We ELI LILLY AND COMPANY

of 307 East McCarty Street, City of Indianapolis, State of Indiana, United States of America

hereby apply for the grant of a Patent for an invention entitled "7-(ALPHA-FUROYLURIDIOARYL AND CYCLOHEXAKIENYLACETAMIDO) CEPHALOSPORIN ANTI-BIOTICS"

which is described in the accompanying complete specification. This application is a Convention Application and is based on the application numbered 456,491 for a patent or similar protection made in United States of America on 1st April, 1974

My address for service is:

Care: SPRUSON & FERGUSON PATENT ATTORNEYS ESSO HOUSE, 1, KENT STREET SYDNEY, NEW SOUTH WALES, AUSTRALIA.

Dated this ELEVENTH day of MARCH, 1975.

ELI LILLY AND COMPANY

By: __________________________

Signature of Applicant

Registered Patent Attorney
COMMONWEALTH OF AUSTRALIA
PATENTS ACT 1952-1966

DECLARATION IN SUPPORT OF CONVENTION APPLICATION
FOR A PATENT OR PATENT OF ADDITION

(The declaration shall be made by the applicant, or, if the applicant is a body corporate, by a person authorized by the body corporate to make the declaration on its behalf.)

In support of the Convention Application made for a patent or patent of addition for an invention entitled

"7-(ALPHA-4-FUROYLUREIDOARYL AND CYCLOHEXAKI ENYLACETAMIDO) CEPHALOSPORIN ANTIBIOTICS"

I declare as follows:-

1. The applicant is

Everet Foy Smith
3026 Glenview Drive
Indianapolis, Indiana
United States of America

and the actual inventor is

ROBIN DAVID GREY COOPER
6740 Dover Road, Indianapolis,
Indiana 46220, United States of America

2. The basic application as defined by Section 141 of the Act was made in the United States of America on the 01 April 1974 by Robin David Grey Cooper.

3. (a) I am the applicant for the patent or patent of addition.

(b) I am authorized by Eli Lilly and Company, a corporation of the State of Indiana, United States of America, to make this declaration on its behalf.

4. The basic application referred to in paragraph 2 of this Declaration was the first application made in a Convention country in respect of the invention the subject of the application.

Declared at Indiana this 10th day of February 1975

ELI LILLY AND COMPANY

BY:

Assistant Secretary
Name of Applicant: ELI LILLY AND COMPANY

Address of Applicant: 307 East McCarty Street, City of Indianapolis, State of Indiana, United States of America

Actual Inventor: ROBIN DAVID GREY COOPER


Complete Specification for the invention entitled:

"7-(ALPHA-FUROYLUREIDOARYL AND CYCLOHEXAKIENYLACETAMIDO) CEPHALOSPORIN ANTIBIOTICS"

The following statement is a full description of this invention, including the best method of performing it known to us:
The present invention relates to novel ureido substituted cephalosporin compounds which have a broad antibiotic spectrum against both the gram-positive and gram-negative microorganisms.

Several antibiotics of the cephalosporin class have achieved an important status in the treatment and control of infectious diseases of man. For example, the well known cephalosporin antibiotics, cephalothin, cephaloglycin, cephaloridine, and cephalexin have been widely used in the treatment of infections in man. Considerable effort continues to be extended in the development of new cephalosporin antibiotics with increased antibiotic activity and particularly with an expanded spectrum of activity against the gram-negative microorganisms.

Cephalosporin compounds having a substituted α-amino group in the 7-arylacetamido side chain have been previously described. For example, U.S. Patent No. 3,646,024 describes certain 7-[α-(3-imidoylureido)arylacetamido]cephalosporanic acids. α-Ureido phenylacetamidocephalosporanic acids have been described in U.S. Patent No. 3,673,183.

The compounds provided by the process of this invention differ structurally from the compounds of the prior art in that the cephalosporin dihydrothiazine ring is substituted in the 3-position with a heterocyclic-thiomethyl group. In addition, the cephalosporin antibiotics described herein can be characterized as expanded spectrum cephalosporin antibiotics in that they not only possess the usual high level of activity against gram-positive microorganisms but they also possess a high level of activity.
against a broad spectrum of gram-negative microorganisms which
the prior art compounds did not possess.

