1,152,544

PATENT SPECIFICATION

NO DRAWINGS

1,152,544

Inventor: MILON WALKER BULLOCK

Date of Application and filing Complete Specification: 17 May, 1967.

No. 22977/67.

Application made in United States of America (No. 554,307) on 1 June, 1966.

Complete Specification Published: 21 May, 1969.

© *Crown Copyright 1969.*

Index at acceptance:—C2 C(3C4, B4E, B4J, B4M)

Int. Cl.:—C 07 d 99/10

COMPLETE SPECIFICATION

Resolution of dl-6-Phenyl-2,3,5,6-Tetrahydroimidazo [2,1-b] Thiazole

We, AMERICAN CYANAMID COMPANY, a corporation and existing under the Laws of the State of Maine, United States of America, of Berdan Avenue, Township of Wayne, State of New Jersey, United States of America, do hereby declare the invention, for which we pray that a patent may be granted to us, and the method by which it is to be performed, to be particularly described in and by the following statement:—

This invention relates to the preparation and isolation of the optically active chemical compounds *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b]thiazole free or substantially free of contaminating dextro-isomer, and *d*(+)6 - phenyl - 2,3,5,6-tetrahydroimidazo[2,1-b]thiazole free or substantially free of *l*-isomer, and novel compounds resulting therefrom.

The optically inactive *dl* 6-phenyl-2,3,5,6-tetrahydroimidazo[2,1-b]thiazole has been found to be a broad-spectrum anthelmintic compound extremely effective for the treatment of helmenthiasis in domestic animals. A method for the preparation of this *dl*-compound and reports of useful biological activity have been published in British Patent Specification No. 1,043,489. This *dl*-compound has a good margin of safety for many host species but greater margins of safety are needed for some species. In attempts to further develop the usefulness of this compound and improve its margin of safety, considerable effort has been expended to resolve the racemic compound. We have now found a process for resolving the racemic compound into the dextro compound or laevo compound each free or substantially free of contamination by the other. Heretofore, all attempts to achieve resolution have ended in failure, and previously, there has been no means available for obtaining the optically active *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo[2,1-b]thiazole as a pure chemical compound free of the *d*-isomer, nor has there been means available for isolating the optically active *d*(+)6 - phenyl - 2,3,5,6-tetrahydroimidazo[2,1-b]thiazole in a purified form free or substantially free of the contaminating *l*-isomer.

It is therefore surprising to discover that the dextro and laevo antipodes of this compound can be produced in substantially pure forms essentially free of the other isomer. It is also surprising to discover that the desired anthelmintic activity is associated only with the laevo form and that the *l*(−)6 - phenyl-2,3,5,6 - tetrahydroimidazo[2,1 - b]thiazole has a much greater margin of safety in animals than does the known racemic *dl*-compound. With the discovery of the increased margin of safety of the *l*-compound, it is contemplated that the *l*-compound and the hydrogen halide salts thereof will be useful in warm-blooded animals such as specifically exemplified hereinafter for the control of helminths. Although the dextro form had no anthelmintic activity, we have found it to be toxic. In comparing the *dl*-compound with the *l*-compound, we find that the *dl*-compound has a melting point of 90° C. while the *l* or *d*-compounds have a melting point of 60°—61.5° C. and treatments using the laevo form have twice the margin of safety of treatments prepared from the racemic *dl*-compound.

Furthermore, we have found that although the dextro form is ineffective anthelmintically, it has its own utility as an intermediate in a new process for the preparation of the racemic *dl*-compound and subsequently the optically active anthelmintically effective *l*-antipode.

In accordance with the present invention, the laevo compound may be prepared as a crystalline compound or solution free of the

[*Price 4s. 6d.*]

Price 25p

dextro compound. Similarly, the dextro compound may be prepared in crystalline form or as a solution free of the laevo antipode.

The process which we have discovered for the resolution of *dl* 6-phenyl-2,3,5,6-tetrahydroimidazo[2,1-b]thiazole into its dextro and laevo forms takes advantage of an unexpected and unpredictable property of the camphor-10-sulfonic acid salt. At temperatures above about 45° C. both the dextro and laevo 6 - phenyl - 2,3,5,6 - tetrahydroimidazo[2,1-b]thiazoles form salts with *d*-10-camphorsulfonic acid which are extremely soluble in chloroform. As the temperature of the solution is lowered below 35° C. the dextro 6 - phenyl - 2,3,5,6 - tetrahydroimidazo[2,1 - b]thiazole *d* - 10 - camphorsulfonate undergoes a transition to a solvate containing apparently three molecules of chloroform. This solvate is very insoluble in chloroform and crystallizes out as a nicely crystalline compound while the laevo-isomer does not crystallize from chloroform. The recovery of the *d*-acid *d*-base salt can amount to approximately 90% of the theoretical amount and it is essentially optically pure. It can be recrystallized from chloroform with recoveries of about 95%. The laevo 6-phenyl-2,3,5,6-tetrahydroimidazo[2,1-b]thiazole *d*-10-camphorsulfonate can be recovered from the mother liquor from which its diasterio-isomer has crystallized, by evaporation of most of the chloroform and adding hot acetone to the syrupy residue. An insoluble racemic salt separates from the hot solution and this salt is composed of approximately two molecules of the acid and one molecule of *d*-base and one of *l*-base. Filtration of the hot solution removes this salt which contains the balance of the *d*-base along with some of the *l*-base. This racemic salt can be separated into its diasterio-isomers by repeating the recrystallization from chloroform.

