, J. S.; Gwebu, E. T.; Sharma, H. M.; Cornwell, D. G. *Prostaglandins* 1977, 14, 261.
- (9) Carter, G. W.; Young, P. R.; Albert, D. H.; Boucka, J. B.; Dyer, R. D.; Bell, R. L.; Summers, J. B.; Brooks, D. W.; Gunn, B. P.; Rubin, P.; Kesterson, J. In *Leukotrienes and Prostanoids in Health and Disease. New Trends in Lipid Mediators Research*; Zor, U., Naor, Z., Danon, A., Eds.; Karger: Basel, 1989; Vol. 3, pp 50–55.

- (10) Higgs, G. A.; Flower, R. J.; Vane, J. R. *Biochem. Pharmacol.* 1979, 28, 1959.
- (11) Vanderhoek, J. K.; Bryant, R.; Bailey, J. M. *Biochem. Pharmacol.* 1982, 31, 3463.
- (12) Musser, J. H.; Chakraborty, U. R.; Sciortino, S.; Gordon, R. J.; Khandwala, A.; Neiss, E. S.; Pruss, T. P.; Van Inwegen, R.; Weinryb, L.; Coutta, S. M. *J. Med. Chem.* 1987, 30, 96.
- (13) Corey, E. J.; Monroe, J. E. *J. Am. Chem. Soc.* 1982, 104, 1752.

2-(2-Furyl)-4-hydroxy-5-methylthiazole (11): 52% yield; mp 173–174 °C (EtOH);  $^1\text{H}$  NMR (300 MHz, DMSO- $d_6$ )  $\delta$  2.21 (s, 3 H), 6.64–6.68 (m, 1 H), 6.86–6.89 (m, 1 H), 7.78–7.82 (m, 1 H), 10.46 (s, 1 H); MS  $m/e$  181 (M $^+$ ). Anal. ( $\text{C}_8\text{H}_7\text{NO}_2\text{S}$ ) C, H, N.

2-(2-Pyridyl)-4-hydroxy-5-methylthiazole (12): 72% yield; mp 201–202 °C (MeOH) (lit.<sup>14</sup> mp 198–202 °C);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.25 (s, 3 H), 7.35–7.62 (m, 2 H), 8.05–8.35 (m, 2 H), 10.45 (s, 1 H); MS  $m/e$  192 (M $^+$ ). Anal. ( $\text{C}_9\text{H}_6\text{N}_2\text{OS}$ ) C, H, N.

2-(4-Pyridyl)-4-hydroxy-5-methylthiazole (13): 55% yield; mp 223–224 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.28 (s, 3 H), 7.67–7.84 (m, 2 H), 8.67–8.85 (m, 2 H), 10.65 (s, 1 H); MS  $m/e$  192 (M $^+$ ). Anal. ( $\text{C}_9\text{H}_6\text{N}_2\text{OS}$ ) C, H, N.

2-[2-(6-Methoxybenzothiazolyl)]-4-hydroxy-5-methylthiazole (14): 65% yield; mp 249–250 °C (EtOH) (lit.<sup>14</sup> mp 250–253 °C);  $^1\text{H}$  NMR (300 MHz, DMSO- $d_6$ )  $\delta$  2.28 (s, 3 H), 3.86 (s, 3 H), 7.11–7.17 (m, 1 H), 7.68–7.72 (m, 1 H), 7.90–7.96 (m, 1 H), 10.11 (s, 1 H); MS  $m/e$  278 (M $^+$ ). Anal. ( $\text{C}_{12}\text{H}_{10}\text{N}_2\text{O}_2\text{S}_2$ ) C, H, N.

2-(3-Quinolyl)-4-hydroxy-5-methylthiazole (15): 62% yield; mp 279–280 °C (EtOH);  $^1\text{H}$  NMR (300 MHz, DMSO- $d_6$ )  $\delta$  2.28 (s, 3 H), 7.75–7.82 (m, 1 H), 7.60–7.68 (m, 1 H), 8.02–8.06 (s, 1 H), 8.10–8.15 (m, 1 H), 8.69–8.72 (m, 1 H), 9.32–9.35 (m, 1 H), 10.55 (s, 1 H); MS  $m/e$  242 (M $^+$ ). Anal. ( $\text{C}_{13}\text{H}_{10}\text{N}_2\text{O}_2\text{S}$ ) C, H, N.

2-(3-Pyrazolyl)-4-hydroxy-5-methylthiazole (16): 42% yield; mp 124–125 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.16 (s, 3 H), 8.00 (s, 1 H), 8.35 (s, 1 H), 9.02 (s, 1 H); MS  $m/e$  181 (M $^+$ ). Anal. ( $\text{C}_7\text{H}_7\text{N}_3\text{OS}$ ) C, H, N.

2-(Carbomethoxymethyl)-4-hydroxy-5-methylthiazole (18): 64% yield; mp 132–133 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  1.16 (t, 3 H,  $J = 7$  Hz), 2.20 (s, 3 H), 4.08 (q, 2 H,  $J = 7$  Hz), 5.51 (s, 2 H), 10.55 (s, 1 H); MS  $m/e$  201 (M $^+$ ). Anal. ( $\text{C}_8\text{H}_{11}\text{NO}_3\text{S}$ ) C, H, N.

2-Carbomethoxy-4-hydroxy-5-methylthiazole (19). The title compound was prepared from ethyl cyanoformate and thiolactic acid in 65% yield; mp 163–164 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  1.18 (t, 3 H,  $J = 7$  Hz), 2.25 (s, 3 H), 4.35 (q, 2 H,  $J = 7$  Hz), 8.87 (s, 1 H); MS  $m/e$  187 (M $^+$ ). Anal. ( $\text{C}_7\text{H}_9\text{NO}_3\text{S}$ ) C, H, N.

2-Carboxy-4-hydroxy-5-methylthiazole (20). The title compound was prepared by treatment of 19 with LiOH in 85% yield; dec >170 °C;  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.32 (s, 3 H), 9.52 (br s, 1 H); MS  $m/e$  159 (M $^+$ ). Anal. ( $\text{C}_6\text{H}_6\text{NO}_3\text{S}$ ) C, H, N.

2-(4-Fluorophenyl)-4-hydroxy-5-methylthiazole (23): 68% yield; mp 173–174 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.20 (s, 3 H), 7.16–8.00 (m, 4 H), 10.0 (s, 1 H); MS  $m/e$  209 (M $^+$ ). Anal. ( $\text{C}_{10}\text{H}_8\text{FNOS}$ ) C, H, N.

2-(4-Bromophenyl)-4-hydroxy-5-methylthiazole (24): 51% yield; mp 206–207 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.25 (s, 3 H), 7.50–7.90 (m, 4 H), 10.32 (s, 1 H); MS  $m/e$  269 (M $^+$ ). Anal. ( $\text{C}_{10}\text{H}_8\text{BrNOS}$ ) C, H, N.