This invention relates to new cephalosporin anti-
biotics represented by the following general formula

\[
\begin{align*}
&\text{O} \quad \text{OH} \quad \text{OH} \\
\text{C-N-C-N-CH-C-N} \\
&\text{R'} \quad \text{R} \\
&\text{CH}_2-S-R_1 \\
&\text{COOR}_2 \\
\end{align*}
\]

wherein Z is O or S

R' is hydrogen or methyl;

R is phenyl, methylphenyl, hydroxyphenyl, halophenyl, hydroxy substituted halophenyl, thiényl, furyl or 1,4-cyclohexadienyl;

R_1 is

\[
\begin{align*}
&\text{R}_3 \\
&\text{N-N} \\
&\text{N-N} \\
&\text{R}_3 \\
\end{align*}
\]

or

\[
\begin{align*}
&\text{R}_3 \\
&\text{N} \\
&\text{S} \\
&\text{R}_3 \\
\end{align*}
\]

wherein R_3 is C_1-C_4 lower alkyl; R_2 is hydrogen, an indanyl group, a phthalidyl group, or an acyloxymethyl group of the

formula

\[
\begin{align*}
&\text{O} \\
&\text{CH}_2-O-C-Y
\end{align*}
\]
wherein Y is C\textsubscript{1}-C\textsubscript{4} alkyl or phenyl;
and when R\textsubscript{2} is hydrogen, the pharmaceutically acceptable salts thereof.

In the above formula I, the term "methylphenyl" refers to the mono and dimethylphenyl groups such as 4-methylphenyl, 3-methylphenyl, 3,4-dimethylphenyl, or 3,5-dimethylphenyl. "Hydroxyphenyl" refers to the 3- and 4-monohydroxyphenyl groups, and to the 3,4-dihydroxy- and 2,4-dihydroxyphenyl groups. "Halophenyl" refers to the fluoro, chloro, and bromophenyl groups such as 4-chlorophenyl, 3-chlorophenyl, 3,4-dichlorophenyl, 4-bromophenyl, or 4-fluorophenyl. "Hydroxy substituted halophenyl" refers to 3-chloro-4-hydroxyphenyl, 3,5-dichloro-4-
hydroxyphenyl, or 3,5-dibromo-4-hydroxyphenyl. "Thienyl" and "furyl" refer to the respective 2- and 3- isomers thereof.

As described above, the heterocyclic radical in the 3-position of the cephem ring is substituted with a C₁-C₄ lower alkyl group. Representative of these groups are the 1-methyl-1H-tetrazole-5-yl group, the 1-ethyl-1H-tetrazole-5-yl group, the 5-methyl-1,3,4-thiadiazol-2-yl group, the 5-isopropyl-1,3,4-thiadiazol-5-yl group, and like lower alkyl substituted tetrazole and thiadiazole groups.

The compounds of formula I wherein R' is hydrogen are prepared by reacting a 7-phenylglycylamido, a substituted phenylglycylamido or a 1,4-cyclohexadienylglycylamido 3-tetrazolethiomethyl or thiadiazolethiomethyl substituted cephalosporin of the following formula II with furoyl or thenoyl isocyanate as illustrated by the following reaction scheme:

![Reaction scheme]

wherein R, R₁ and Z are as previously defined.

The compounds represented when R' is methyl are prepared by acylating the compound of the formula II with N-(α-furoyl)-N-methylcarbamoyl chloride or N-(α-thenoyl)-N-
methylcarbamoyl chloride represented by the formula

\[
\begin{array}{c}
\text{O} \\
\text{C-N=C=O} \\
\text{CH}_3
\end{array}
\]

The carbamoyl chloride is prepared by reacting N-methyl-2-furamide or N-methylthiophene-2-carboxamide with n-butyllithium at -78°C. to generate the lithium salt followed by the reaction of the lithium salt with phosgene. The reaction is carried out in the cold (-78°C.) in an inert solvent such as tetrahydrofuran.

The acylation of the glycylamido cephalosporin of formula II with the carbamoyl chloride is carried out in an inert solvent at a temperature between about -15 and 10°C. in the presence of a hydrogen halide acceptor.

Inert solvents such as acetonitrile and tetrahydrofuran can be used conveniently. Hydrogen halide acceptors such as the tertiary amines, triethylamine, and pyridine; and the alkylene oxides such as propylene oxide and butylene oxide can be used. Equimolar amounts of the starting material and the carbamoyl chloride are used. In an example of the preparation of a compound of formula I wherein R' is methyl, 7-(D-phenylglycylamido)-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid is suspended in dry tetrahydrofuran and solubilized by adding bis-(trimethylsilyl)acetamide to the suspension. The solution is cooled to about 0°C. and an equimolar amount of N-(α-furoyl)-N-methylcarbamoyl chloride in tetrahydrofuran is added. The mixture is stirred in the cold for about 2 hours, is allowed to warm to room temperature, and
the product extracted with an organic solvent such as ethyl acetate.

