

## Synthesis and Fasciolicidal Activity of 5-Chloro-2-methylthio-6-(1-naphthoxy)-1H-benzimidazole\*\*

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**The synthesis and fasciolicidal activity of 5-chloro-2-methylthio-6-(1-naphthoxy)-1H-benzimidazole (6) is described. Compound 6 showed 100% activity *in vitro* at 146.70 and 29.34  $\mu\text{M}$  concentrations. It also completely removed 3-d and 10-week-old *Fasciola hepatica* in sheep at a dose of 15 mg/kg.**

**Key words** fasciolicidal activity; bioisostere; benzimidazole synthesis; *Fasciola hepatica*

Fasciolosis, caused by *Fasciola hepatica* and *Fasciola gigantica*, is a very serious parasitic disease, which is responsible for heavy economic losses in sheep and cattle production in many countries of the world.<sup>1)</sup>

Besides domestic animals, other animal species and man may be infected as well.<sup>2)</sup> There are effective strategies for the control of fasciolosis which are largely based on drug (fasciolicide) use on the definitive host.<sup>3)</sup> So far the only effective drug against immature and adult flukes alike is triclabendazole (TCBZ, Fig. 1), a potent fasciolicide with a better bioavailability than that of the benzimidazole carbamates, such as albendazole. It differs structurally from the latter in having a 2-methylthio group instead of a 2-methylcarbamate group in the benzimidazole ring.<sup>4)</sup>

TCBZ appears to be highly specific for flukes, possessing 97–100% activity against all stages of *Fasciola* spp. The usual oral dose of TCBZ for the elimination of immature and adult forms of *F. hepatica* is 5–10 mg/kg for sheep and goats, and 12 mg/kg for cattle.<sup>5)</sup>

For safety and efficacy, TCBZ is the drug of choice for the treatment of human fasciolosis.<sup>6–10)</sup> Unlike nematocides, no new fasciolicides have been marketed recently and apparently no compounds are yet under development.<sup>1)</sup> Considering that fasciolosis is also a health problem in Mexico, a research project was undertaken in our laboratory with the purpose of developing new compounds with potential fasciolicidal activity.<sup>11,12)</sup>

For the design of one of the new compounds described in this paper, we use as a model pronethalol, an adrenergic blocker which was formed by replacing the 3,4-dichlorophenyl group in dichloroisoproterenol (DCI, Fig. 1) by the 2-naphthyl group.<sup>13)</sup> DCI is also an adrenergic blocker with some agonist activity thus establishing the fact that pronethalol is a bioisostere of DCI.

In this paper we report the synthesis and preliminary fasciolicidal activity, *in vitro* and *in vivo*, against *F. hepatica*, of 5-chloro-2-methylthio-6-(1-naphthoxy)-1H-benzimidazole (6), a TCBZ analog. In this compound, the 1-naphthyl group replaces the 3,4-dichlorophenyl group in TCBZ, a moiety change, which is analogous to that done with pronethalol-DCI.

### Chemistry

The synthetic sequence in the preparation of 6 is shown in

Chart 1. In the first step, 4,5-dichloro-2-nitroaniline (1) was subjected to a nucleophilic substitution reaction with 1-naphthol (2) under known conditions,<sup>14)</sup> and the ether obtained (3) was reduced with  $\text{SnCl}_2 \cdot 2\text{H}_2\text{O}$ .<sup>15)</sup> The corresponding *o*-phenylenediamine (4) formed was immediately cyclocondensed with  $\text{CS}_2$  in an EtOH–KOH solution to give the 2-mercaptobenzimidazole (5).<sup>16)</sup> In the last step, 5 was monoalkylated with  $\text{CH}_3\text{I}$  in acetone and KOH solution<sup>17)</sup> to afford the title compound (6).

The synthesized compounds were purified by recrystallization and their physical constants were determined. The structure of all new compounds was established by spectroscopic and spectrometric data.