2-(4-Chlorophenyl)-4-hydroxy-5-methylthiazole (25): 66% yield; mp 198–199 °C (EtOH);  $^1\text{H}$  NMR (300 MHz, DMSO- $d_6$ )  $\delta$  2.23 (s, 3 H), 7.48–7.54 (m, 2 H), 7.75–7.83 (m, 2 H), 10.40 (br s, 1 H); MS  $m/e$  225 (M $^+$ ). Anal. ( $\text{C}_{10}\text{H}_8\text{ClNOS}$ ) C, H, N.

2-(4-Cyanophenyl)-4-hydroxy-5-methylthiazole (27): 62% yield; mp 220–221 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.26 (s, 3 H), 7.85–8.10 (m, 4 H), 10.55 (br s, 1 H); MS  $m/e$  216 (M $^+$ ). Anal. ( $\text{C}_{11}\text{H}_8\text{N}_2\text{OS}$ ) C, H, N.

2-(4-Carbomethoxyphenyl)-4-hydroxy-5-methylthiazole (28): 65% yield; mp 207–208 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  1.25 (t, 3 H,  $J = 7$  Hz), 2.35 (s, 3 H), 4.32 (q, 2 H,  $J = 7$  Hz), 7.85–8.15 (m, 4 H), 10.41 (br s, 1 H); MS  $m/e$  263 (M $^+$ ). Anal. ( $\text{C}_{13}\text{H}_{13}\text{NO}_3\text{S}$ ) C, H, N.

2-(4-Carbomethoxyphenyl)-4-hydroxy-5-methylthiazole (29): 64% yield; mp 219–220 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.35 (s, 3 H), 3.80 (s, 3 H), 7.85–8.15 (m, 4 H), 10.41 (br s, 1 H); MS  $m/e$  249 (M $^+$ ). Anal. ( $\text{C}_{12}\text{H}_{11}\text{NO}_3\text{S}$ ) C, H, N.

2-[4-(Phenethyloxy)carbonyl]phenyl]-4-hydroxy-5-methylthiazole (30): 58% yield; mp 251–252 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.25 (s, 3 H), 3.08 (t, 2 H,  $J = 7$  Hz),

4.55 (t, 2 H,  $J = 7$  Hz), 7.32–7.50 (m, 5 H), 7.95–8.05 (m, 4 H), 10.35 (br s, 1 H); MS  $m/e$  339 (M $^+$ ). Anal. ( $\text{C}_{19}\text{H}_{17}\text{NO}_3\text{S}$ ) C, H, N.

2-(4-Acetylphenyl)-4-hydroxy-5-methylthiazole (32): 64% yield; mp 219–220 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.25 (s, 3 H), 2.59 (s, 3 H), 7.89–7.95 (m, 2 H), 8.0–8.6 (m, 2 H), 10.50 (s, 1 H); MS  $m/e$  233 (M $^+$ ). Anal. ( $\text{C}_{12}\text{H}_{11}\text{NO}_2\text{S}$ ) C, H, N.

2-(4-Nitrophenyl)-4-hydroxy-5-methylthiazole (33): 52% yield; mp 244–248 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.25 (s, 3 H), 7.85–8.50 (m, 4 H), 10.41 (br s, 1 H); MS  $m/e$  236 (M $^+$ ). Anal. ( $\text{C}_{10}\text{H}_8\text{N}_2\text{O}_3\text{S}$ ) C, H, N.

2-(4-Sulfamoylphenyl)-4-hydroxy-5-methylthiazole (34): 45% yield; mp 256–257 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.20 (s, 3 H), 6.65 (s, 2 H), 7.32–7.66 (m, 2 H), 7.85–8.10 (m, 2 H); MS  $m/e$  270 (M $^+$ ). Anal. ( $\text{C}_{10}\text{H}_{10}\text{N}_2\text{O}_5\text{S}_2$ ) C, H, N.

2-[4-(Trifluoromethyl)phenyl]-4-hydroxy-5-methylthiazole (35): 48% yield; mp 232–233 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.20 (s, 3 H), 7.66–8.10 (m, 4 H), 10.50 (s, 1 H); MS  $m/e$  259 (M $^+$ ). Anal. ( $\text{C}_{11}\text{H}_8\text{F}_3\text{NOS}$ ) C, H, N.

2-[4-(Trifluoromethyl)thio]phenyl]-4-hydroxy-5-methylthiazole (36): 64% yield; mp 178–179 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.18 (s, 3 H), 3.32 (br s, 1 H), 7.66–8.05 (m, 4 H), 9.75 (br s, 1 H); MS  $m/e$  291 (M $^+$ ). Anal. ( $\text{C}_{11}\text{H}_8\text{F}_3\text{NOS}_2$ ) C, H, N.

2-(4-Carbamoylphenyl)-4-hydroxy-5-methylthiazole (37): 74% yield; mp 274–277 °C dec (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.21 (s, 3 H), 7.30–8.10 (m, 5 H), 10.15 (br s, 1 H); MS  $m/e$  234 (M $^+$ ). Anal. ( $\text{C}_{11}\text{H}_{10}\text{N}_2\text{O}_2\text{S}$ ) C, H, N.

2-(4-Aminophenyl)-4-hydroxy-5-methylthiazole (38): 42% yield; mp 168–169 °C (EtOH);  $^1\text{H}$  NMR (300 MHz, DMSO- $d_6$ )  $\delta$  1.53 (d, 3 H,  $J = 7$  Hz), 4.35 (q, 1 H,  $J = 7$  Hz), 6.63–6.72 (m, 4 H), 7.75–7.85 (m, 2 H); MS  $m/e$  206 (M $^+$ ). Anal. ( $\text{C}_{10}\text{H}_{10}\text{N}_2\text{OS}$ ) C, H, N.

2-(4-Carboxyphenyl)-4-hydroxy-5-methylthiazole (39): 54% yield; mp 276 °C dec (EtOH);  $^1\text{H}$  NMR (300 MHz, DMSO- $d_6$ )  $\delta$  2.25 (s, 3 H), 7.88–7.94 (m, 2 H), 7.98–8.04 (m, 2 H), 10.49 (br s, 1 H), 12.07 (br s, 1 H); MS  $m/e$  235 (M $^+$ ). Anal. ( $\text{C}_{11}\text{H}_8\text{NO}_3\text{S}$ ) C, H, N.

2-(4-Biphenyl)-4-hydroxy-5-methylthiazole (40): 45% yield; mp 265–266 °C (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  2.16 (s, 3 H), 7.32–8.00 (m, 9 H), 10.25 (s, 1 H); MS  $m/e$  267 (M $^+$ ). Anal. ( $\text{C}_{16}\text{H}_{13}\text{NOS}$ ) C, H, N.

2-(2-Fluorophenyl)-4-hydroxy-5-methylthiazole (41): 48% yield; mp 159–160 °C (EtOH);  $^1\text{H}$  NMR (300 MHz, DMSO- $d_6$ )  $\delta$  2.25 (s, 3 H), 7.30–7.51 (m, 4 H), 8.05–8.15 (m, 1 H), 10.43 (br s, 1 H); MS  $m/e$  209 (M $^+$ ). Anal. ( $\text{C}_{10}\text{H}_8\text{FNOS}$ ) C, H, N.