When the acetone filtrate from which the insoluble racemic salt was recovered is allowed to cool, crystals of the laevo 6-phenyl-2,3,5,6-tetrahydroimidazo[2,1 - b]thiazole *d* - 10-camphorsulfonate separate. The yield of this salt can amount to about 70% of the theoretical amount. It is obtained in a high state of optical purity but can be recrystallized from acetone if desired.

The diasterio-isomer salts, *d*(+)6-phenyl-2,3,5,6 - tetrahydroimidazo[2,1 - b]thiazole *d*-10-camphor sulphonate and *l*(−)6-phenyl-2,3,5,6 - tetrahydroimidazo[2,1 - b]thiazole *d*-10-camphorsulfonate, can be converted to their free bases by treatment of the salts, or preferably aqueous solutions of the salts, with an amine, ammonia or an alkali metal hydroxide or carbonate, extracting the mixture with chloroform and then recovering from the chloroform mixture the *d*(+)-base substantially free of the *l*-isomer or the *l*(−)-base substantially free of the *d*-isomer. The liberated dextro or laevo 6-phenyl-2,3,5,6-tetrahydroimidazo[2,1-b]thiazole bases can be converted to pharmaceutically acceptable salts by methods familiar to those skilled in the art. For example, the hydrogen halide salts can be prepared by treating the *l*(−)- or *d*(+)-base with acetone and admixing a lower alcoholic hydrogen halide with the acetone solution. The camphorsulfonic acid can also be recovered by procedures familiar to those skilled in the art and used in subsequent resolutions.

For simplicity the process has so far been described for the use of *d*-10-camphorsulfonic acid. The process can be operated in exactly the same way with *l*-10-camphorsulfonic acid. The compounds obtained with this isomer will be identical in all physical and chemical properties to those obtained with the *d*-isomer except that they will rotate the plane of polarized light in the opposite direction.

Although both optically active forms of the thiazole base can be obtained readily from either *d*- or *l*-10-camphorsulfonic acid, the *d*-acid gives the best yield of the *d*-base and the *l*-acid gives the best yield of the desired *l*-base. We have found that the process can be operated to resolve *dl*-10-camphorsulfonic acid into its *d* and *l* forms. The *l*-10-camphorsulfonic acid is the preferred form for preparing *l*(−)6 - phenyl - 2,3,5,6-tetrahydroimidazo[2,1-b]thiazole. The resolution of *dl*-base with an optically active 10-camphorsulfonic acid can be carried out as described above and the optically active bases obtained can in turn be used to resolve *dl*-10-camphorsulfonic acid. The resolution of the *dl*-base and the *dl*-10-camphorsulfonic acid can be carried out in the same operation by the following procedure. The *dl*-base is treated with at least enough *d*-10-camphorsulfonic acid in chloroform to cause most of the *d*-base to be crystallized out by cooling to below 35° C. as the chloroform solvate of *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo-[2,1-b]thiazole *d* - 10 - camphorsulfonate. After the crystals have been collected, the mother liquor is concentrated to a syrup and hot acetone is added which causes the separation of an insoluble racemic salt leaving substantially pure *l*(−)base and its *d*-10-camphorsulfonate in solution. The base is recovered as the free base and combined with *dl*-10-camphorsulfonic acid in hot chloroform. The product that crystallizes on cooling is *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo[2,1-b]thiazole *l*-10-camphorsulfonate which separates as the highly insoluble chloroform solvate. After the crystals are collected, the chloroform is evaporated and replaced by hot acetone. Now the *l*(−)6-phenyl - 2,3,5,6 - tetrahydroimidazo[2,1 - b]-thiazole *d*-10-camphorsulfonate crystallizes

from the solution on cooling. The chemical and physical properties of the salts obtained on resolving *dl*-10-camphorsulfonic acid are identical to those obtained in resolving the *dl*-base with the *d*-acid except that the *l*-base *l*-acid salt will rotate the plane of polarized light in the opposite direction from the *d*-base *d*-acid salt. There is no racemic double salt formed in the resolution of the racemic acid with optically active base.

When the various 10-camphorsulfonates are decomposed, both the *d*- and the *l*-base and the *d* and the *l*-camphorsulfonic acids are obtained. Some modifications, such as a shift in the acid-base ratio and the use of mixtures of solvents or other solvents for some of the crystallizations are premissible but appear to offer no advantage.

The following examples illustrate the process aspects of the present invention and the novel products produced thereby. Example 1 illustrates the preparation of the *dl*-base from its hydrochloride, and Example 2 is a comparative example.