### Pharmacology

Compound 6 was evaluated *in vitro* against newly excysted *F. hepatica* metacercariae at 146.70, 29.34, 9.68, 3.22, and 1.08  $\mu\text{M}$  concentrations.<sup>18,19)</sup> In this experiment we used the deacetylated (amine) metabolite of diamphenetide (DAMD), as a reference standard. Diamphenetide is a phenoxyalkane compound which is highly effective against early immature flukes from 1 d up to 6 weeks, at concentrations of 1.08–3.81  $\mu\text{M}$ .<sup>19)</sup> The activity was assessed by comparing the survival of flukes with the untreated control after a 4-d period of exposure to 6; the results of these studies are shown in Table 1.

For the *in vivo* evaluation two experiments were carried out. In the former, 27 crossbred sheep were infected each with 150 metacercariae; ten weeks after infection the sheep were randomly divided in three groups of 9 animals each.

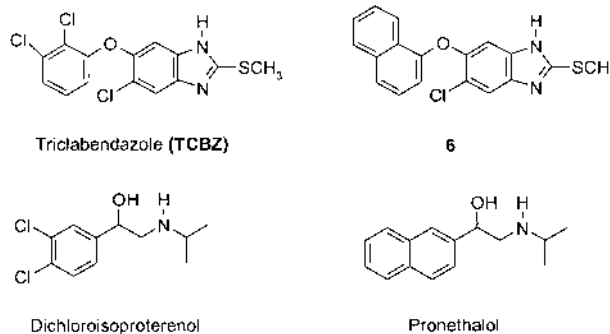


Fig. 1. Isosteric Substitution in TCBZ and Dichloroisoproterenol (DCI) That Led to 6 and Pronethalol, Respectively

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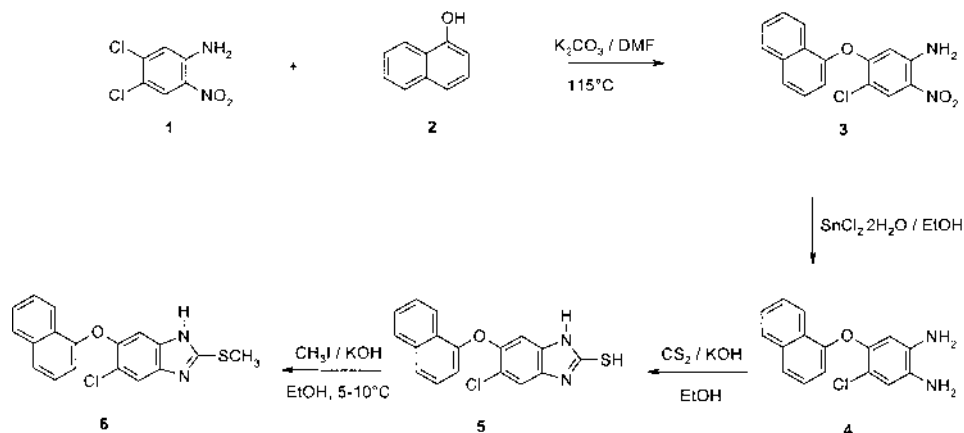


Chart 1. Synthesis of 5-Chloro-2-methylthio-6-(1-naphthoxy)-1*H*-benzimidazole (**6**)

Groups 1 and 2 were treated orally with a dose of 10 and 15 mg/kg of compound **6**, respectively, in gelatin capsules. Group 3 was the untreated control. Two weeks after treatment, all sheep were killed; the flukes were collected from the liver and counted. Efficacy was determined as a percentage of fluke reduction by the treatment in comparison with the control (Table 2).<sup>12)</sup>

In a second experiment, sixty 10 to 12 month-old cross-bred sheep were orally infected, each with 150 metacercariae of *F. hepatica*. The animals were divided into 12 groups of 5 each. Groups 1, 3, 5, 7, 9, and 11 with 3-d, 2-week, 4-week, 6-week, 8-week and 10-week-old flukes, respectively, were orally treated with 15 mg of **6** in a freshly prepared suspension,<sup>20,21)</sup> while groups 2, 4, 6, 8, 10, and 12 remained as untreated controls. Four weeks after treatment, the animals were killed in order to collect and count the flukes in the liver. Efficacy (Table 3) was assessed as a percentage of fluke reduction in the treated groups in comparison with the control according to the formula of Forey.<sup>22)</sup>

$$\% \text{ efficacy} = \frac{A-B}{A} \times 100$$

Where *A* is the mean number of flukes in the control group; *B* is the mean number of flukes in the treated group.