2-(3-Fluorophenyl)-4-hydroxy-5-methylthiazole (42): 44% yield; mp 162–163 °C (EtOH);  $^1\text{H}$  NMR (300 MHz, DMSO- $d_6$ )  $\delta$  2.25 (s, 3 H), 7.23–7.31 (m, 1 H), 7.48–7.66 (m, 3 H), 10.45 (s, 1 H); MS  $m/e$  209 (M $^+$ ). Anal. ( $\text{C}_{10}\text{H}_8\text{FNOS}$ ) C, H, N.

2-(4-Carboxyphenyl)-4-hydroxy-5-phenylthiazole (74): 35% yield; mp >300 °C dec (EtOH);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  7.32–8.15 (m, 9 H), 11.32 (br s, 1 H); MS  $m/e$  297 (M $^+$ ). Anal. ( $\text{C}_{16}\text{H}_{11}\text{NO}_3\text{S}$ ) C, H, N.

**Method B. 2-Phenyl-4-hydroxy-5-methylthiazole (1).** Ethyl bromopropionate (3.96 g, 21.9 mmol) was added dropwise to a solution of thiobenzamide (3.00 g, 21.9 mmol) and pyridine (7 mL, 87.5 mmol) in toluene (200 mL) at 23 °C. The reaction mixture was heated to 80 °C for 2 h and allowed to cool to 23 °C. The precipitate was collected and recrystallized from ethanol to afford 3.3 g (81%) of product; mp 192–193 °C (EtOH) (lit.<sup>15</sup> mp 190–191 °C);  $^1\text{H}$  NMR (300 MHz, DMSO- $d_6$ )  $\delta$  2.20 (s, 3 H), 7.32–7.55 (m, 3 H), 7.75–7.82 (m, 2 H), 10.31 (s, 1 H); MS  $m/e$  191 (M $^+$ ). Anal. ( $\text{C}_{10}\text{H}_9\text{NOS}$ ) C, H, N.

The following 4-hydroxythiazoles were prepared in a manner similar to that of 1 with the appropriately substituted benzamide or  $\alpha$ -bromo ester.

2-Phenyl-4-thiazolinone (2): 42% yield; mp 107 °C (ether) (lit.<sup>15</sup> mp 106–108 °C);  $^1\text{H}$  NMR (60 MHz, DMSO- $d_6$ )  $\delta$  3.70 (s, 2 H), 7.30–7.95 (m, 5 H); MS  $m/e$  177 (M $^+$ ). Anal. ( $\text{C}_9\text{H}_7\text{NOS}$ ) C, H, N.

2,5-Diphenyl-4-hydroxythiazole (7): 68% yield; mp 212–213 °C (EtOH) (lit.<sup>16</sup> mp 215 °C);  $^1\text{H}$  NMR (300 MHz,  $\text{CDCl}_3$ )  $\delta$

(14) Suzuki, N.; Goto, T. *Agric. Biol. Chem. Jpn.* 1972, 36, 2213.

(15) Jensen, K. A.; Crossland, I. *Acta Chem. Scand.* 1963, 17, 144.

7.22–7.30 (m, 2 H), 7.38–7.53 (m, 4 H), 7.82–7.87 (m, 2 H), 7.92–8.00 (m, 2 H); MS *m/e* 253 (M<sup>+</sup>). Anal. (C<sub>15</sub>H<sub>11</sub>NOS) C, H, N.

2,5-Dimethyl-4-hydroxythiazole (17): 48% yield; mp 101–103 °C; <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>) δ 2.20 (s, 3 H), 2.56 (s, 3 H), 8.65 (br s, 1 H); MS *m/e* 129 (M<sup>+</sup>). Anal. (C<sub>8</sub>H<sub>7</sub>NOS) C, H, N.

2-Amino-4-hydroxy-5-methylthiazole (21): 44% yield; mp 175–176 °C (EtOH); <sup>1</sup>H NMR (60 MHz, CDCl<sub>3</sub>) δ 2.25 (s, 3 H), 7.25–7.38 (m, 5 H), 9.85 (br s, 1 H), 8.55 (br s, 2 H); MS *m/e* 130 (M<sup>+</sup>). Anal. (C<sub>6</sub>H<sub>8</sub>N<sub>2</sub>OS) C, H, N.

2-Hydrazino-5-methyl-4-thiazolinone (22): 58% yield; mp 242–243 °C (EtOH); <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>) δ 1.50 (d, 3 H, *J* = 7 Hz), 4.53 (q, 1 H, *J* = 7 Hz), 5.82 (br s, 3 H), 11.05 (br s, 1 H); MS *m/e* 145 (M<sup>+</sup>). Anal. (C<sub>4</sub>H<sub>7</sub>N<sub>3</sub>OS), C, H, N.

2-(4-Methylphenyl)-4-hydroxy-5-methylthiazole (26): 71% yield; mp 172–174 °C (EtOH); <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>) δ 2.15 (s, 3 H), 2.25 (s, 3 H), 7.15–7.35 (m, 2 H), 7.55–7.85 (m, 2 H), 9.82 (s, 1 H); MS *m/e* 205 (M<sup>+</sup>). Anal. (C<sub>11</sub>H<sub>11</sub>NOS) C, H, N.

2-(4-Methoxyphenyl)-4-hydroxy-5-methylthiazole (31): 33% yield; mp 149–150 °C (EtOH); <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>) δ 2.18 (s, 3 H), 7.00–7.20 (m, 2 H), 7.67–7.97 (m, 2 H), 10.41 (br s, 1 H); MS *m/e* 221 (M<sup>+</sup>). Anal. (C<sub>11</sub>H<sub>11</sub>NO<sub>2</sub>S) C, H, N.

2-Phenyl-4-hydroxy-5-propylthiazole (51): 56% yield; mp 86–87 °C (EtOH); <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>) δ 0.92 (t, 3 H, *J* = 7 Hz), 1.50–1.65 (m, 2 H), 2.62 (t, 2 H, *J* = 7 Hz), 7.35–7.55 (m, 3 H), 7.75–7.83 (m, 2 H), 10.28 (s, 1 H); MS *m/e* 219 (M<sup>+</sup>). Anal. (C<sub>12</sub>H<sub>13</sub>NOS) C, H, N.

2-Phenyl-4-hydroxy-5-butylthiazole (60): 71% yield; mp 69–71 °C (EtOH); <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>) δ 0.90 (t, 3 H, *J* = 7 Hz), 1.28–1.41 (m, 2 H), 1.48–1.60 (m, 2 H), 2.63 (t, 2 H, *J* = 7 Hz), 7.40–7.50 (m, 3 H), 7.82–7.85 (m, 2 H), 10.35 (s, 1 H); MS *m/e* 233 (M<sup>+</sup>). Anal. (C<sub>13</sub>H<sub>15</sub>NOS) C, H, N.