EXAMPLE 1.

dl 6 - phenyl - 2,3,5,6 - tetrahydroimidazo[2,1-b]thiazole hydrochloride, 188 grams (0.785 mole), is suspended in a mixture of 500 milliliters of water and 500 milliliters of methylene chloride. The suspension is stirred mechanically while 20% sodium hydroxide solution is added until the solution is basic. Ice is added from time to time to keep the temperature below the boiling point of the methylene chloride. The methylene chloride layer is separated, washed with water, dried over potassium carbonate and evaporated. The oily residue crystallizes with the evolution of the heat when poured into a beaker containing 100 milliliters of ether. The free base is washed with ether. The yield of *dl* 6-phenyl-2,3,5,6-tetrahydroimidazo[2,1-b]thiazole is 151.4 grams (0.746 mole), 94%. The product has a melting point of 90° C.

EXAMPLE 2.

A mixture of 2.04 grams (10 m. mole) of *dl* 6 - phenyl - 2,3,5,6 - tetrahydroimidazo-[2,1-b]thiazole and 2.32 grams (10 m. mole) of *d*-10-camphorsulfonic acid is dissolved in 10 milliliters of 95% ethanol. On standing large crystals separate which are recovered by filtration and washed with ethanol. This salt has a melting point of 195°—196° C. and a rotation of + 14.1° (conc. 16%, H_2O). It is *dl* 6 - phenyl - 2,3,5,6 - tetrahydroimidazo-[2,1-b]thiazole *d*-10-camphorsulfonate. This salt recrystallized unchanged from acetonitrile, nitromethane, ethylene chloride and several other solvents. It is very insoluble in methyl isobutyl ketone and acetone. It is a racemic compound and when the base is recovered from this salt, it is found to be totally unresolved.

EXAMPLE 3.

A solution of 204.3 grams (1 mole) of *dl* 6 - phenyl - 2,3,4,5 - tetrahydroimidazo[2,1-b]thiazole and 232.3 grams (1 mole) of *d*-10-camphorsulfonic acid in 1750 milliliters of chloroform is allowed to crystallize overnight at − 28° C. The solvate is recovered by filtration and washed with ice cold chloroform (400 milliliters). The solvate is dried (decomposed) under nitrogen seven hours and then in air overnight. The yield of *d*(+)6-phenyl-2,3,5,6 - tetrahydroimidazo[2,1 - b]thiazole *d*-10-camphorsulfonate is 202.5 grams (0.464 mole) 92.8%, melting point 139°—140° C, $[\alpha]_D^{25}$ + 82.6 (conc. 16%, H_2O).

The trisolvate separates only below its transition temperature which is about 35° C. Cooling to 0° C is about as good as − 28° C. It separates as fine needles which are very easy to filter and wash. The solubility in chloroform is only 0.34 grams per 100 milliliters at 0° C. The solvate becomes sticky in air on humid days but can be dried in a stream of air on dry days. It dries to the non-solvated apparently anhydrous salt. The solvate melts about 35° C and is soluble in methylene chloride and in acetone. It can be recrystallized from 2 milliliters or less per gram of chloroform with a recovery of 95%. Recrystallized *d*(+)-base *d*-acid melts at 140°—141° C and has $[\alpha]_D^{25}$ + 83 (conc. 15%, H_2O).

Following the procedure above and substituting *l*-10-camphorsulfonic acid for *d*-10-camphorsulfonic acid yields the *l*(−)6-phenyl - 2,3,5,6 - tetrahydroimidazo[2,1 - b]-thiazole *l*-10-camphorsulfonate.

EXAMPLE 4.

l(−)6-Phenyl-2,3,5,6-tetrahydroimidazo-[2,1-b]thiazole *d*-10-camphorsulfonate.

The mother liquor from which the salt is prepared in Example 3 is taken and is evaporated to a syrup. The syrup which is approximately one-half chloroform by weight is treated with 1500 milliliters of hot acetone. The resulting solution is kept near the boiling point for ten minutes during which time a solid crystallizes out. The crystals are collected by filtering the hot solution. The material on the filter is washed with 200 milliliters of hot acetone and dried. This racemic salt weighs 24.2 grams (0.055 mole) and melts at 186°—192° C $[\alpha]_D^{25}$ − 14.7 (conc. 16%, H_2O). It is not an optically pure salt and can be resolved by recrystallization from chloroform. The mother liquor (acetone filtrate) is stored at − 15° C overnight. The crystalline product is recovered by filtration and washed with 150 milliliters of cold acetone. The yield of *l*(−)6-phenyl-2,3,5,6-

tetrahydroimidazo[2,1 - b]thiazole *d* - 10-camphorsulfonate is 168.1 grams (0.385 mole), 77%, melting point 130°—132° C $\alpha]_D^{25}$ – 54.7 (conc. 16%, H_2O).

The mother liquor is evaporated leaving 22.8 grams of syrup. The hot syrup is dissolved in 100 milliliters of hot methyl isobutyl ketone. The product crystallizes within a few minutes. The mixture is cooled to room temperature and the product recovered by filtration and washed with methyl isobutyl ketone. The yield of *l*(–)-base *d*-acid from this fraction is 16.74 grams (0.038 mole), 7.7%, melting point 128°—130° C (cloudy) $\alpha]_D^{25}$ – 57.1° (conc. 16%, H_2O).