## Results and Discussion

The synthesis of compound **3** implies a nucleophilic substitution which can be carried out either by melting **1** and **2** in the presence of KOH,<sup>23)</sup> or by heating the mixture at 115 °C in a polar solvent (*N,N*-dimethylformamide (DMF)), and a base ( $K_2CO_3$ ).<sup>14)</sup> We used the second method; which gave higher yield % of the desired product, however, it was severely decreased by recrystallization. Washing the crude product with cold solvent gave a product of high quality and a good yield %. Compound **4** was obtained in good yield %, but the crude amine had to be stabilized with acid to avoid a fast decomposition. Although it was possible to isolate **4**, we preferred to use it immediately in the next reaction. The preparation of 2-mercaptobenzimidazoles is also a known process;<sup>16,17)</sup> it was adapted here to obtain **5**. Although this compound had a poor solubility in most organic solvents, it was possible to purify it by recrystallization from AcOEt–EtOH. The reaction to obtain **6** was very fast. In this final

Table 1. Biological Activity *in Vitro* of **6** against *F. hepatica* Metacercariae<sup>a)</sup>

Concentration ( $\mu M$ )	(% Efficacy)	
	Compound <b>6</b>	Diamphenetide deacetylated
146.7	100 <sup>b)</sup>	nd
34.68 <sup>c)</sup>	nd	100
29.34 <sup>c)</sup>	100 <sup>b)</sup>	nd
9.68	77.5	nd
3.81	nd	100 <sup>d)</sup>
3.22	0.0	nd
1.08	0.0	100 <sup>d)</sup>

a) Experiment was carried out in triplicate. b) Caused lysis of the metacercariae. c) Dose equivalent to 10  $\mu g/ml$ . d) Ref. 19. nd=not determined.

Table 2. Efficacy of **6** against 10 Week-Old *F. hepatica* in Experimentally Infected Sheep at Two Dose Levels

Group (n=9)	Dose (mg/kg/bw) <sup>a)</sup> per os	Efficacy (%)
1	10	80.6
2	15	86.9
3	Non treated control	0.0

a) Body weight.

Table 3. Efficacy of **6** against *F. hepatica* of Different Ages in Experimentally Infected Sheep

Group (n=5)	Age of <i>F. hepatica</i> at the moment of treatment	Efficacy (%) at 15 mg/kg <sup>a)</sup>
1	3 d	100
2	Control	0
3	2 weeks	100
4	Control	0
5	4 weeks	100
6	Control	0
7	6 weeks	100
8	Control	0
9	8 weeks	100
10	Control	0
11	10 weeks	100
12	Control	0

a) Previously formulated.<sup>21)</sup>

step, the amount of reagent and the temperature were carefully controlled; otherwise dimethylation products were formed.

The *in vitro* assay (Table 1) showed that compound **6** killed early immature flukes (1–4 d) at concentrations of 29.34–9.68  $\mu\text{M}$  (77.5%). These results motivated us to carry out *in vivo* tests.

The preliminary *in vivo* assay (Table 2) showed that compound **6** had an efficacy of 86.90% at a dose of 15 mg/kg against 10-week-old flukes. However, compound **6** was not formulated in our assay; this fact might as well have reduced significantly the efficacy.

The third trial showed that compound **6** given in an oral suspension was 100% effective (Table 3) against all stages of *F. hepatica* (3 d–12 weeks) at a dose of 15 mg/kg.

## Conclusions

The substitution of the 1-naphthyl group for the 2,3-dichlorophenyl group in TCBZ gives a bioisostere of this potent fasciolicide. The *in vitro* assay showed that compound **6** is very active and capable of disrupting the metacercariae. Compound **6** showed good activity when it was administered in a suspension. At a dose of 15 mg/kg it was 100% effective against 3-d and 10-week-old flukes. The results so far obtained are promising, since most fasciolicidal agents such as rafoxanide, nitroxylin, closantel or clorsulon act only against 6-week-old flukes or older; or at the immature stage, as diamphenetide.<sup>1)</sup> Further studies are in progress in order to get a better formulation and to optimize the dose. The determination of the efficacy of **6** in cattle is in progress as well.