2-(4-Biphenyl)-4-hydroxy-5-phenylthiazole (63): 67% yield; mp 242–243 °C (EtOH); <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>) δ 7.15–8.05 (m, 14 H), 11.25 (br s, 1 H); MS *m/e* 329 (M<sup>+</sup>). Anal. (C<sub>21</sub>H<sub>15</sub>NOS) C, H, N.

2-Phenyl-4-hydroxy-5-ethylthiazole (65): 52% yield; mp 175–177 °C dec (MeOH); <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>) δ 1.18 (t, 3 H, *J* = 7 Hz), 2.66 (q, 2 H, *J* = 7 Hz), 7.40–7.50 (m, 3 H), 7.75–7.85 (m, 2 H), 9.52 (br s, 1 H); MS *m/e* 205 (M<sup>+</sup>). Anal. (C<sub>11</sub>H<sub>11</sub>NOS) C, H, N.

2-Phenyl-4-hydroxy-5-phenethylthiazole (66): 68% yield; mp 127–128 °C (EtOH); <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>) δ 2.88 (s, 4 H), 7.21–7.90 (m, 10 H), 9.84 (br s, 1 H); MS *m/e* 281 (M<sup>+</sup>). Anal. (C<sub>17</sub>H<sub>15</sub>NOS) C, H, N.

2-Phenyl-4-hydroxy-5-(carboxymethyl)thiazole (67). The title compound was prepared by treatment of 68 with LiOH in 96% yield; mp 196 °C dec (EtOH); <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>) δ 3.65 (s, 2 H), 7.35–7.55 (m, 3 H), 7.72–7.89 (m, 2 H), 10.55 (br s, 1 H), 12.52 (br s, 1 H); MS *m/e* 235 (M<sup>+</sup>). Anal. (C<sub>11</sub>H<sub>9</sub>NO<sub>3</sub>S) C, H, N.

2-Phenyl-4-hydroxy-5-(carboxymethoxymethyl)thiazole (68): 62% yield; mp 114–116 °C (EtOH); <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>) δ 3.65 (s, 2 H), 3.80 (s, 3 H), 7.35–7.55 (m, 3 H), 7.72–7.89 (m, 2 H), 10.50 (s, 1 H); MS *m/e* 249 (M<sup>+</sup>). Anal. (C<sub>12</sub>H<sub>11</sub>NO<sub>3</sub>S) C, H, N.

2-Phenyl-4-hydroxy-5-[(*N*-methyl-*N*-hydroxyamino)-carbonyl]methylthiazole (69). The title compound was prepared from the corresponding acid chloride by treatment with methylhydroxyamine hydrochloride and triethylamine in 95% yield; mp 156–157 °C (ether); <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>) δ 3.12 (s, 3 H), 3.80 (s, 2 H), 7.38–7.52 (m, 3 H), 7.75–7.86 (m, 2 H), 10.10 (br s, 1 H), 10.51 (s, 1 H); MS *m/e* 264 (M<sup>+</sup>). Anal. (C<sub>12</sub>H<sub>12</sub>N<sub>2</sub>O<sub>3</sub>S) C, H, N.

2-Phenyl-4-hydroxy-5-carbomethoxythiazole (70): 62% yield; mp 91–92 °C (EtOH) (lit.<sup>17</sup> mp 91–92 °C); <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>) δ 1.18 (t, 3 H, *J* = 7 Hz), 4.25 (q, 2 H, *J* = 7 Hz), 7.35–8.05 (m, 5 H); MS *m/e* 249 (M<sup>+</sup>). Anal. (C<sub>12</sub>H<sub>11</sub>NO<sub>3</sub>S) C, H, N.

2-Methyl-4-hydroxy-5-phenylthiazole (71): 41% yield; mp 208–211 °C (EtOH) (lit.<sup>18</sup> 210–211 °C); <sup>1</sup>H NMR (300 MHz,

DMSO-*d*<sub>6</sub>) δ 2.56 (s, 3 H), 7.13–7.20 (m, 1 H), 7.30–7.40 (m, 2 H), 7.59–7.65 (m, 4 H), 8.50 (br s, 1 H); MS *m/e* 191 (M<sup>+</sup>). Anal. (C<sub>10</sub>H<sub>9</sub>NOS) C, H, N.

2-(4-Fluorophenyl)-4-hydroxy-5-phenylthiazole (72): 52% yield; mp 231–233 °C (EtOH); <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>) δ 7.20–8.05 (m, 9 H), 11.10 (br s, 1 H); MS *m/e* 271 (M<sup>+</sup>). Anal. (C<sub>15</sub>H<sub>10</sub>FNOS) C, H, N.

2-(4-Methylphenyl)-4-hydroxy-5-phenylthiazole (73): 71% yield; mp 252–255 °C (EtOH); <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>) δ 2.32 (s, 3 H), 7.20–7.95 (m, 9 H), 10.75 (br s, 1 H); MS *m/e* 267 (M<sup>+</sup>). Anal. (C<sub>15</sub>H<sub>13</sub>NOS) C, H, N.

2-(4-Methoxyphenyl)-4-hydroxy-5-phenylthiazole (75): 52% yield; mp 218–220 °C (EtOH); <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>) δ 3.84 (s, 3 H), 7.0–8.0 (m, 9 H), 11.05 (br s, 1 H); MS *m/e* 283 (M<sup>+</sup>). Anal. (C<sub>16</sub>H<sub>13</sub>NO<sub>2</sub>S) C, H, N.

2-(4-Pyridyl)-4-hydroxy-5-phenylthiazole (76): 44% yield; mp 280 °C dec (EtOH); <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>) δ 7.15–7.95 (m, 9 H), 8.55 (br s, 1 H); MS *m/e* 254 (M<sup>+</sup>). Anal. (C<sub>14</sub>H<sub>10</sub>N<sub>2</sub>OS) C, H, N.

2-(3-Pyridyl)-4-hydroxy-5-phenylthiazole (77): 41% yield; mp 273–276 °C (EtOH); <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>) δ 7.20–7.30 (m, 1 H), 7.35–7.48 (m, 2 H), 7.52–7.80 (m, 3 H), 8.25–8.40 (m, 1 H), 8.63–8.80 (m, 1 H), 9.01–9.15 (m, 1 H), 10.20 (br s, 1 H); MS *m/e* 254 (M<sup>+</sup>). Anal. (C<sub>14</sub>H<sub>10</sub>N<sub>2</sub>OS) C, H, N.

