possible that the β-ecdysone isolated was not elaborated by the nematode but was obtained from vegetable material ingested by the parasite from the intestine of the host animal. However, this seems unlikely and the possibility that ecdysones play a role in nematode development now warrants further investigation.

Résumé. Une hormone de mue des arthropodes, très probablement la β-ecdysone, a été trouvée dans un extrait du nématode parasite *Ascaris lumbricoides*, obtenus des intestins du cochons.

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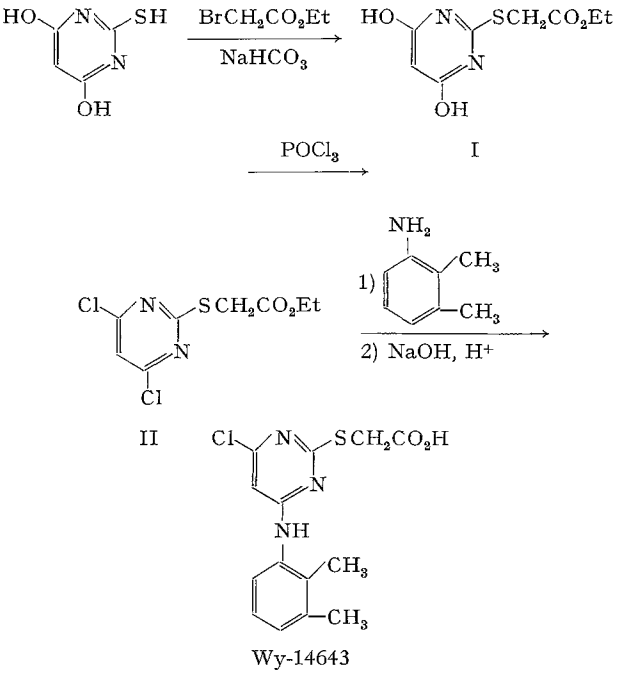
A Potent Antihypercholesterolemic Agent: [4-Chloro-6-(2,3-xylidino)-2-pyrimidinylthio]acetic Acid (Wy-14643)

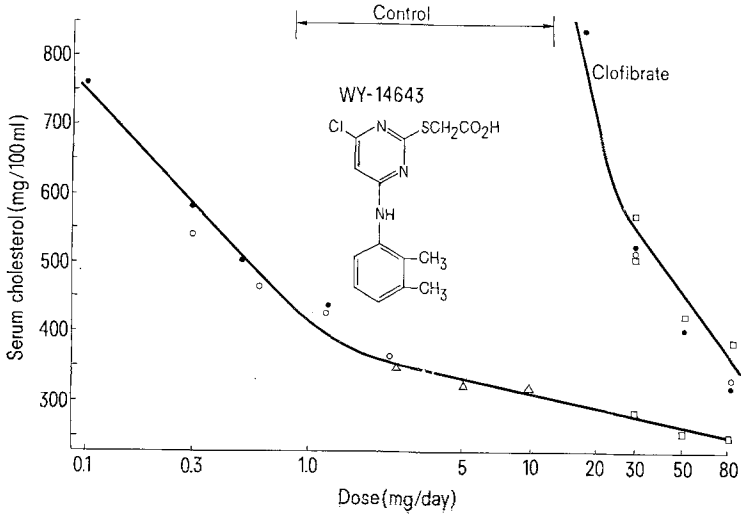
The search for more efficacious drugs to control hypercholesterolemia, a major risk factor in coronary heart disease, has been intensified over the past decade. One of the most widespread agents used in the clinical management of hyperlipidemia is ethyl *p*-chlorophenoxyisobutyrate (clofibrate)[1]. A number of 2-pyrimidinylthioacetic acid derivatives recently synthesized in this laboratory were compared with clofibrate for ability to lower the serum cholesterol in rats with hypercholesterolemia of dietary origin. The pertinent points of this assay procedure have previously been described by one of us[2]. Since these pyrimidine derivatives represent a novel class of hypocholesterolemic agents, it is the purpose of this communication to outline the synthesis and biological activity of one of the more potent and interesting members of the series, [4-chloro-6-(2,3-xylidino)-2-pyrimidinylthio]acetic acid (Wy-14643).

The scheme illustrates the route used to prepare the title compound. The alkylation of sodium 2-thiobarbiturate with an equivalent of ethyl bromoacetate in aqueous ethanol at 60° for 1.5 h gave a 72% yield of I: m.p. 194–197°/ethanol; analysis for $C_8H_{10}N_2O_4S$, Calc: C, 41.37; H, 4.38; N, 12.16. Found: C, 41.75; H, 4.46; N, 12.27.