The first crop of crystals, 168 grams, is recrystallized from 5 milliliters per gram of acetone (cooled to – 15° C). The recovery is 158 grams, 94%, melting point 130°—131° C $\alpha]_D^{25}$ – 56.3 (conc. 15%, H_2O).

Where the mother liquor from the process of Example 3 is obtained from the process employing the *l*-camphorsulfonic acid and said mother liquor is treated in the manner above, *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo-[2,1-b]thiazole *l*-10-camphorsulfonate is obtained.

EXAMPLE 5.

d(+)6-Phenyl-2,3,5,6-tetrahydroimidazo-[2,1-b]thiazole.

A solution of 150 grams (0.344 mole) of *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo-[2,1-b]thiazole. *d*-10-camphorsulfonate in water is treated with 15.5 grams (0.378 mole) of 98% sodium hydroxide and the liberated base extracted with chloroform. The chloroform solution is washed with water followed by sodium chloride solution and dried over magnesium sulfate. Evaporation of the solvent left 72.1 grams of residue which crystallized shortly. The desired free base hereby obtained has a melting point of 60°—61.5° C. and an optical rotation $\alpha]_D^{25}$ + 85.1 (conc. 10%, $CHCl_3$).

EXAMPLE 6.

d(+)6-Phenyl-2,3,5,6-tetrahydroimidazo-[2,1-b]thiazole hydrochloride.

The free base *d*(+)6-phenyl-2,3,5,6-tetrahydroimidazo[2,1-b]thiazole is dissolved in 112 milliliters of acetone and 178 milliliters of isopropanolic hydrogen chloride is added all at once. The hydrochloride crystallizes at once. After cooling to below 0° C, the salt is recovered by filtration and washed with acetone. The desired hydrochloride weighs 75.2 grams (0.312 mole), 91%, from the camphorsulfonate, melting point 227°—227.5° C $\alpha]_D^{25}$ + 123.1° (conc. 15%, H_2O).

EXAMPLE 7.

l(–)6-Phenyl-2,3,5,6-tetrahydroimidazo-[2,1-b]thiazole.

A solution is made by dissolving 158.1 grams (0.36 mole) of *l*(–)6-phenyl-2,3,5,6-tetrahydroimidazo[2,1 - b]thiazole *d* - 10-camphorsulfonate in 400 milliliters of water. The solution is clarified by treating with a little "Darco G 60" ("Darco G" is a registered Trade Mark) charcoal and filtering. The salt is decomposed by adding a solution of 16.3 grams (0.4 mole) of 98% sodium hydroxide in 100 milliliters of water. The free base is recovered by three extractions with chloroform. The chloroform extracts are combined, washed with water and with saturated sodium chloride solution, and dried over magnesium sulfate. Evaporation of the solvent left 75 grams of the desired free base having a melting point 60°—61.5° C. and an optical rotation $\alpha]_D^{25}$ – 85.1° (conc. 10%, $CHCl_3$).

EXAMPLE 8.

l(–)6-Phenyl-2,3,5,6-tetrahydroimidazo-[2,1-b]thiazole hydrochloride.

The free base *l*(–)6-phenyl-2,3,5,6-tetrahydroimidazo[2,1-b]thiazole is dissolved in 125 milliliters of acetone. The turbidity is removed by mixing in a little activated charcoal and filtering. The acetone solution is treated all at once with 110 milliliters of 3.82 N isopropanolic hydrogen chloride. The hydrochloride crystallizes rapidly. After cooling, it is recovered by filtration and washed with acetone. The yield is 84.0 grams (0.349 mole), 96.9%, based on the *d*-10-camphorsulfonate. The desired hydrochloride has a melting point of 227°—229° C and has $\alpha]_D^{25}$ – 122.9° (conc. 15%, H_2O).

EXAMPLE 9.

Resolution of *dl*-10-camphorsulfonic acid.

A warm solution prepared by dissolving 8.5 grams (0.0416 mole) of *l*(–)6-phenyl-2,3,5,6-tetrahydroimidazo[2,1-b]thiazole and 9.67 grams (0.416 mole) of *dl*-10-camphorsulfonic acid in 83 milliliters of chloroform is allowed to crystallize below 0° C overnight. The chloroform solvate is recovered by filtration and washed with cold chloroform and decomposed by drying in a nitrogen atmosphere. The yield is 8.66 grams (0.0198 mole), 95%, of the theoretical amount of *l*(–)6 - phenyl - 2,3,5,6 - tetrahydroimidazo-[2,1-b]thiazole *l*-10-camphorsulfonate. The product has a melting point of 135°—138° C and has $\alpha]_D^{25}$ – 72.7 (conc. 15%, H_2O). The product is the mirror image of *d*(+)6-phenyl-2,3,5,6 - tetrahydroimidazo[2,1 - b]thiazole *d*-10-camphorsulfonate and has a melting point of 141° C and $\alpha]_D^{25}$ – 84° after purification.