**Preparation of Acetates.** Method C. 2,5-Diphenyl-4-acetoxythiazole (46). The title compound was prepared by reacting hydroxythiazole (7) (2.53 g, 10 mmol) in methylene chloride (250 mL) with pyridine (0.8 g, 10 mmol) and acetic anhydride (1.5 g, 15 mmol) at 23 °C for 10 h. The reaction mixture was washed with water and dried (MgSO<sub>4</sub>). Removal of solvent, followed by recrystallization of the crude residue provided the product (2.72 g, 92%); mp 101–103 °C (EtOAc/hexane); <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>) δ 2.40 (s, 3 H), 7.38–7.62 (m, 8 H), 7.86–7.96 (m, 2 H); MS *m/e* 295 (M<sup>+</sup>). Anal. (C<sub>17</sub>H<sub>13</sub>NO<sub>2</sub>S) C, H, N. The following compounds were prepared in a similar manner from the corresponding 4-hydroxythiazole as compound 46.

2-Phenyl-4-acetoxy-5-propylthiazole (52): 92% yield; colorless oil; <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>) δ 0.90 (t, 2 H, *J* = 7 Hz), 1.50–1.65 (m, 2 H), 2.28 (s, 3 H), 7.62 (t, 2 H, *J* = 7 Hz), 7.35–7.55 (m, 3 H), 7.75–7.83 (m, 2 H); MS *m/e* 261 (M<sup>+</sup>). Anal. (C<sub>14</sub>H<sub>15</sub>NO<sub>2</sub>S) C, H, N.

2-Phenyl-4-acetoxy-5-butylthiazole (61): 81% yield; colorless oil; <sup>1</sup>H NMR (60 MHz, CDCl<sub>3</sub>) δ 0.85 (t, 3 H, *J* = 7 Hz), 1.0–1.65 (m, 4 H), 2.20 (s, 3 H), 2.55 (t, 2 H, *J* = 7 Hz), 7.32–7.50 (m, 3 H), 7.80–8.00 (m, 2 H); MS *m/e* 275 (M<sup>+</sup>). Anal. (C<sub>15</sub>H<sub>17</sub>NO<sub>2</sub>S) C, H, N.

2-(4-Carboxyphenyl)-4-acetoxy-5-methylthiazole (62): 91% yield; mp 227–230 °C (EtOH); <sup>1</sup>H NMR (60 MHz, CDCl<sub>3</sub>) δ 2.30 (s, 3 H), 2.32 (s, 3 H), 7.84–8.20 (m, 4 H); MS *m/e* 277 (M<sup>+</sup>). Anal. (C<sub>13</sub>H<sub>11</sub>NO<sub>4</sub>S) C, H, N.

2-(4-Biphenyl)-4-acetoxy-5-phenylthiazole (64): 89% yield; mp 146–147 °C (EtOH); <sup>1</sup>H NMR (60 MHz, CDCl<sub>3</sub>) δ 2.30 (s, 3 H), 7.30–8.05 (m, 14 H); MS *m/e* 371 (M<sup>+</sup>). Anal. (C<sub>23</sub>H<sub>17</sub>NO<sub>2</sub>S) C, H, N.

**Preparation of Esters.** Method D. 2,5-Diphenyl-4-[(ethoxysuccinyl)oxy]thiazole (47). The title compound was prepared by reacting compounds 7 (913 mg, 3.6 mmol) in methylene chloride (50 mL) with 4-(dimethylamino)pyridine (710 mg, 5.8 mmol) and ethyl succinyl chloride (924 mg, 5.61 mmol) at 23 °C under nitrogen for 2 h. The organic layer was washed with water and dried (Na<sub>2</sub>SO<sub>4</sub>). Removal of solvent, followed by recrystallization of the crude residue with EtOAc/hexane provided the product (1.34 g, 97%); mp 55–58 °C; <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 1.25 (q, *J* = 7 Hz, 3 H), 2.73 (t, *J* = 7 Hz, 2 H), 2.98 (t, *J* = 7 Hz, 2 H), 4.15 (q, *J* = 7 Hz, 2 H), 7.45 (m, 6 H), 7.55 (m, 2 H), 7.92 (m, 2 H); MS *m/e* 381 (M<sup>+</sup>). Anal. (C<sub>21</sub>H<sub>15</sub>NO<sub>4</sub>S) C, H, N.

The following compounds were prepared in a manner similar to that of compound 47 using the corresponding 4-hydroxythiazole and an appropriately substituted acid chloride.

2,5-Diphenyl-4-(hexanoyloxy)thiazole (48): 96% yield; mp 70–72 °C (EtOH); <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 0.90 (t, 3 H, *J* = 7 Hz), 1.23–1.40 (m, 4 H), 1.65–1.80 (m, 2 H), 2.61 (t, 2 H, *J*

(16) Ferrey, M.; Robert, A.; Foucaud, A. *Synthesis* 1976, 261.

(17) Duro, F. *Gazz. Chim. Ital.* 1963, 93, 215.

(18) Reeve, W.; Barrow, E. R. *J. Org. Chem.* 1976, 40, 1917.

= 7 Hz), 7.28–7.58 (m, 8 H), 7.88–7.96 (m, 2 H); MS *m/e* 351 (M<sup>+</sup>). Anal. (C<sub>21</sub>H<sub>21</sub>NO<sub>2</sub>S) C, H, N.

**2,5-Diphenyl-4-(trimethylacetoxymethyl)thiazole (49):** 85% yield; 134–136 °C (EtOAc/hexane); <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 1.38 (s, 9 H), 7.40 (m, 6 H), 7.56 (m, 2 H), 7.92 (m, 2 H); MS *m/e* 337 (M<sup>+</sup>). Anal. (C<sub>20</sub>H<sub>19</sub>NO<sub>2</sub>S) C, H, N.

**Preparation of Carbamates. Method E. 2-Phenyl-4-[(N-methylcarbamyl)oxy]-5-propylthiazole (53).** The title compound was prepared by reacting compound 51 (2.33 g, 10 mmol) in benzene (250 mL) with triethylamine (1.0 g, 10 mmol) and methyl isocyanate (0.65 mL, 10 mmol) at 23 °C under nitrogen for 20 h. The reaction mixture was washed with saturated aqueous NH<sub>4</sub>Cl and water and dried (MgSO<sub>4</sub>). The organic solvent was removed and the residue recrystallized from toluene to afford the product (2.4 g, 86%); mp 55–58 °C (toluene); <sup>1</sup>H NMR (60 MHz, CDCl<sub>3</sub>) δ 0.90 (t, 3 H, *J* = 7 Hz), 1.67 (m, 2 H), 2.66 (t, 2 H, *J* = 7 Hz), 2.85 (d, 3 H, *J* = 7 Hz), 5.50 (br s, 1 H), 7.75 (m, 2 H), 7.40 (m, 3 H); MS *m/e* 276 (M<sup>+</sup>). Anal. (C<sub>14</sub>H<sub>16</sub>N<sub>2</sub>O<sub>2</sub>S) C, H, N.

The following compounds were prepared in a similar manner to that of compound 53 from the corresponding 4-hydroxythiazole and an appropriately substituted isocyanate.