The 7-thienylglycylamido, 7-furylglycylamido, 7-phenylglycylamido and 7-substituted phenylglycylamido-3-heterocyclic thiomethyl cephalosporin starting materials of the formula II are prepared by acylating a 7-amino-3-heterocyclic-thiomethyl-substituted cephalosporin nucleus compound with an active derivative of phenylglycine or a substituted phenylglycine, for example, the acid chloride, in the presence of a hydrogen halide acceptor such as triethylamine or sodium carbonate, to provide the acylated phenylglycylamido cephalosporin starting material.

The compounds of the formula II wherein R is the 1,4-cyclohexadienyl-1-yl group are prepared by acylation of the 7-amino-3-(1-lower alkyl-1H-tetrazole-5-ylthiomethyl) or (5-lower alkyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid with an active derivative of α-amino-α-(1,4-cyclohexadienyl) acetic acid. The cyclohexadienyl acetic acid is converted to an active derivative such as that formed with chloroethyl formate for use as the acylating reagent.

The reaction of the starting material of the formula II with furoyl or thenoyl isocyanate is carried out in the following manner. The starting material of the formula II is suspended in an inert solvent at about 20-25°C. and a silylating agent such as bis-(trimethylsilyl)acetamide (BSA) or monosilylacetamide (MSA) is added in excess to form a homogenous solution. Inert solvents such as tetrahydrofuran, dichloromethane, chloroform, or dioxane can be used. After obtaining a solution of the silylated derivative of the starting material, the reaction mixture is cooled in a dry ice-acetone bath to
a temperature of approximately -75 to -80°C. To the cold solution is added, in excess, the isocyanate. The reaction mixture is then allowed to stir in the cold for about 3 hours and is thereafter allowed to warm to room temperature. Methanol is added to the reaction mixture to decompose excess silylating agent and the mixture is then evaporated under reduced pressure to remove the volatile solvents. The ureido reaction product is then extracted from the residue with ethyl acetate. The product is purified with an acid-base wash and can be further purified by recrystallization.

By way of illustration of the above preparation methods, 7-amino-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid is acylated with phenylglycyl chloride hydrochloride in the presence of sodium carbonate to yield 7-phenylglycylamido-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid. The above acylation product is then reacted with furoylisocyanate following the solubilization of the phenylglycylamido acylation product in tetrahydrofuran with MSA, to provide a compound of the invention wherein R is phenyl, R₁ is the 1-methyl-1H-tetrazole substituent, R' is H and R₂ is hydrogen.

The compounds represented by formula I wherein R₂ is an acyloxymethyl group are prepared by reacting a salt, for example, an alkali metal salt of the free acid compound of formula I with a lower alkanoyloxymethyl halide or with a halomethyl benzoate. Lower alkanoyloxymethyl halides which can be employed are, for example, chloromethyl acetate, chloromethyl propionate, bromomethyl acetate, bromomethyl butyrate, chloromethyl pivaloate, and like halomethyl esters of the lower alkyl straight and branched chain C₁-C₄ alkyl carboxylic acids.
When Y is phenyl, bromo or chloromethylbenzoate can be used in like manner to prepare the benzoyloxymethyl ester. The reaction is carried out by reacting the salt of a cephalosporin acid of the formula I, for example, the sodium or potassium salt with the halomethyl ester in an inert solvent from about 20°C to about 55°C. Inert solvents which can be employed include, for example, dimethylformamide (DMF), dimethylacetamide (DMAC), tetrahydrofuran, and dioxane. For example, sodium 7-[[α-(3-furoyl-1-ureido)phenylacetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephe-4-carboxylate is reacted in aqueous DMF with chloromethyl acetate to provide the acetoxymethyl ester of the cephalosporin acid.

The 5-indanyl esters of the formula I are prepared by esterifying the cephalosporin acid with the phenolic 5-indanol. The indanyl esters can be prepared by the conventional procedures used for preparing phenolic esters of carboxylic acids. For example, an active derivative of the cephalosporin acid such as is formed with ethylchloroformate is reacted with 5-indanol.

The phthalidyl esters of the formula I are prepared by reacting bromophthalide with a salt of the cephalosporin acid, for example, the sodium or potassium salt. Bromophthalide of the formula

\[
\begin{array}{c}
\text{Br} \\
\text{O} \\
\text{O} \\
\end{array}
\]

is prepared in known manner by the reaction of phthalide with N-bromosuccinimide.
The acyloxyethyl esters of the formula I are orally effective forms of the antibiotic acids.