## Experimental

Melting points were determined on a Büchi B-540 melting point apparatus and are uncorrected. Reactions were monitored by TLC on 0.2 mm pre-coated silica gel 60 F<sub>254</sub> plates (E. Merck). IR spectra were recorded on a Perkin Elmer FT-IR-1600 spectrometer as KBr disks. <sup>1</sup>H-NMR spectra were measured with a Varian EM-390 (300 MHz) spectrometer. Chemical shifts are given in ppm relative to Me<sub>4</sub>Si ( $\delta=0$ ) in DMSO-*d*<sub>6</sub>. *J* values are given in Hz. The following abbreviations are used: s, singlet; d, doublet; dd, doublet of doublet; t, triplet; m, multiplet; br, broad. MS were recorded on a JEOL JMS-SX102A spectrometer by electron impact (EI) low and high resolution (HR). Elemental analyses were carried out on a Fisons EA11108 CHNSO analyzer. Starting material **1** was synthesized in our laboratory from the commercially available 3,4-dichloroaniline via acetylation, nitration and hydrolysis of the nitroacetanilide.

**4-Chloro-5-(1-naphthoxy)-2-nitroaniline (3)** A suspension of **1** (100 g, 0.483 mol), **2** (70.33 g, 0.487 mol), K<sub>2</sub>CO<sub>3</sub> (100 g, 0.724 mol), and DMF (360 ml) was stirred at 115 °C for 3.5 h. Then the mixture was cooled at room temperature and filtered by suction. The residue was washed with hot DMF (300 ml) and the solvent removed *in vacuo*. The crude product was washed with cold methanol (300 ml) twice, and the orange solid was airdried to give 108.0 g (71%) of essentially pure **3**. An analytical sample was recrystallized from toluene–ethanol affording 98% of yellow crystals. mp 145–146 °C. IR (KBr) cm<sup>-1</sup>: 3462, 3347, 1628, 1560, 1387, 1320, 1225. <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>)  $\delta$ : 6.22 (1H, s, H-6), 7.37 (1H, dd, *J*<sub>1</sub>=7.5 Hz, *J*<sub>2</sub>=1.0 Hz, H-2'), 7.44 (2H, br, NH<sub>2</sub>), 7.55–7.64 (3H, m, H-3, H-6', H-7'), 7.79 (1H, dd, *J*<sub>1</sub>=8.0 Hz, *J*<sub>2</sub>=2.4 Hz, H-5'), 7.93 (1H, d, *J*=8.4 Hz, H-4'), 8.05 (1H, dd, *J*<sub>1</sub>=7.2 Hz, *J*<sub>2</sub>=1.8 Hz, H-8'), 8.18 (1H, s, H-3). <sup>13</sup>C-NMR (DMSO-*d*<sub>6</sub>)  $\delta$ : 103.59 (C-6), 109.54 (C-4), 117.14 (C-2'), 120.73 (C-5'), 125.69 (C-8'a), 125.79 (C-4'a), 129.99 (C-4'), 126.26 (C-6'), 127.06 (C-3'), 127.12 (C-3), 127.15 (C-7'), 128.26 (C-8'), 134.77 (C-2), 146.91 (C-1), 149.14 (C-1'), 159.04 (C-5). MS EI *m/z* 314 (M<sup>+</sup>), 279, 233. HR-MS (EI) Calcd for C<sub>16</sub>H<sub>11</sub>ClN<sub>2</sub>O<sub>3</sub> (M<sup>+</sup>) *m/z*: 314.0458. Found: 314.0458. Anal. Calcd for C<sub>16</sub>H<sub>11</sub>ClN<sub>2</sub>O<sub>3</sub>: C, 61.06; H, 3.52; N, 8.90. Found: C, 61.18; H, 3.58; N, 8.62.