**2-Phenyl-4-[(N-tert-butylcarbamyl)oxy]-5-propylthiazole (54):** 84% yield; mp 97–98 °C (benzene); <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 0.89 (t, 3 H, *J* = 7 Hz), 1.21–1.45 (m, 2 H), 0.99 (s, 9 H), 2.55 (t, 2 H, *J* = 7 Hz), 7.05–7.10 (m, 1 H), 7.30–7.60 (m, 4 H); MS *m/e* 318 (M<sup>+</sup>). Anal. (C<sub>17</sub>H<sub>22</sub>N<sub>2</sub>O<sub>2</sub>S) C, H, N.

**2-Phenyl-4-[(N-phenylcarbamyl)oxy]-5-propylthiazole (55):** 65% yield; mp 104–105 °C (ether); <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 0.99 (t, 3 H, *J* = 7 Hz), 1.58–1.63 (m, 2 H), 2.61 (t, 2 H, *J* = 7 Hz), 7.05–7.16 (m, 1 H), 7.30–7.50 (m, 8 H), 7.80–7.90 (m, 2 H); MS *m/e* 338 (M<sup>+</sup>). Anal. (C<sub>19</sub>H<sub>18</sub>N<sub>2</sub>O<sub>2</sub>S) C, H, N.

**Preparation of Carbonates. Method F. 2-Phenyl-4-[(ethoxycarbonyl)oxy]-5-propylthiazole (56).** The title compound was prepared by reacting compound 51 (2.3 g, 10 mmol) and pyridine (0.9 mL, 10 mmol) in toluene (200 mL) with ethyl chloroformate (1 mL, 10 mmol) at 23 °C under nitrogen for 2 h. The organic solvent was washed with water and dried (Na<sub>2</sub>SO<sub>4</sub>). The organic solvent was removed and the crude residue chromatographed (silica gel, CHCl<sub>3</sub>) to afford the product (2.1 g, 76%) as a colorless oil; <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 1.00 (t, 3 H, *J* = 7 Hz), 1.40 (t, 3 H, *J* = 7 Hz), 1.62–1.76 (m, 2 H), 2.69 (t, 2 H, *J* = 7 Hz), 4.35 (q, 2 H, *J* = 7 Hz), 7.40 (m, 3 H), 7.87 (m, 2 H); MS *m/e* 291 (M<sup>+</sup>). Anal. (C<sub>15</sub>H<sub>17</sub>NO<sub>3</sub>S) C, H, N.

The following compounds were prepared in a manner similar to that of compound 56 from the corresponding 4-hydroxythiazole and the appropriately substituted chloroformate.

**2-Phenyl-4-[(benzoxycarbonyl)oxy]-5-propylthiazole (57):** 88% yield; mp 62–64 °C (toluene); <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>) δ 7.15–8.05 (m, 14 H), 11.25 (br s, 1 H); MS *m/e* 329 (M<sup>+</sup>). Anal. (C<sub>20</sub>H<sub>19</sub>NO<sub>3</sub>S) C, H, N.

**2-Phenyl-4-[(isopropoxycarbonyl)oxy]-5-propylthiazole (58):** 96% yield; colorless oil; <sup>1</sup>H NMR (60 MHz, CDCl<sub>3</sub>) δ 0.85 (t, 2 H, *J* = 7 Hz), 1.25 (d, 6 H, *J* = 7 Hz), 1.60–1.87 (m, 2 H), 2.55 (t, 2 H, *J* = 7 Hz), 5.0 (m, 1 H), 7.30–7.50 (m, 3 H), 7.65–7.95 (m, 2 H); MS *m/e* 305 (M<sup>+</sup>). Anal. (C<sub>18</sub>H<sub>19</sub>NO<sub>3</sub>S) C, H, N.

**2,5-Diphenyl-4-oxazolinone (8).** The title compound was prepared by the method of Rao and Filler.<sup>19</sup> 53% yield; mp 159–161 °C (lit.<sup>19</sup> mp 161 °C); <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>) δ 6.65 (s, 1 H), 7.28–7.70 (m, 8 H), 8.00–8.15 (m, 2 H); MS *m/e* 237 (M<sup>+</sup>). Anal. (C<sub>15</sub>H<sub>11</sub>NO<sub>2</sub>) C, H, N.

**2,5-Diphenyl-4-hydroxyimidazole (9).** The title compound was prepared by the method of Jeffrey.<sup>20</sup> 21% yield; mp 162–164 °C (lit.<sup>20</sup> mp 162–164 °C); <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>) δ 7.00–8.20 (m, 10 H), 11.8 (br s, 1 H); MS *m/e* 236 (M<sup>+</sup>). Anal. (C<sub>15</sub>H<sub>12</sub>N<sub>2</sub>O) C, H, N.

**2,5-Diphenyl-4-aminothiazole (43).** The title compound was prepared by reacting α-cyanobenzyl benzenesulfonate (21 g, 80 mmol) in EtOH (200 mL) with thiobenzamide (11 g, 80 mmol) at 0 °C for 20 h. The yellow precipitate was collected and recrystallized from ethanol to afford the benzenesulfonate salt (12.8

g). The salt (12.8 g) was dissolved in concentrated NH<sub>4</sub>OH/H<sub>2</sub>O 1/1 (200 mL) and stirred for 1 h. The precipitate was collected and recrystallized from H<sub>2</sub>O/EtOH to give the product (7.4 g, 38%); mp 92–94 °C (lit.<sup>21</sup> mp 103–104 °C); <sup>1</sup>H NMR (60 MHz, DMSO-*d*<sub>6</sub>) δ 5.83 (br s, 2 H), 7.25–8.00 (m, 10 H); MS *m/e* 252 (M<sup>+</sup>). Anal. (C<sub>15</sub>H<sub>12</sub>N<sub>2</sub>S) C, H, N.

**2,5-Diphenyl-4-methoxythiazole (44).** The title compound was prepared by reacting hydroxythiazole (7) (180 mg, 0.711 mmol) in ethanol (20 mL) at 0 °C with excess ethereal diazomethane. The reaction mixture was stirred for 2 h and the solvent removed. Recrystallization of the crude residue from EtOAc/hexane afforded the product (181 mg, 95%); mp 82–83 °C (EtOAc/hexane); <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 4.19 (s, 3 H), 7.18–7.25 (m, 1 H), 7.35–7.45 (m, 5 H), 7.70–7.82 (m, 2 H), 7.90–7.96 (m, 2 H); MS *m/e* 267 (M<sup>+</sup>). Anal. (C<sub>16</sub>H<sub>13</sub>NOS) C, H, N.

**2,5-Diphenyl-4-[(dimethylphosphono)oxy]thiazole (45).** Phosphorus oxychloride (4 mL) was added to compound 7 (2.5 g, 10 mmol) in benzene (40 mL) at 23 °C under nitrogen and the reaction stirred for 6 h. The benzene was removed and methanol (40 mL) added. The reaction mixture was stirred for 1 h and the solvent removed to afford a crude material. Recrystallization from ether gave the product (2.3 g, 66%); mp 98–99 °C (ether); <sup>1</sup>H NMR (300 MHz, CDCl<sub>3</sub>) δ 3.93 (s, 3 H), 3.98 (s, 3 H), 7.30–7.50 (m, 6 H), 7.62–7.71 (m, 2 H), 7.88–7.95 (m, 2 H); MS *m/e* 361 (M<sup>+</sup>). Anal. (C<sub>17</sub>H<sub>16</sub>NO<sub>2</sub>PS) C, H, N.