The mother liquor from which the solvate is filtered is evaporated to a syrup and 63 milliliters of hot acetone is added. The solution is filtered to remove a trace of insoluble material and allowed to cool to – 5° C. The crystals which separate are collected by filtration and washed with cold acetone. The yield is 6.0 grams (0.137 mole), 63%, melting point 131.5°—132.5° C (cloudy), $\alpha]_D^{25}$ + 50.0

(conc. 15%, H_2O). The salt is *l*(−)6-phenyl-2,3,5,6 - tetrahydroimidazo[2,1 - b]thiazole *d*-10-camphorsulfonate. This is the same product as prepared in Example 4.

EXAMPLE 10.

Recovery of Ammonium *d*-10-camphorsulfonate.

A solution is prepared by dissolving 277.6 grams (0.63 mole) of *l*(−)6-phenyl-2,3,5,6-tetrahydroimidazo[2,1 - b]thiazole *d* - 10-camphorsulfonate in 750 milliliters absolute ethanol by warming. Gaseous ammonia is added to the stirred solution to decompose the salt and precipitate the *d*-10-camphorsulfonic acid as the ammonium salt. The salt is recovered by filtration and washed with absolute ethanol followed by acetone. The recovered salt weights 102.6 grams (0.41 mole). The filtrate is evaporated to a syrup which is partitioned between chloroform and ammonium hydroxide solution. The aqueous layer is separated and evaporated to dryness yielding an additional 44.7 grams (0.179 mole) of ammonium *d*-10-camphorsulfonate bringing the total recovery to 93.5%. The *l*(−)6-phenyl - 2,3,5,6 - tetrahydroimidazo[2,1 - b]-thiazole is recovered by evaporation of the chloroform phase.

Other solvents can be substituted for ethanol or the ammonium salt can be obtained by evaporation of the water from an ammonium hydroxide/chloroform partition. The ammonium salt can also be salted out of aqueous solution.

EXAMPLE 11.

Mice were experimentally inoculated with thirty infectious larvae of *Nematospiroides dubius*. After 19 days, when the worms had matured, randomly selected groups of eight mice for each dosage level of the three compounds (*l*)-, (*dl*)-, and (*d*)-6-phenyl-2,3,5,6-tetrahydroimidazo[2,1 - b]thiazole received single oral doses by gavage. Three days after treatment, the mice were sacrificed, the small intestine removed, and all worms present were enumerated. Average worm numbers were determined for the group of eight treated mice on each regimen, and the percent efficacy of each dosage was calculated by comparison with average worm numbers for 10 or 20 control mice which had not received drug treatment using the customary formula:

$$\% \text{ Efficacy} = \frac{\text{Avg. Worms in Controls} - \text{Avg. Worms in Treated}}{\text{Avg. Worms in Controls.}} \times 100$$

Test B indicates that the dextro compound is markedly inferior to the others, showing less than 50% activity at 48 mg./kg orally, while 150 mg./kg. orally is highly lethal to mice (see later). Tests A and B show that the laevo compound (ED_{50} about 5 mg./kg.) is approximately twice as active anthelmintically as the dextro-laevo compound (ED_{50} about 10 mg./kg.). The following Table I summarizes the above results.

TABLE I

% Efficacy in Mice

Oral Dose mg./kg.	Test A		Test B		
	laevo	dextro-laevo	laevo	dextro-laevo	dextro
2	0	—	0	—	—
4	22	—	47	—	—
6	—	0	69	10	—
8	88	36	79	31	—
12	—	71	97	61	—
16	—	93	—	83	0
32	—	—	—	—	22
48	—	—	—	—	36

EXAMPLE 12.

The above results on the comparable activity of 5 mg./kg. of the laevo and 10 mg./kg. of the dextro-laevo compound have been confirmed in additional tests by similar procedures. The laevo compound at 5 mg./kg. removed on the average 68% of adult *N. dubius*, whereas the dextro-laevo at twice this dose removed on the average 74% of the worms. Again (Test F) the dextro compound at a much higher dose, 40 mg./kg. removed only 34% of the worms. Testing results are shown in the following Table II.

TABLE II — MICE

Test	Compound	No. Mice per Dose	% Efficacy at Doses Shown mg./kg. 5	10	20	40
C	dl	8	—	74	99	—
C	l	8	65	97	99	—
D	dl	8	—	77	—	—
D	l	8	74	—	—	—
E	dl	8	12	70	—	—
E	l	40	64	—	—	—
F	dl	5	—	—	98	—
F	d	17	—	—	—	34

EXAMPLE 13.

The acute oral and subcutaneous toxicity of the above three compounds was compared in white mice. All mice were held for four or more days after dosing. Mortality due to drug occurred predominately within the first four hours after administration. The following Table III shows that there are no significant differences between the acute oral toxicity of the three compounds. The acute subcutaneous toxicity are also similar, but the laevo compound may be slightly less toxic than the others. The mouse data on similar toxicity and dissimilar activity of the laevo compared to the dextro-laevo compound indicate that the former gives a twofold improvement in the chemotherapeutic index, i.e. in safety, over the latter.