**4-Chloro-5-(1-naphthoxy)-1,2-phenylenediamine (4)** A mixture of **3** (100 g, 0.317 mol), SnCl<sub>2</sub>·2H<sub>2</sub>O (430 g, 1.9 mol), and EtOH (640 ml) was stirred at 70 °C for 3 h in a nitrogen atmosphere. After cooling, the mixture

was basified (pH=9) with a 50% NaOH solution. The solvent was removed carefully *in vacuo*, and the solid residue was extracted with AcOEt (200 ml×3). The combined organic extracts were washed with brine, dried with anhydrous Na<sub>2</sub>SO<sub>4</sub> and concentrated *in vacuo* to give 89.6 g (99.08%) of **4** as an oil, which was immediately cyclocondensed in the next step. An analytical sample, which was dissolved in anhydrous ethanol, was converted into the monohydrochloride by treatment with HCl gas; the solid formed was separated *in vacuo*, washed with ethanol and recrystallized from ethanol to give yellow crystals. mp 195–197 °C. IR (KBr) cm<sup>-1</sup>: 3461, 3345, 1629, 1493, 1227, 766. <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>)  $\delta$ : 3.80 (4H, br, 2NH<sub>2</sub>), 6.50 (1H, s, H-6), 6.89 (1H, dd, *J*<sub>1</sub>=7.6, *J*<sub>2</sub>=0.9, H-2'), 7.53 (1H, s, H-3), 7.47 (1H, t, *J*<sub>1</sub>=8.1, *J*<sub>2</sub>=7.8 Hz, H-3'), 7.50–7.61 (2H, m, H-6', H-7'), 7.73 (1H, d, *J*=8.4 Hz, H-4'), 7.95–8.02 (2H, m, H-5', H-8'). <sup>13</sup>C-NMR (DMSO-*d*<sub>6</sub>)  $\delta$ : 108.51 (C-6), 112.38 (C-4), 112.90 (C-2'), 119.10 (C-8'a), 121.12 (C-8'), 123.67 (C-3), 123.82 (C-4'), 125.39 (C-4'a), 126.13 (C-3'), 126.52 (C-6'), 126.93 (C-7'), 127.98 (C-5'), 134.59 (C-2), 137.96 (C-1), 150.16 (C-1'), 151.58 (C-5). MS (EI) *m/z*: 284 (M<sup>+</sup>), 232. HR-MS (EI) Calcd for C<sub>16</sub>H<sub>13</sub>ClN<sub>2</sub>O (M<sup>+</sup>) *m/z*: 284.0716. Found: 284.0716. Anal. Calcd for C<sub>16</sub>H<sub>13</sub>ClN<sub>2</sub>O·HCl: C, 59.83; H, 4.39; N, 8.72. Found: C, 59.80; H, 4.38; N, 8.67.

**5-Chloro-2-mercapto-6-(1-naphthoxy)-1H-benzimidazole (5)** A solution of **4** (50 g, 0.175 mol) in 250 ml of EtOH:H<sub>2</sub>O (80:20), KOH (14.77 g, 0.263 mol), and CS<sub>2</sub> (15.84 ml, 20.05 g) was stirred at 70 °C for 3 h under a N<sub>2</sub> atmosphere. During this time a yellow precipitate was formed. The cold mixture was poured into 600 ml of H<sub>2</sub>O and treated with 20% AcOH solution to a pH 5–6. The precipitate was separated by suction filtration, washed with water and dried in an oven at 90 °C. The crude product was purified by recrystallization from AcOEt–EtOH and activated carbon twice to give 41.82 g (73%) of **5** as a white powder. mp 273–275 °C. IR (KBr) cm<sup>-1</sup>: 3380, 3053, 1460, 1391, 1328, 1229, 1157. <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>)  $\delta$ : 6.75 (1H, dd, *J*<sub>1</sub>=6.6, *J*<sub>2</sub>=2.0, H-2'), 6.84 (1H, s, H-7), 7.34 (1H, s, H-4), 7.40 (1H, t, *J*<sub>1</sub>=8.1 Hz, *J*<sub>2</sub>=8.1 Hz, H-3'), 7.54–7.62 (2H, m, H-6', H-7'), 7.68 (1H, d, *J*=8.4 Hz, H-4'), 7.95–8.00 (1H, m, H-5'), 8.13–8.17 (1H, m, H-8'), 12.60 (1H, s, NH or SH), 12.72 (1H, s, NH or SH). <sup>13</sup>C-NMR (DMSO-*d*<sub>6</sub>)  $\delta$ : 102.27 (C-7), 110.60 (C-2'), 110.95 (C-4'), 118.96 (C-5), 121.19 (C-6'), 123.04 (C-4), 124.97 (C-8'a), 126.01 (C-8'), 126.31 (C-7'), 126.89 (C-5'), 127.82 (C-3'), 129.57 (C-4'a), 132.04 (C-3a), 134.46 (C-7a), 147.10 (C-1'), 152.66 (C-6), 169.66 (C-2). MS (EI) *m/z* 326 (M<sup>+</sup>), 291, 290. HR-MS (EI) Calcd for C<sub>17</sub>H<sub>11</sub>ClN<sub>2</sub>OS (M<sup>+</sup>) *m/z*: 326.0280. Found: 326.0281. Anal. Calcd for: C<sub>17</sub>H<sub>11</sub>ClN<sub>2</sub>OS: C, 62.48; H, 3.39; N, 8.57; S, 9.81. Found: C, 62.49; H, 3.29; N, 8.44; S, 9.93.