**2,4-Diphenyl-5-hydroxythiazole (50).** The title compound was prepared by reacting methyl thionobenzoate (3.86 g, 25 mmol) in ether (20 mL) with phenylglycine (4.22 g, 27 mmol) in 3 N NaOH (30 mL) at 23 °C under nitrogen for 18 h. The aqueous layer was separated and acidified slowly with 3 N HCl. A yellow precipitate formed and was collected. The yellow precipitate was then dissolved in acetic anhydride (25 mL) and heated at 100 °C for 1 h. After cooling to 23 °C, the acetic anhydride was removed in vacuo at 60 °C. The crude residue was dissolved in isopropyl alcohol/water 2/1 (60 mL) and lithium hydroxide (0.56 g, 13.6 mmol) added. After stirring at 23 °C for 4 h, water was added and the mixture extracted with methylene chloride. Chromatography (silica gel, CH<sub>2</sub>Cl<sub>2</sub>/pentane 3/1) afforded the product (2.6 g, 42%); mp 133–136 °C (EtOAc) (lit.<sup>22</sup> mp 134–135 °C); <sup>1</sup>H NMR (300 MHz, DMSO-*d*<sub>6</sub>) δ 7.30–7.60 (m, 8 H), 7.90–8.00 (m, 2 H); MS *m/e* 253 (M<sup>+</sup>). Anal. (C<sub>18</sub>H<sub>11</sub>NOS) C, H, N.

**2-Phenyl-4-[(methylsulfonyl)oxy]-5-propylthiazole (59).** The title compound was prepared by reacting compound 51 (650 mg, 2.96 mmol) in methylene chloride (20 mL) with triethylamine (0.42 mL, 3.00 mmol) and methanesulfonyl chloride (0.25 mL, 3.26 mmol) at 23 °C for 1 h. The reaction mixture was washed with water and dried (Na<sub>2</sub>SO<sub>4</sub>). Removal of solvent followed by recrystallization from ether afforded the product (550 mg, 82%); mp 82–84 °C; <sup>1</sup>H NMR (60 MHz, CDCl<sub>3</sub>) δ 0.90 (t, 3 H, *J* = 7 Hz), 1.67 (m, 2 H), 2.80 (t, 2 H, *J* = 7 Hz), 3.50 (s, 3 H), 7.25–8.00 (m, 5 H); MS *m/e* 297 (M<sup>+</sup>). Anal. (C<sub>13</sub>H<sub>15</sub>NO<sub>3</sub>S<sub>2</sub>) C, H, N.

**RBL-1 5-Lipoxygenase Assay.** 5-Lipoxygenase activity was measured in the 20000g supernatant from homogenized rat basophilic leukemia (RBL-1) cells. Inhibitors or vehicle (2% Me<sub>2</sub>SO) were preincubated for 20 min with the RBL-1 supernatant (7.5 × 10<sup>6</sup> cell equiv/mL) at 37 °C in pH 6.8 buffer (10 mM BES, 10 mM PIPES, 1 mM EDTA, 0.1 M NaCl, 0.7 mM CaCl<sub>2</sub>) prior to initiating the 5-lipoxygenase reaction by addition of 66 μM [<sup>14</sup>C]arachidonic acid. [<sup>3</sup>H]-5-HETE added to the reaction mixture served as a recovery standard. Reactions were terminated by acidification to pH 3 and the mixtures were extracted with diethyl ether. The ether extracts were evaporated under nitrogen and the reaction products were separated from nonconverted substrate by thin-layer chromatography. Radioactivity comigrating with 5-HETE was measured by liquid scintillation counting and corrected for recovery of [<sup>3</sup>H]-5-HETE. Inhibition was calculated as the percent reduction from control levels of [<sup>14</sup>C]-5-HETE formation. Concentrations causing 50% inhibition (IC<sub>50</sub>'s) and their 95% confidence limits were calculated as the 50% intercept and their fiducial limits from linear-regression analysis of percent inhibition vs log concentration plots.

(19) Rao, Y. S.; Filler, R. *J. Chem. Soc. Chem. Commun.* 1970, 1622.

(20) Jeffrey, E. A.; Johansen, O.; Meisters, A. *Aust. J. Chem.* 1978, 31, 79.

(21) Taylor, E.; Anderson, J.; Berchtold, G. *J. Amer. Chem. Soc.* 1955, 77, 5444.

(22) Barrett, G. C.; Khokhar, A. R. *J. Chem. Soc.* 1969, 8, 1117.

Inhibitory potencies against related enzymes were determined in assays similar to that described above,<sup>23</sup> 15-lipoxygenase from soybean (Sigma) or cyclooxygenase from sheep seminal vesicles in place of the RBL-1 preparation.

**Rat PMNL 5-Lipoxygenase Assay.** Rat PMNL were obtained from the pleural cavity of nonfasted male rats (Sprague-Dawley, 200-250 g) injected intrapleurally with 200  $\mu$ L of 0.05% (w/v) carrageenan. Cells were pooled and centrifuged for 15 min at 500g. The cell pellet was resuspended in PBS-H, washed by centrifugation, and then briefly resuspended in water for 25 s to lyse contaminating erythrocytes. The cells were washed two more times with PBS-H and suspended at a concentration of  $2 \times 10^7$  cells/mL in 20 mM *N*-(2-hydroxyethyl)piperazine-*N'*-2-ethanesulfonic acid (Sigma Chemical Co., St. Louis, MO) in Earle's balanced salts (GIBCO Laboratories, Grand Island, NY), pH 7.0, containing 1 mg/mL BSA (EH-BSA). About  $7.5 \times 10^7$  cells were recovered per rat and greater than 90% of the cells were polymorphonuclear leukocytes (PMNL) as determined by differential counting using Wright's stain. Test compounds or DMSO vehicle (final concentration 2%) were preincubated with cell suspensions ( $5 \times 10^6$  cells/250  $\mu$ L) for 15 min at 37  $^{\circ}$ C. Cellular arachidonate metabolism was initiated by adding calcium ionophore A23187 (final concentration 8.3  $\mu$ M) and terminated after 10 min by rapid cooling in an ice bath. Cell supernatants were extracted with two volumes of methanol and analyzed for LTB<sub>4</sub> content by HPLC.

**Human Whole Blood 5-Lipoxygenase Assay.** Heparinized (20 units/mL) human blood (0.7 mL) was preincubated with test compounds or vehicle for 15 min at 37  $^{\circ}$ C. Eicosanoid biosynthesis was initiated by adding calcium ionophore A23187 in DMSO (final concentration 50  $\mu$ M) and terminated after 30 min by rapid cooling

of the blood in an ice bath and centrifuging at 3  $^{\circ}$ C for 10 min at 2500g. The plasma was mixed with 4 volumes of methanol and allowed to stand for at least 2 h at 3  $^{\circ}$ C prior to centrifuging at 1800g for 30 min. The level of LTB<sub>4</sub> in aliquots of the methanol-plasma extract was analyzed by RIA (Amersham Corp., Arlington Heights, IL) or by EIA (Cayman Chemical Co., Ann Arbor, MI).