TABLE III

mg./kg.	Dead/Total Mice laevo	dextro-laevo	dextro
		Oral Dosage	
100	0/10	1/10	1/10
150	16/30	8/30	8/10
200	16/20	15/20	8/10
300	9/10	10/10	9/10
		Subcutaneous Dosage	
20	0/10	0/10	0/10
40	0/30	1/30	2/10
60	7/30	20/30	10/10
80	23/30	18/20	—

EXAMPLE 14.

Young lambs were experimentally inoculated with infectious larve of *Trichostrongylus colubriformis*. After three weeks, when mature adult worms had developed, the sheep were allocated at random to various dosage groups containing four sheep each, and the animals treated by the oral drench method. All lambs were slaughtered several days after treatment, and the numbers of worms present in the small intestine were determined. The five unmedicated lambs used for this trial had an average of 3,520 worms (range 2,830 to 3,870). The percent efficacy for treated groups was determined by the conventional method of "controlled tests" (See Example 11) with four treated sheep per medicated treatment level.

The results indicate that on a dosage basis the laevo compound is approximately twice as active as the dextro-laevo compound against this important sheep helminth nematode.

In other trials on subcutaneous toxicity, two sheep received 40 mg/kg. of the laevo compound, and two other sheep received 40 mg./kg. of the dextro-laevo compound. All four sheep showed similar transient mild side reactions. Thus, the two compounds have similar subcutaneous toxicity at dosages in the neighborhood of the maximum tolerated dosage. In other subcutaneous toxicity trials with a dosage of the dextro-laevo compound of 80 mg./kg. killed two of two sheep, and one of 60 mg./kg. produced moderate to severe side reactions in three sheep, and mild reactions in three others. Data summarized in the following Table IV.

TABLE IV

Sheep Activity Data

Compound	Oral Dose mg./kg.	% Efficacy T. colubriformis
laevo	2.5	72%
laevo	3.75	92%
dextro-laevo	2.5	33%
dextro-laevo	5.0	83%
dextro-laevo	7.5	97%

EXAMPLE 15.

In comparative toxicity trials with calves, four calves each received 40 mg./kg. intramuscularly of the *l* (two doses), the *dl* (one dose), and the *d* compound (one dose) using a "cross-over" design and with an interval of one week between the four successive doses. The side reactions observed occurred promptly, no animals died, and all recovered within several hours. The laevo compound may be less toxic than the others at the above dosage to judge by the following tabulation.

TABLE V

Incidence and Severity of Side Reactions

Compound 40 mg./kg.	Mild	Mild to Moderate	Moderate	Moderate to Severe
l	4	4	0	0
dl	1	1	1	1
d	0	1	2	1

Two weeks after the last of the above series of 40 mg./kg. doses, all four calves were treated with 60 mg./kg. intramuscularly with results as follows: one calf which received the dextro compound showed very severe reactions and died within two hours; one calf which received the dextro-laevo compound showed moderate transient side reactions: two calves which received the laevo compound showed mild to moderate transient symptoms. Twenty days following the above 60 mg./kg. doses, two of the surviving calves received 90 mg./kg. intramuscularly of either the laevo or the dextro-laevo compound—both developed very severe reactions and died in 105 and 50 minutes, respectively.

The above data at 60 and 90 mg./kg. further indicates that the three compounds have relatively similar toxicity to cattle, but the laevo compound may be somewhat less toxic at doses below 90 mg./kg. intramuscularly.

A comparison between the oral toxicity for cattle of the laevo compound and the dextro-laevo compound was made at 40 mg./kg. by oral drench. A "cross-over" design was used, i.e., two animals each first received the laevo and two the dextro-laevo compound. Several weeks later the four animals were treated again, but the compounds used were interchanged. Similar transient and mild side reactions were observed after the above four treatments with each of the two compounds, primarily intermittent restlessness for several hours. The dosage of 40 mg./kg. used approaches the maximum tolerated dose for the dextro-laevo compound. The other cattle toxicity trials using the oral drench procedure, two of six animals receiving 60 mg./kg. of the dextro-laevo compound died after treatment, whereas the other four animals showed mild to moderate side reactions.

EXAMPLE 16.

Cattle with naturally acquired nematode infections were divided into groups of five animals which had similar average numbers of nematode eggs in their stools. Five animals were treated by the oral drench procedure at each of various dosages of the dextro-laevo or of the laevo compound, and two groups of five animals each were retained as controls. All animals were sacrificed three or four days after treatment, necropsied, the worms present in their abomasa counted, and the average percent efficacy determined by comparison with ten unmedicated controls.

The results (Table VI) below show that the laevo compound is considerably more active than the dextro-laevo compound against *T. axei* on a dosage basis, as 2.5 mg./kg. of the former had efficacy at least equivalent to 5.0 mg./kg. and probably to 7.5 mg./kg. of the latter. Similar results were also obtained against *Haemonchus* and *Ostertagia*. However, since 5 mg./kg. of the dextro-laevo compound was highly effective against both these helminths, unlike *T. axei*, quantitative comparison between the two compounds cannot be made.