Pharmacetically acceptable salts of the compounds represented by formula I are prepared by methods commonly practiced in the cephalosporin art. Representative pharmaceutically acceptable salts include the alkali metal salts, for example, the sodium, potassium, and lithium salts, the calcium salt, the ammonium salt, the lower aliphatic ammonium salts, for example, those salts formed with methylamine, dimethylamine, diethylamine, or di-n-propylamine; and the hydroxyalkyl ammonium salts, for example, those formed with ethanolamine or diethanolamine. Preferred pharmaceutically acceptable salts include the alkali metal salts, for example, the sodium salt and the potassium salt. The pharmaceutically acceptable salts of the compounds of formula I are prepared by methods well known in the cephalosporin art. For example, the free acid form of the antibiotic is neutralized with an alkali metal hydroxide or carbonate or with ammonium hydroxide or with the desired alkylamine or ethanolamine to form the salt.

The compounds represented by formula I are illustrated by the following compounds.

7-[α-(3-α-furoyl-1-ureido)-α-phenylacetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-1-ureido)-α-(4-hydroxyphenyl)-acetamido]-3-(5-methyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-1-ureido)-α-(α-thienyl)acetamido]-3-(1-ethyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,
7-[α-(3-α-furoyl-l-ureido)-α-(α-furyl)acetamido]-3-(5-isopropyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-l-ureido)-α-(3-chloro-4-hydroxyphenyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-l-ureido)-α-(3-hydroxyphenyl)acetamido]-3-(5-methyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-l-ureido)-α-(1,4-cyclohexadien-1-yl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-l-ureido)-α-(4-chlorophenyl)acetamido]-3-(5-methyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-l-ureido)-α-(4-methylphenyl)acetamido]-3-(5-ethyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-l-ureido)-α-(3,5-dichloro-4-hydroxyphenyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-l-ureido)-α-(3-bromophenyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-thenoyl-l-ureido)-α-phenylacetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-thenoyl-l-ureido)-α-(4-hydroxyphenyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,
7-[α-(3-α-thienoyl-1-ureido)-α-(α-thienyl)acetamido]-3-(5-methyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-thienoyl-3-methyl-1-ureido)-α-phenylacetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-β-thienoyl-1-ureido)-α-phenylacetamido]-3-(5-methyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-3-methyl-1-ureido)-α-phenylacetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-3-methyl-1-ureido)-α-(α-thienyl)acetamido]-3-(5-methyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-3-methyl-1-ureido)-α-(3-chloro-4-hydroxyphenyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-3-methyl-1-ureido)-α-(4-hydroxyphenyl)acetamido]-3-(5-methyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid, and the pharmaceutically acceptable non-toxic salts thereof.

The compounds represented by formula I and the pharmaceutically acceptable salts thereof inhibit the growth of microorganisms pathogenic to animals and man. In particular, these compounds inhibit the growth of a broad spectrum of gram-negative and gram-positive microorganisms. They are further active in inhibiting the growth of penicillin resistant Staphylococcus organisms. Accordingly, the compounds of the invention are useful in combating infections in animals and man.
man attributable to gram-positive and gram-negative microorganisms. The furoyl and thenoylureido-cephalosporins are effective against gram-negative microorganisms of the indole-positive and indole-negative Proteus sp., the Aerobacter sp., the Pseudomonas, the Enterobacter sp., the Serratia, e.g., S. marcescens, Escherichia coli, and Klebsiella. They are also highly effective against the Streptococcus D group of bacteria as well as Staphylococcus aureus and penicillin resistant strains of Staphylococcus.

The compounds of formula I can be administered by the parenteral route, for example, intramuscularly or intravenously. When administered in non-toxic doses ranging between about 25 and about 1,000 mg. per kg. of the patient's body weight, the compounds are effective in the treatment of infectious diseases attributable to both the gram-positive and gram-negative microorganisms. The compounds of this invention can be formulated for such administrative routes as aqueous suspensions or solutions suitable for injection. For example, the compounds of the invention, as the alkali metal salts, can be employed in sterile aqueous solutions for injection or they can be prepared as sterile suspensions in an inert pharmaceutical carrier suitable for injection. When administered intravenously, the salt form of the compound of the invention, for example, the sodium salt, can be dissolved in one of the standard clinical I.V. solutions, for example, I.V. dextrose, for administration via I.V. drip.

Preferred compounds are those represented by formula I wherein R is phenyl, hydroxyphenyl, halophenyl, hydroxy substituted halophenyl, or thiienyl.
An especially preferred group of compounds are those represented when R is phenyl, hydroxyphenyl, or hydroxy substituted halophenyl especially hydroxy substituted chlorophenyl, and the pharmaceutically acceptable non-toxic salts thereof. The preferred compounds described above are illustrated by:

7-[α-(3-α-furoyl-1-ureido)-α-phenylacetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-1-ureido)-α-(4-hydroxyphenyl)-acetamido]-3-(5-methyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-1-ureido)-α-(3-hydroxyphenyl)-acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-1-ureido)-α-(3-chloro-4-hydroxyphenyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-1-ureido)-α-(3,5-dichloro-4-hydroxyphenyl)acetamido]-3-(5-methyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid,

7-[α-(3-α-furoyl-3-methyl-1-ureido)-α-(4-hydroxyphenyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid, and the pharmaceutically acceptable non-toxic salts thereof.