**Acknowledgment.** We wish to thank E. Otis, D. Bornemeier, and T. Clark for technical assistance.

**Registry No.** 1, 70547-26-1; 5, 131786-48-6; 7, 59484-42-3; 10, 131786-51-1; 11, 131786-50-0; 12, 131786-47-5; 13, 70547-50-1; 14, 39785-53-0; 15, 131786-49-7; 16, 133833-90-6; 17, 133833-91-7; 18, 133833-92-8; 19, 133833-93-9; 20, 133833-94-0; 21, 133833-95-1; 22, 133850-30-3; 23, 131786-53-3; 24, 131786-54-4; 25, 131786-55-5; 26, 131786-76-0; 27, 131786-64-6; 28, 131786-67-9; 29, 131786-61-3; 30, 131786-57-7; 31, 131786-75-9; 32, 131786-62-4; 33, 131786-56-6; 34, 133833-96-2; 35, 131786-60-2; 36, 131786-66-8; 37, 131786-58-8; 38, 133833-97-3; 39, 131786-63-5; 40, 131786-59-9; 41, 131786-68-0; 42, 131786-69-1; 43, 77077-60-2; 44, 133833-98-4; 45, 133833-99-5; 46, 65752-44-5; 47, 131786-91-9; 48, 131786-89-5; 49, 131786-90-8; 50, 133834-00-1; 51, 131786-77-1; 52, 131786-97-5; 53, 131786-93-1; 54, 131786-95-3; 55, 131786-96-4; 56, 131786-92-0; 57, 131786-94-2; 58, 133834-01-2; 59, 133834-02-3; 60, 131786-78-2; 61, 131787-05-8; 62, 131787-04-7; 63, 131786-84-0; 64, 131787-03-6; 65, 104223-98-5; 66, 131786-79-3; 67, 133834-03-4; 68, 131786-80-6; 69, 133011-84-4; 70, 70547-29-4; 71, 55073-97-7; 72, 131786-86-2; 73, 131786-85-1; 74, 131786-74-8; 75, 131786-83-9; 76, 131786-82-8; 77, 131786-81-7; thiolactic acid, 79-42-5; 3-cyanopyridine, 100-54-9; ethyl cyanofornate, 623-49-4; ethyl  $\alpha$ -bromopropionate, 535-11-5; thio-benzamide, 2227-79-4; methyl isocyanate, 624-83-9; ethyl chloroformate, 541-41-3;  $\alpha$ -cyanobenzyl benzenesulfonate, 22259-85-4; methyl thionobenzoate, 5873-86-9; phenyl glycine, 69-91-0; 5-lipoxygenase, 80619-02-9.

(23) Aharony, D.; Smith, J. B.; Silver, M. J. *Prostaglandins Med.* 1981, 6, 237.

## Synthesis of an (Iodovinyl)misonidazole Derivative for Hypoxia Imaging

Joseph E. Biskupiak,\* John R. Grierson, Janet S. Rasey, Gary V. Martin, and Kenneth A. Krohn

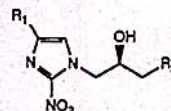
Departments of Radiology and Radiation Oncology, Imaging Research Laboratory, RC-05, University of Washington, Seattle, Washington 98195. Received December 10, 1990

Nitroimidazoles undergo a bioreduction in viable hypoxic tissue, resulting in trapping within these tissues, as demonstrated by misonidazole. A radioiodinated analogue of misonidazole (IVM, (E)-5-(2-Nitroimidazolyl)-4-hydroxy-1-iodopent-1-ene, 3) has been synthesized by halodestannylation, for evaluation as an imaging agent for hypoxia. A key step in the synthetic sequence involves the use of the Lewis acid  $\text{BF}_3 \cdot \text{Et}_2\text{O}$  to promote the nucleophilic ring opening of glycidyl tosylate with (E)-1-lithio-2-(tributylstannyl)ethylene. Direct comparison of IVM versus F-MISO (2) another misonidazole type hypoxic cell marker, in several in vitro cell culture studies, indicates that IVM behaves in analogous fashion to F-MISO and has promise as a hypoxia imaging agent for SPECT.

Our interest in misonidazole (MISO, 1) and its derivatives is due to the ability of viable hypoxic tissues, as might be found in ischemic heart and brain and the central regions of tumors, to metabolically trap these compounds following a bioreduction reaction.<sup>1,2</sup> This property was the impetus for the synthesis of  $^{18}\text{F}$ -labeled fluoromisonidazole (F-MISO, 2) for in vivo nuclear imaging of hypoxic tissues by positron emission tomography (PET).<sup>3-5</sup> We now report the synthesis of an (iodovinyl)misonidazole derivative (IVM, 3) which has been labeled with  $^{131}\text{I}$  and is being evaluated biologically as a hypoxia imaging agent.

Table I. Partition Coefficients

|               | R <sub>1</sub> | R <sub>2</sub>   | partition coefficient |
|---------------|----------------|------------------|-----------------------|
| MISO (1)      | H              | OCH <sub>3</sub> | 0.41                  |
| F-MISO (2)    | H              | F                | 0.40                  |
| IVM (3)       | H              | CH=CHI           | 2.52                  |
| 4-Br-MISO (4) | Br             | OCH <sub>3</sub> | 2.87                  |



The success of these efforts will directly lead to radio-labeling with  $^{123}\text{I}$ , useful with more common nuclear imaging instrumentation, including single photon emission computed tomography (SPECT).

### Chemistry Results and Discussion

In addition to the synthesis of F-MISO, other work in our laboratory involved the synthesis of [ $^{82}\text{Br}$ ]-4-Br-MISO

- (1) Chapman, J. D. *N. Eng. J. Med.* 1979, 301, 1429.
- (2) Rasey, J. S.; Grunbaum, Z.; Magee, S.; Nelson, N. J.; Olive, P. L.; Durand, R. E.; Krohn, K. A. *Radiat. Res.* 1987, 111, 292.
- (3) Grierson, J. R.; Link, J. M.; Mathis, C. A.; Rasey, J. S.; Krohn, K. A. *J. Nucl. Med.* 1989, 30, 343.
- (4) Koh, W.; Rasey, J. S.; Krohn, K. A. Radiation Research Society 37th Annual Meeting, March 18-23, 1989, Seattle, WA.
- (5) Koh, W.; Rasey, J. S.; Grierson, J. R.; Link, J. M.; Woods, S. D.; Graham, M. M.; Lewellen, T. K.; Shields, A. F.; Modell, H. I.; Krohn, K. A. *J. Nucl. Med.* 1989, 30, 789.