TABLE VI

Compound	Oral Dose mg./kg.	% Efficacy (Avg. Worm Number) Trichostrongylus axei	Haemonchus	Ostertagia
None	—	(10,500)	(370)	(1,600)
laevo	2.5	93% (716)	99% (5)	96% (64)
	5.0	94% (680)	100% (0)	91% (136)
dextro-laevo	5.0	60% 4,150	100% (0)	90% (165)
	7.5	94% (640)	100% (0)	96% (72)

The data cited in Examples 14—16 indicate that the laevo and dextro-laevo compounds have similar toxicity in sheep and cattle, and that the quantitative activity of the laevo compound is greater against at least some important nematodes of these ruminants. Therefore, the laevo compound can be used with greater assurance of safety, than the dextro-laevo compound.

The present invention also includes

anthelmintic compositions comprising *l*(−)6-phenyl - 2,3,5,6 - tetrahydroimidazo[2,1 - b]-thiazole free or substantially free of the *d*-isomer, or a hydrogen halide salt of this optical isomer, and an inert, non-toxic carrier.

WHAT WE CLAIM IS:—

1. *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole free or substantially free of the *d*-isomer.

2. *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole free or substantially free of the *l*-isomer.

3. *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole having a rotation of $\alpha]_D^{25} - 85.1°$ (concentration 10%, $CHCl_3$) and melting point 60°—61.5° C.

4. *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole having a rotation of $\alpha]_D^{25} + 85.1°$ (concentration 10%, $CHCl_3$) and melting point 60°—61.5° C.

5. A hydrogen halide salt of either of the compounds defined in Claims 3 and 4.

6. The hydrogen chloride salt of either of the compounds defined in Claims 3 and 4.

7. A method of resolving *dl* 6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole comprising: treating *dl* 6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole with chloroform and *l*-10- or *d*-10-camphorsulphonic acid; cooling the mixture obtained to below 35° C; separating the crystallized chloroform solvate of *d*(+)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *d*-10-camphorsulfonate or *l*-6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *l*-camphorsulfonate from the mother liquor; evaporating the mother liquor; treating the residue with hot acetone; separating an insoluble racemic salt consisting of *dl* 6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *d*-10-camphorsulfonate or *dl* 6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *l*-10-camphorsulfonate; after cooling, separating from the acetone filtrate *l*(−)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *d*-10-camphorsulfonate or *d*(+)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *l*-10-camphorsulfonate; treating the *l*(−)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *d*- or *l*-10-camphorsulfonates or the *d*(+)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *d*- or *l*-10-camphorsulfonates with an amine, ammonia or an alkali metal hydroxide or carbonate; extracting the mixture obtained with chloroform; and recovering from the chloroform extract *d*(+)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole free or substantially free of the *l*-isomer or *l*(−)6-phenyl - 2,3,5,6 - tetrahydroimidazo [2,1 - b] thiazole free or substantially free of the *d*-isomer.

8. A method of preparing a hydrogen halide salt of *d*(+) or *l*(−)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole, comprising treating the *d*(+) or *l*(−) free base with acetone and admixing a lower alcoholic hydrogen halide with said acetone solution to produce the desired hydrogen halide salt.

9. *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole *d*-10-camphorsulfonate.

10. *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole *l*-10-camphorsulfonate.

11. *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole *d*-10-camphorsulfonate.

12. *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole *l*-10-camphorsulfonate.

13. A method of resolving *dl*-10-camphorsulfonic acid, comprising treating *dl*-10-camphorsulfonic acid with chloroform and *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole or *l*(−)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole; cooling the mixture obtained to below 35° C; separating the chloroform solvate *d*(+)6-phenyl - 2,3,5,6 - tetrahydroimidazo [2,1 - b] thiazole *d*-10-camphorsulfonate or *l*(−)6-phenyl - 2,3,5,6 - tetrahydroimidazo [2,1 - b] thiazole *l*-10-camphorsulfonate from the mother liquor; evaporating the mother liquor; treating the concentrated mother liquor with hot acetone; cooling the acetone mixture to crystallize the product; recovering *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole *d*-10-camphorsulfonate or the *d*(+)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *l*-10-camphorsulfonate; and if desired treating the product thus formed with ammonia to give the ammonium salt.

14. A method of resolving *dl*-6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole, substantially as hereinbefore described.

15. *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole free or substantially free of the *l*-isomer, whenever obtained by a method according to Claim 7 or Claim 14.

16. *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole free or substantially free of the *d*-isomer, whenever obtained by a method according to Claim 7 or Claim 14.

17. A hydrogen halide salt of *d*(+) or *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole, whenever prepared by a method according to Claim 8.

18. A method of controlling helminths in warm-blooded animals, other than human beings, comprising treating the animals with *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole free or substantially free of the *d*-isomer, or a hydrogen halide salt of this optical isomer.