The antibiotic activity of the compounds of formula I is illustrated by the in vitro data presented in the following Tables I and II for two of the preferred compounds. In the tables, the minimum inhibitory concentrations (MIC) of the test compounds versus the indicated gram-positive and gram-negative microorganisms is presented. The MIC values were determined by
the gradient plate method which is essentially the method described by Bryson and Szybalski, *Science*, 116, 45-46 (1952).

Table I lists the in vitro antibiotic activity demonstrated by the test compounds against representative gram-negative microorganisms. Table II lists the inhibitory activity in terms of MIC values against clinical isolates of penicillin resistant *Staphylococcus* microorganisms both in the presence of and in the absence of serum.

**TABLE I**

Antibiotic Activity of 7-[[α-(3-Hydroxyphenyl)acetamido]phenyl-(3-furoyl-l-ureido)phenyl]-3-cephalosporins vs. Gram-

<table>
<thead>
<tr>
<th>Test Organism</th>
<th>Test Compound A</th>
<th>Test Compound B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Shigella sp.</td>
<td>5.5</td>
<td>5.5</td>
</tr>
<tr>
<td><em>Escherichia coli</em></td>
<td>7.0</td>
<td>5.8</td>
</tr>
<tr>
<td><em>Klebsiella pneumoniae</em></td>
<td>5.0</td>
<td>6.3</td>
</tr>
<tr>
<td><em>Aerobacter aerogenes</em></td>
<td>7.5</td>
<td>6.5</td>
</tr>
<tr>
<td><em>Salmonella heidelberg</em></td>
<td>6.8</td>
<td>5.8</td>
</tr>
<tr>
<td><em>Pseudomonas aeruginosa</em></td>
<td>12.3</td>
<td>10.7</td>
</tr>
<tr>
<td><em>Serratia marcescens</em></td>
<td>19.5</td>
<td>14.5</td>
</tr>
</tbody>
</table>

1 Test Compound A=7-[[α-(3-α-furoyl-l-ureido)-α-phenylacetamido]-3-(1-methyl-1H-tetrazole-5-ythiomethyl)-3-cephem-4-carboxylic acid.

2 Test Compound B=7-[[α-(3-α-furoyl-l-ureido)-α-(4-hydroxyphenyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ythiomethyl)-3-cephem-4-carboxylic acid.
### TABLE II

Antibiotic Activity of 7-\{a-(3-Furoyl-l-ureido)-(hydroxyphenyl)acetamidocephalosporins vs.

<table>
<thead>
<tr>
<th>Resistant Staph.</th>
<th>Test Compound</th>
<th>MIC (mcg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>A</td>
<td>B</td>
</tr>
<tr>
<td>NS²</td>
<td>S³</td>
<td>NS</td>
</tr>
<tr>
<td>V-41</td>
<td>3.0</td>
<td>8.0</td>
</tr>
<tr>
<td></td>
<td>5.0</td>
<td>&gt;20</td>
</tr>
<tr>
<td>V-32</td>
<td>4.5</td>
<td>8.0</td>
</tr>
<tr>
<td></td>
<td>13.7</td>
<td>&gt;20</td>
</tr>
<tr>
<td>X-400</td>
<td>&gt;20</td>
<td>&gt;20</td>
</tr>
<tr>
<td>V-84</td>
<td>0.4</td>
<td>1.0</td>
</tr>
<tr>
<td></td>
<td>1.0</td>
<td>1.0</td>
</tr>
<tr>
<td>X1.1</td>
<td>0.4</td>
<td>1.0</td>
</tr>
<tr>
<td></td>
<td>1.0</td>
<td>1.0</td>
</tr>
</tbody>
</table>

1 Test compounds A & B are respectively the test compounds of Table I.

2 Compound tested in the absence of serum.

3 Compound tested in the presence of serum.

As indicated by the in vitro data presented above for two of the preferred compounds, the furoylureido cephalosporin compounds disclosed herein are resistant to the action of the enzymes, penicillinase and cephalosporinase, generated respectively by the penicillin-resistant Staphylococci and the gram-negative organisms.

The preparation of the compounds represented by formula I is further illustrated by the following examples.