Ethyl (4,6-dichloro-2-pyrimidinylthio)acetate (II) was obtained in 71% yield by treating I in boiling phosphorus oxychloride containing N,N-diethylaniline for 4 h. Removal of the phosphorus oxychloride and treatment of the residue with ice gave II: m.p. 61–62°/petroleum ether; analysis for $C_8H_8Cl_2N_2O_2S$, Calc: C, 35.97; H, 3.02; N, 10.48. Found: C, 35.95; H, 3.00; N, 10.30.

Treatment of II with an equivalent of 2,3-dimethylaniline and sodium carbonate in boiling ethanol for 4 h, followed by filtration, and addition of water to the filtrate gave 61% yield of ethyl [4-chloro-6-(2,3-xylidino)-2-pyrimidinylthio]acetate: m.p. 87–91°/ethanol (95%); analysis for $C_{16}H_{18}ClN_3O_2S$, Calc: C, 54.62; H, 5.15; N, 11.94. Found: C, 54.80; H, 5.08; N, 12.00.





Effect of Wy-14643 and clofibrate on the serum cholesterol of hypercholesterolemic male rats (250–350 g body weight). The data from 4 bioassays are combined; values represented by a given symbol are from the same bioassay.

Hydrolysis was effected by warming for a few min a solution of the ester in 30% sodium hydroxide solution containing ethanol for solubilization. Acidification gave Wy-14643 in 69% yield: m.p. 150–153°/ethyl acetate; analysis for $C_{14}H_{14}ClN_3O_2S$, Calc: C, 51.93; H, 4.36; N, 12.98. Found: C, 52.01; H, 4.41; N, 12.99.

A structure-activity study has been carried out and will be reported in greater detail in a subsequent paper.

Biological activity. The hypocholesterolemic response of Wy-14643, like that of clofibrate, was linear when plotted against log dose. The dose-response curves for both agents are shown in the Figure. Because of the non-parallel relationship between the two curves, relative potencies were dose dependent, e.g.

Dose (mg/rat/day)	Reduction of cholesterol (%)	Activity vs clofibrate
50	68	1.9×
1	50	60×
0.1	12	180×

A more extensive evaluation of this drug is currently in progress.

Zusammenfassung. Es wird die Synthese von [4-Chlor-6-(2, 3-xylidino)-2-pyrimidinylthio]-essigsäure und seine antihypercholesterolämische Aktivität beschrieben.

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Diurnal Rhythm of Choline-^{14}C Incorporation into Lecithin in Ehrlich-Lettré Ascites Tumour Cells

A diurnal rhythm, especially in the concentration of plasma[1] and liver fatty acids[2], in the level of serum triglycerides[3] and phospholipids[4], and in compounds other than lipids[5,6], has been observed in recent years. Generally, the diurnal rhythm can be looked upon as an expression of the fluctuation of the metabolic activity of a given organism. The present study describes such a rhythm for the choline-^{14}C incorporation into lecithin in Ehrlich-Lettré ascites tumour cells (EAT). A preliminary note has been presented elsewhere[7].

During the course of our studies on choline transport and its conversion into lecithin in ascites cells[8], we observed marked differences of ^{14}C-labelled choline incorporation into lecithin with a few hours time interval between killing the animals. Therefore, we began to follow up the ^{14}C-incorporation over a 24-h period in combination with a concomitant thymidine-^{3}H incorporation into DNA, since it has been argued that the mitotic index might also show a rhythmic pattern.

Methods. For these studies, the glycogen-free strain of the hyperdiploid Ehrlich-Lettré mouse ascites tumour 7 days after transplantation was used. On the day of inoculation, randomly selected mice were put in cages in groups of 10 animals. Lab chow diet was given ad libitum and water renewed at least every other day. In the case of choline-free diet, the feed was given as a slurry mixed with water, which was prepared fresh every day and given at 09.00. Every 3rd day the mice were put into fresh cages. The animals were kept under 12–14 h of daylight from 06.00 to 20.00 h without any artificial lighting or darkening treatment. Usually 3 animals were killed every 4 h for a period of 24 h and radioactivity

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Table I. Analysis of total lipid phosphorus and of lecithin phosphorus after TLC-separation of an aliquot of the lipid extract corresponding to 2×10^6 cells

	Time of the day	Lipid phosphorus[a] (nmoles)	Lecithin phosphorus[a] (nmoles)	Difference probability for lecithin (P)
Experiment 1	24.00–09.00	164.1 ± 80.5	78.7 ± 28.1	< 0.01
	12.00–21.00	173.1 ± 78.0	49.1 ± 18.9	
Experiment 2	24.00–09.00	200.6 ± 29.8	75.4 ± 7.1	< 0.01
	12.00–21.00	214.0 ± 51.1	46.3 ± 23.4	

[a] Mean values plus SD from 12 (experiment 1) and 9 (experiment 2) animals for each time interval. Student's t-test gave t-values of 3.03 (experiment 1) and of 3.37 (experiment 2).