19. An anthelmintic composition comprising *l*(−)6-phenyl-2,3,5,6-tetrahydroimidazo

anthelmintic compositions comprising *l*(−)6-phenyl - 2,3,5,6 - tetrahydroimidazo[2,1 - b]-thiazole free or substantially free of the *d*-isomer, or a hydrogen halide salt of this optical isomer, and an inert, non-toxic carrier.

WHAT WE CLAIM IS:—

1. *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole free or substantially free of the *d*-isomer.

2. *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole free or substantially free of the *l*-isomer.

3. *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole having a rotation of $\alpha]_D^{25} - 85.1°$ (concentration 10%, $CHCl_3$) and melting point 60°—61.5° C.

4. *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole having a rotation of $\alpha]_D^{25} + 85.1°$ (concentration 10%, $CHCl_3$) and melting point 60°—61.5° C.

5. A hydrogen halide salt of either of the compounds defined in Claims 3 and 4.

6. The hydrogen chloride salt of either of the compounds defined in Claims 3 and 4.

7. A method of resolving *dl* 6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole comprising: treating *dl* 6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole with chloroform and *l*-10- or *d*-10-camphorsulphonic acid; cooling the mixture obtained to below 35° C; separating the crystallized chloroform solvate of *d*(+)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *d*-10-camphorsulfonate or *l*-6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *l*-camphorsulfonate from the mother liquor; evaporating the mother liquor; treating the residue with hot acetone; separating an insoluble racemic salt consisting of *dl* 6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *d*-10-camphorsulfonate or *dl* 6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *l*-10-camphorsulfonate; after cooling, separating from the acetone filtrate *l*(−)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *d*-10-camphorsulfonate or *d*(+)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *l*-10-camphorsulfonate; treating the *l*(−)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *d*- or *l*-10-camphorsulfonates or the *d*(+)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *d*- or *l*-10-camphorsulfonates with an amine, ammonia or an alkali metal hydroxide or carbonate; extracting the mixture obtained with chloroform; and recovering from the chloroform extract *d*(+)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole free or substantially free of the *l*-isomer or *l*(−)6-phenyl - 2,3,5,6 - tetrahydroimidazo [2,1 - b] thiazole free or substantially free of the *d*-isomer.

8. A method of preparing a hydrogen halide salt of *d*(+) or *l*(−)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole, comprising treating the *d*(+) or *l*(−) free base with acetone and admixing a lower alcoholic hydrogen halide with said acetone solution to produce the desired hydrogen halide salt.

9. *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole *d*-10-camphorsulfonate.

10. *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole *l*-10-camphorsulfonate.

11. *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole *d*-10-camphorsulfonate.

12. *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole *l*-10-camphorsulfonate.

13. A method of resolving *dl*-10-camphorsulfonic acid, comprising treating *dl*-10-camphorsulfonic acid with chloroform and *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole or *l*(−)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole; cooling the mixture obtained to below 35° C; separating the chloroform solvate *d*(+)6-phenyl - 2,3,5,6 - tetrahydroimidazo [2,1 - b] thiazole *d*-10-camphorsulfonate or *l*(−)6-phenyl - 2,3,5,6 - tetrahydroimidazo [2,1 - b] thiazole *l*-10-camphorsulfonate from the mother liquor; evaporating the mother liquor; treating the concentrated mother liquor with hot acetone; cooling the acetone mixture to crystallize the product; recovering *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole *d*-10-camphorsulfonate or the *d*(+)6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole *l*-10-camphorsulfonate; and if desired treating the product thus formed with ammonia to give the ammonium salt.

14. A method of resolving *dl*-6-phenyl-2,3,5,6-tetrahydroimidazo [2,1-b] thiazole, substantially as hereinbefore described.

15. *d*(+)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole free or substantially free of the *l*-isomer, whenever obtained by a method according to Claim 7 or Claim 14.

16. *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole free or substantially free of the *d*-isomer, whenever obtained by a method according to Claim 7 or Claim 14.

17. A hydrogen halide salt of *d*(+) or *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole, whenever prepared by a method according to Claim 8.

18. A method of controlling helminths in warm-blooded animals, other than human beings, comprising treating the animals with *l*(−)6 - phenyl - 2,3,5,6 - tetrahydroimidazo [2,1-b] thiazole free or substantially free of the *d*-isomer, or a hydrogen halide salt of this optical isomer.

19. An anthelmintic composition comprising *l*(−)6-phenyl-2,3,5,6-tetrahydroimidazo

* C L M S *

[2,1-b] thiazole free or substantially free of the *d*-isomer, or a hydrogen halide salt of this optical isomer, and an inert, non-toxic carrier.

TREGEAR, THIEMANN & BLEACH,
Chartered Patent Agents,
Melbourne House, Aldwych, London, W.C.2,
Agents for the Applicants.

Printed for Her Majesty's Stationery Office by the Courier Press, Leamington Spa, 1969.
Published by the Patent Office, 25 Southampton Buildings, London, W.C.2, from which copies may be obtained.