Example 1

To a suspension of 0.6693 g. of 7-phenylglycyl-amido-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid in 100 ml. of dry tetrahydrofuran was added an excess of monosilyl acetamide to form a solution. Linde 4A molecular sieve was added and the mixture was cooled in...
dry ice-acetone bath. An excess of furoyl isocyanate was added to the cold mixture with stirring. Stirring was continued in the cold for 2 hours and then the mixture was allowed to warm to room temperature. Fifty milliliters of methanol were added and the reaction mixture was filtered. The filtrate was evaporated under reduced pressure to remove volatile solvents. The residue was dissolved in aqueous sodium bicarbonate and the solution was extracted with ethyl acetate. The aqueous phase was acidified to about pH 1.5-2.0 with dilute hydrochloric acid and was extracted with ethyl acetate. The extract was concentrated and was then diluted with about an equal volume of petroleum ether to precipitate the product, 7-[α-(3-α-furoyl-1-ureido)-α-phenylacetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid.

The nuclear magnetic resonance spectrum of the product run in deuterated dimethyl sulfoxide was in agreement showing peaks at δ5.10 and δ5.70 for the C\textsubscript{6} and C\textsubscript{7} β-lactam protons; multiplets at δ4.32 and δ3.65 for the methylene protons and a singlet at δ4.00 for the N-methyl protons of the tetrazole group.

Example 2

To a suspension of 0.955 g. of 7-(4-hydroxyphenylglycylamido)-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid in 100 ml. of tetrahydrofuran was added excess monosilylacetamide. To the resultant solution was added molecular sieve (Linde 4A molecular sieve) and the mixture cooled in a dry-ice acetone bath. A slight excess of furoyl isocyanate was then added with stirring. The reaction mixture was stirred in the cold for 3 hours.
and was then allowed to warm to room temperature. Methanol, 100 ml., was added and the mixture was then evaporated under reduced pressure to remove volatile solvents. The residue was dissolved in an aqueous solution of sodium bicarbonate and the solution was washed with ethyl acetate. The solution was then acidified with dilute hydrochloric acid to about pH 2 and extracted with ethyl acetate. The extract was concentrated and the concentrate diluted with petroleum ether to precipitate the reaction product, 7-[(α-(3-furoyl-1-ureido)-4-hydroxyphenylacetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)]-3-cephem-4-carboxylic acid. The product was further purified by crystallization from methanol-diethyl ether-pentane (Skelly-solve-B).

Example 3

Following the procedures of Example 1, furoyl isocyanate is reacted with 7-phenylglycylamido-3-(5-methyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid to obtain 7-[(α-(3-furoyl-1-ureido)-α-phenylacetamido]-3-(5-methyl-1,3,4-thiadiazole-2-ylthiomethyl)-3-cephem-4-carboxylic acid.

Example 4

7-[(α-(3-furoyl-1-ureido)-α-(3-chloro-4-hydroxyphenyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid is prepared by the reaction of furoyl isocyanate with 7-(3-chloro-4-hydroxyphenylglycylamido)-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid.

Example 5

To a suspension of 461 mg 7-phenylglycylamido-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid X-4283A...
acid in 8 ml. of dry acetonitrile containing 2 ml. of propylene oxide was added with stirring 1 ml. of bis-(trimethylsilyl)acetamide (BSA). The resulting orange solution was cooled to 0°C. and a solution of a slight molar excess of N-(2-furoyl)-N-methylcarbamoyl chloride in 2 ml. of dry acetonitrile was added. The reaction mixture was stirred for 2 hours in the cold and was then allowed to warm to room temperature.

The reaction mixture was filtered and the methanol was added to the filtrate to destroy any excess BSA present. The filtrate was evaporated and the residue was dissolved in a mixture of ethyl acetate and water. The pH of the mixture was adjusted to 2 and the organic layer separated. The organic layer was washed with water, dried, and evaporated to yield the reaction product, 7-[α-(3-α-furoyl-3-methyl-1-ureido)-α-phenylacetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid. The product was crystallized from acetone-ether to yield 156 mg. of purified product.