**5-Chloro-2-methylthio-6-(1-naphthoxy)-1H-benzimidazole (6)** A suspension of 100 g (0.3059 mol) of **5** in acetone (300 ml) was treated with a solution of KOH (18.8 g, 0.336 mol) in water (50 ml), at room temperature, under a N<sub>2</sub> atmosphere. The dark solution formed was cooled to 0–5 °C and treated slowly (1 h) with a solution of CH<sub>3</sub>I (43.41 g, 0.3059 mol) in acetone (43 ml). The mixture was stirred at 10 °C for 30 min, neutralized with 20% HCl solution, and the solvent was eliminated *in vacuo*. The solid residue was taken up with 400 ml of AcOEt, washed with brine, dried with anhydrous Na<sub>2</sub>SO<sub>4</sub>, and the solvent was evaporated up to 200 ml. To the concentrated solution was added an equal volume of methanol and it was stirred in a cold bath. The resulting precipitate was collected, washed with methanol and dried. The crude product was recrystallized from CHCl<sub>3</sub>–MeOH to give 86.22 g (82.70%) of **6** as white powder. mp 192–195 °C. IR (KBr) cm<sup>-1</sup>: 3402, 2720, 1453, 1316, 1157. <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>)  $\delta$ : 2.84 (3H, s, S–CH<sub>3</sub>), 6.48 (1H, br, NH), 6.77 (1H, d, *J*=7.8 Hz, H-2'), 7.29 (1H, s, H-4), 7.40 (1H, t, *J*<sub>1</sub>=7.8 Hz, *J*<sub>2</sub>=8.1 Hz, H-3'), 7.53–7.66 (2H, m, H-6', H-7'), 7.70 (1H, d, *J*=8.1 Hz, H-4'), 7.89 (1H, s, H-7), 7.94–8.20 (1H, m, H-5'), 8.12–8.22 (1H, m, H-8'). <sup>13</sup>C-NMR (DMSO-*d*<sub>6</sub>)  $\delta$ : 13.46 (S–CH<sub>3</sub>), 102.95 (C-7), 110.20 (C-2'), 113.16 (C-4), 119.70 (C-8'), 120.83 (C-5), 122.04 (C-4'), 123.80 (C-8'a), 124.12 (C-3'), 124.66 (C-6'), 125.25 (C-7'), 126.21 (C-5'), 128.72 (C-4'a), 131.34 (C-3a), 133.08 (C-7a), 148.05 (C-1'), 150.75 (C-2), 152.03 (C-6). MS (EI) *m/z* 340 (M<sup>+</sup>), 305, 290. HR-MS (EI) Calcd for C<sub>18</sub>H<sub>13</sub>ClN<sub>2</sub>OS (M<sup>+</sup>) *m/z*: 340.0437. Found: 340.0437. Anal. Calcd for C<sub>18</sub>H<sub>13</sub>ClN<sub>2</sub>OS: C, 63.43; H, 3.84; N, 8.22; S, 9.41. Found: C, 62.82; H, 3.69; N, 7.97; S, 9.86.

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## References and Notes

- 1) Fairweather I., Boray J. C., *Vet. J.*, **158**, 81—112 (1999).
- 2) Chen M. G., Mott K. E., *Trop. Dis. Bull.*, **1990**, R1—R37 (1990).
- 3) Boray J. C. "Immunology, Pathobiology and Control of Fasciolosis," ed. by Boray J. C., MSD AGVET Rahway, New Jersey, 1997, pp. 83—89.
- 4) Thorpe P. J., *Drugs Future*, **9**, 593—594 (1984).
- 5) Boray J. C., Crowfoot P. D., Strong M. B., Allison J. R., Schellenbaum M., Von Orelli M., Sarisin G., *Vet. Rec.*, **113**, 315—317 (1983).
- 6) Wolf K., Eckert J., Schneider G., Lutz H., *Vet. Parasitol.*, **13**, 145—150 (1983).
- 7) Craig T. M., Huey R. L., *Am. J. Vet. Res.*, **45**, 1644—1645 (1984).
- 8) Arjona R., Riancho J. A., Aguado J. M., Salesa R., Gonzalez-Macias J., *Medicine* (Baltimore), **74**, 13—23 (1995).
- 9) Apt W., Aguilera X., Vega F., Miranda C., Zulantay I., Perez C., Gabor M., Apt P., *Am. J. Trop. Med. Hyg.* **52**, 532—535 (1995).
- 10) el-Karakasy H., Hassanein B., Okasha S., Behairy B., Gadallah I., *J. Trop. Pediatr.*, **45**, 135—138 (1999).
- 11) Ibarra-Velarde F., García-Sánchez E., Fernández-Ruvalcaba M., Vera-Montenegro Y., Castillo R., Hernández-Campos A., *Vet. Méx.* **28**, 291—296 (1997).
- 12) Ibarra-Velarde F., Vera-Montenegro Y., Hernández-Campos A., Castillo R., *R. Vet. Méx.* **28**, 297—301 (1997).
- 13) Black J. W., Stephenson J. S. *Lancet*, **ii**, 311—314 (1962).
- 14) Averkin E. A., Beard C. C., Dvorak C. A., Edwards J. A., Fried J. H., Kilian J. G., Schiltz R. A., Kistner T. P., Drudge J. H., Lyons E. T., Sharp M. L., Corwin R. M. J., *J. Med. Chem.*, **18**, 1164—1166 (1975).
- 15) Manjarrez N., Luna H. M., Pérez H. I., Soria O., *Rev. Soc. Quím. Méx.*, **30**, 52—54 (1986).
- 16) Van Allan J. A., Deacon B. D., "Organic Syntheses Collective Volume," Vol. IV, ed. by Rabjohn N., John Wiley and Sons, N.Y., 1976, pp. 569—570.
- 17) Ciba-Geigy A. G., Belg. Pat. 865870 (1978) [*Chem. Abstr.*, **90**, 137817 z (1979)].
- 18) Ibarra O. F., Jenkins D. C., *Z. Parasitenk.*, **70**, 655—671 (1984).
- 19) Sánchez E. B., Ibarra V. F., *Tec. Pec. Méx.*, **25**, 397—403 (1988).
- 20) Ibarra-Velarde F., Montenegro-Cristino N., Flores-Crespo J., Hernández-Campos A., Castillo R., *Vet. Méx.*, **31**, 47—51 (2000).
- 21) The freshly prepared suspension contained: compound **6** (10 g), pectin (1.5 g), low viscosity carboxymethylcellulose (0.5 g), medium viscosity carboxymethylcellulose (0.5 g), USP saccharine (0.2 g), methylparaben (0.5 g), propylparaben (0.03 g), and water to make 100 ml.
- 22) Forey W. S., *Vet. Parasitol.*, **26**, 265—271 (1988).
- 23) Frick W. E., Weiss A. G., Wenger T., Traber W., U.S. Pat. 3506767 (1968) [*Chem. Abstr.*, **69**, 37061w (1968)].
- 24) Bennett J. L., Köhler P., *Exp. Parasitol.*, **63**, 49—57 (1987).