Example 6

To a solution of 511 mg. of 7-(3-chloro-4-hydroxyphenylglycylamido)-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid in 1 ml. of tetrahydrofuran and 1 ml. of bis-(trimethylsilyl)acetamide was added a solution of 2 ml. of propylene oxide in 8 ml. of acetonitrile. The resulting solution was cooled to 0°C. and a solution of 2 ml. of N-(2-furoyl)-N-methylcarbamoyl chloride in 2 ml. of acetonitrile was added. The dark reaction mixture was stirred at 0°C. for 2 hours and was then allowed to warm to room temperature. The mixture was filtered and 3 ml. of methanol were added to the filtrate to destroy any excess
silyl reagent. The filtrate was evaporated and the residue dissolved in a mixture of ethyl acetate and water. With stirring the pH of the mixture was adjusted to 2 with dilute hydrochloric acid. The organic phase was separated and was washed with water, dried, and evaporated under reduced pressure. The reaction product residue, 7-[α-(3-α-furoyl-3-methyl-1-ureido)-α-(3-chloro-4-hydroxyphenyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid was crystallized from a mixture of ethyl acetate-diethyl ether-petroleum ether to yield 74 mg. of crystalline product.

Example 7

7-[α-(3-α-furoyl-1-ureido)-α-(α-thienyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid.

To a suspension of 467 mg of 7-[α-(α-thienyl)-α-amino-acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid in 100 ml of dry THF were added 2 g of mono-(trimethylsilyl)acetamide (MSA). When solution had occurred a small amount of molecular sieve was added to maintain dryness and the solution was cooled to 0°C. To the cold solution was added a solution of an excess of furoyl isocyanate in 2 ml of THF. The reaction mixture was stirred at 0°C. for 3 hours and was then allowed to warm to room temperature. The reaction mixture was filtered and 5 ml of methanol were added to the filtrate. The filtrate was evaporated and the residue was layered with ethyl acetate and water. The pH of the aqueous phase was adjusted to 2 with dilute hydrochloric acid and the organic layer was separated. The organic layer was treated with a dilute solution of sodium bicarbonate to pH 7.2. The aqueous layer
was separated and acidified to pH 2 with dilute hydrochloric acid at ice bath temperature. The acidified solution was extracted with ethyl acetate. The extract was dried and evaporated and the residue recrystallized from a mixture of acetone-diethyl ether-petroleum ether to yield a first crop of product weighing 45 mg, a second crop weighing 83 mg and an additional 24 mg of product from the filtrate.

**Example 8**

7-[α-(3-α-furoyl-3-methyl-1-ureido)-α-(1,4-cyclohexadienyl)-acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid.

By following the procedures described in Example 5, 463 mg of impure 7-[α-(1,4-cyclohexadienyl)-α-amino-acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid was dissolved in 15 ml of methylene chloride containing excess bis-(trimethylsilyl)acetamido (BSA), propylene oxide was added to the solution followed by a slight molar excess of N-(2-furoyl)-N-methylcarbamoyl chloride. After stirring for 2 hours in the cold 74 mg of the product was recovered as an amorphous powder.

**Example 9**

7-[α-(3-α-thenoyl-1-ureido)-α-(α-thienyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid.

To a solution of 467 mg of 7-(α-amino-α-thienyl-acetamido)-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid in 100 ml of dry THF, formed by the addition of 1 ml of BSA, was added at 0°C. a solution containing a molar excess of α-thienylisocyanate in 2 ml of dry THF. The reaction mixture was stirred for 3 hours
at 0°C. and was then allowed to warm to room temperature. The product was recovered from the reaction mixture by following the isolation procedures described in Example 7. The product was recrystallized from a mixture of acetone-diethyl ether-petroleum ether to yield 156 mg.

Example 10

7-[(α-(3-α-furoyl-3-methyl-1-ureido)-α-(2-thienyl)acetamido)-
3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic
acid.

A suspension of 234 mg of 7-[(α-amino-α-(α-thienyl)-
acetamido)-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-
carboxylic acid in 50 ml of dry acetonitrile was solubilized with 5 ml of BSA and the slightly orange solution was cooled to 0°C. To the cold solution were added 2 ml. of propylene oxide and a stoichiometric excess of N-(α-furoyl)-N-methyl-

A suspension of 234 mg of 7-[(α-amino-α-(α-thienyl)-
acetamido)-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-
carboxylic acid in 50 ml of dry acetonitrile was solubilized with 5 ml of BSA and the slightly orange solution was cooled to 0°C. To the cold solution were added 2 ml. of propylene oxide and a stoichiometric excess of N-(α-furoyl)-N-methyl-

carbamoyl chloride. The reaction mixture was stirred 2 hours at 0°C., one hour at room temperature and methanol was added to decompose the silylating agent. The mixture was evaporated and the residue extracted with ethyl acetate at pH2. The extract was washed with water, dried and evaporated under vacuum. The dried residue was recrystallized from a mixture of acetone-diethylether-petroleum ether to yield 114 mg of the product.
The claims defining the invention are as follows:

What is claimed is:

1. Compounds of the formula I

\[
\begin{array}{c}
\text{Z} \\
\text{R'} \\
\text{R} \\
\text{R}_1 \\
\text{R}_2
\end{array}
\]

wherein Z is O or S;

R' is hydrogen or methyl;

R is phenyl, methylphenyl, hydroxyphenyl, halophenyl, hydroxy substituted halophenyl, thienyl, furyl or 1,4-cyclohexadienyl;

R\textsubscript{1} is

\[
\begin{array}{c}
\text{N} \\
\text{N} \\
\text{N} \\
\text{N} \\
\text{R}_3
\end{array}
\]

or

\[
\begin{array}{c}
\text{S} \\
\text{N} \\
\text{R}_3
\end{array}
\]

wherein R\textsubscript{3} is C\textsubscript{1}-C\textsubscript{4} alkyl;

R\textsubscript{2} is hydrogen, indanyl, phthalidyl, or an acyloxy-methyl group of the formula

\[
\begin{array}{c}
\text{O} \\
\text{CH}_2 \text{O-C-Y}
\end{array}
\]

wherein Y is C\textsubscript{1}-C\textsubscript{4} alkyl or phenyl;

and when R\textsubscript{2} is hydrogen, the pharmaceutically acceptable non-toxic salts thereof.

2. The compound of claim 1, wherein R is phenyl, hydroxyphenyl, halophenyl, hydroxy substituted halophenyl, or thienyl.

3. The compound of claims 1 or 2, wherein R is phenyl, hydroxyphenyl, or hydroxy substituted halophenyl, and R\textsubscript{2} is hydrogen.
4. The compound of any of claims 1-3, wherein R₁ is

![Chemical Structure Image]

5. The compound of any of claims 1-4, said compound being 7-[α-(3-α-furoyl-1-ureido)-α-phenylacetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid.

6. The compound of any of claims 1-4, said compound being 7-[α-(3-α-furoyl-1-ureido)-α-(4-hydroxyphenyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid.

7. The compound of any of claims 1-4, said compound being 7-[α-(3-α-furoyl-1-ureido)-α-(3-chloro-4-hydroxyphenyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid.

8. The compound of any of claims 1-4, said compound being 7-[α-(3-α-furoyl-3-methyl-1-ureido)-α-(3-chloro-4-hydroxyphenyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid.

9. The compound of any of claims 1-4, said compound being 7-[α-(3-α-furoyl-3-methyl-1-ureido)-α-phenylacetamido]-3-(1-methyl-1H-tetrazole-5-ylthiomethyl)-3-cephem-4-carboxylic acid.

10. The compound of any of claims 1-3 or 10, wherein R₁ is

![Chemical Structure Image]

11. The compound of any of claims 1-3 or 10, wherein R' is hydrogen and R is phenyl.
12. The compound of any of claims 1-3 or 10, wherein R' is methyl and R is phenyl.

13. The compound of claim 1 wherein R is thiethyl.

14. The compound of claim 13 said compound being 7-\([\alpha-(3-\alpha-furoyl-1-ureido)-\alpha-(\alpha-thienyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiromethyl)-3-cephem-4-carboxylic acid.

15. The compound of claim 13 said compound being 7-\([\alpha-(3-\alpha-thienyl-1-ureido)-\alpha-(\alpha-thienyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiromethyl)-3-cephem-4-carboxylic acid.

16. The compound of claim 13 said compound being 7-\([\alpha-(3-\alpha-furoyl-3-methyl-1-ureido)-\alpha-(\alpha-thienyl)acetamido]-3-(1-methyl-1H-tetrazole-5-ylthiromethyl)-3-cephem-4-carboxylic acid.

17. A process for preparing compounds of formula I wherein Z, R, R_1, R' and R_2 are as defined in claim 1, which comprises reacting a compound of the formula II

\[
\begin{align*}
\text{OH} & \\
\text{H}_2\text{N-CH-C-N} & \\
\text{R} & \\
\text{O} & \\
\text{COOH} & \\
\end{align*}
\]

with a compound of the formula III

\[
\begin{align*}
\text{O} & \\
\text{Z} & \\
\text{C-A} & \\
\text{O} & \\
\text{H} & \\
\text{CH}_3 & \\
\end{align*}
\]

wherein A is \(-N=\equiv C=0\) or \(-N-C=\equiv C\); and if desired converting the acid so obtained wherein R_2 is hydrogen to the corresponding ester wherein R_2 is other than hydrogen.
18. A process for preparing compounds of the formula I wherein Z, R, R', R₁, and R₂ are as defined in claim 1, substantially as hereinbefore described with particular reference to the examples.

19. Compounds of the formula I wherein Z, R, R', R₁, and R₂ are as defined in claim 1, substantially as hereinbefore described with particular reference to the examples.

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