Effects of Sch 9122 HCl, a Hypocholesterolemic Compound, on Mammary Tissue, Libido, and Fertility of Male Rats (34863)

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It has been reported (1) that Sch 9122 HCl (2-(*p*-anisyl)-3-(2-pyridyl)-pentane hydrochloride), a compound structurally related to diethylstilbestrol, lowers serum cholesterol when administered orally to rats. The oral dose which lowers cholesterol by 30% (ED_{30} cholesterol) in both normal male and ovariectomized rats is approximately 5 mg/kg. At the ED_{30} cholesterol, the compound did not reduce seminal vesicle weights and produced no estrogenic stimulation of the uterus or vagina. Sch 9122 HCl was much less estrogenic than conjugated equine estrogen, ethinyl estradiol, or diethylstilbestrol at comparable hypocholesterolemic doses. However, the predominant side-effects that limit the clinical usefulness of estrogenic compounds for the reduction of serum cholesterol are swelling and tenderness of the breast and adverse effects on libido and sexual performance. It was, therefore, decided to compare directly in the rat the effects of chronically-administered Sch 9122 HCl and several estrogens on pertinent activities which might reflect these undesirable side effects: mammary development, mating behavior, and fertility. The results of these investigations are reported below.

Methods. Compounds were administered daily by gavage to normal adult male Charles River rats, CD strain, at doses (ED_{30}) that produced approximately equal hypocholesterolemic responses in acute experiments:

Expt. 1 (75 days)

Treatment	Dose (mg/kg, po)	No. of rats
Sch 9122 HCl	10.0	6
Ethinyl estradiol	0.05	8
Diethylstilbestrol	0.20	6
Controls	—	10

Expt. 2 (100 days)

Treatment	Dose (mg/kg, po)	No. of rats
Sch 9122 HCl	5.0	7
Conjugated equine estrogen	0.5	5
Controls	—	9

The compounds were suspended in an aqueous medium containing carboxymethyl cellulose (0.05%), benzyl alcohol (0.5%), and Tween 80 (0.25%). The controls received the vehicle alone.

After 44 and 65 days in Exp. 1, and after 41 and 90 days in Expt. 2, the male rats were housed individually for 8 days with randomly selected adult female Charles River rats during which time drug treatment was continued. Daily vaginal smears were inspected for the presence of sperm. Females which had mated the previous evening were considered to be in the first day of pregnancy. The uteri were inspected for nidation sites on day 8 of pregnancy as evidence for male fertility. At the end of the treatment period, *i.e.*, 75 or 100 days, the male rats were sacrificed and the testes, prostate, and seminal vesicles were removed and weighed to the nearest milligram. In Expt. 2, these organs were placed in 10% formalin and saved for subsequent histological examination (hemotoxylin and eosin). Samples of subcutaneous fat from the mammary region were also removed in Expt. 2, and the degree of estrogenic stimulation of mammary gland growth or development was determined histologically.

Serum cholesterol (2) and blood glucose (3) were determined using the Technicon AutoAnalyser.

A one-way, completely randomized analy-

TABLE I. Effects of Sh 9122 HCl, Ethinyl Estradiol, and Diethylstilbestrol on Body and Organ Weights, Serum Cholesterol, Blood Glucose, and Fertility of Male Rats (Expt. 1, 75 days).[a]

Treatment (no. of rats)	Dose (mg/kg, po)	Mean (± SE)						% Mating		% Pregnant	
		Wt (g)				Serum cholesterol (mg/100 ml)	Blood glucose (mg/100 ml)	44 days	65 days	44 days	65 days
		Final body	Testes	Seminal vesicles	Prostate						
Sch 9122 HCl (6)	10	432[c] (29.5)	3.20 (0.102)	0.55[c] (0.033)	1.11[c] (0.068)	67[c] (10.7)	134 (3.6)	67	83	67	83
Ethinyl estradiol (8)	0.05	444[c] (12.1)	3.20 (0.134)	0.40[c] (0.033)	0.70[c] (0.088)	77[b] (5.1)	132 (3.9)	25	63	25	50
Diethylstilbestrol (6)	0.20	385[c] (20.6)	2.30[c] (0.332)	0.25[c] (0.047)	0.38[c] (0.089)	76[b] (11.8)	136 (3.7)	14	33	0	33
Controls (10)	—	540 (17.9)	3.60 (0.105)	0.73 (0.044)	1.57 (0.099)	103 (4.8)	135 (4.5)	75	75	75	75

[a] Experimental details in Methods section.

[b] Significantly different from control value, $p<0.05$; [c] $p<0.01$.

sis of variance was used to analyze the differences among the groups (4). If significant differences were indicated, the treatment groups were compared to the controls using Dunnett's t statistic.

Results. Sch 9122 HCl decreased serum cholesterol by 35% at a dose of 10 mg/kg in Expt. 1 (Table I) and by 23% at 5 mg/kg in Expt. 2 (Table II). Ethinyl estradiol (0.05 mg/kg), diethylstilbestrol (0.20 mg/kg), and conjugated equine estrogen (0.5 mg/kg) produced comparable hypocholesterolemic responses at the doses used.

All treatments retarded the rate of weight gain of the animals. Testis weight was reduced by diethylstilbestrol and conjugated equine estrogen, but not by ethinyl estradiol or Sch 9122 HCl. In Expt. 2, where histological examination was made, conjugated equine estrogen produced marked atrophic changes in the testes, seminal vesicles, and prostate gland. At 5 mg/kg, Sch 9122 HCl had no significant effect on seminal vesicle and prostate weight or histology. Though the absolute weight of these organs was reduced at 10 mg/kg (Table I), if the organ weights are calculated as grams per 100 grams of body weight, the seminal vesicle and prostate weights do not differ significantly from the control values. Both Sch 9122 HCl and conjugated equine estrogen reduced the weight of the epididymal fat pad in Expt. 2 where this parameter was measured.

In Expt. 2, where mammary tissue was examined, Sch 9122 HCl had no significant influence upon mammary histology in any of the treated animals whereas conjugated equine estrogen produced hyperplasia in three of five treated animals and stimulation of duct development in one of the remaining rats. When compared to the control animals, Sch 9122 HCl had no effect on mating frequency or fertility in either experiment. All three estrogens (*i.e.*, ethinyl estradiol, diethylstilbestrol, and conjugated equine estrogen) reduced the number of males that mated and therefore the number of females that became pregnant.

Discussion. In a previous report (1), we demonstrated that Sch 9122 HCl is an effective cholesterol-lowering agent that is less es-

TABLE II. Effects of Sch 9122 HCl and Conjugated Equine Estrogen on Body and Organ Weights, Serum Cholesterol and Fertility of Male Rats (Expt. 2, 100 days).[a]

Treatment (no. of rats)	Dose (mg/kg, po)	Mean (± SE)						% Mating		% Pregnant	
		Wt (g)					Serum cholesterol (mg/100 ml)	41 days	90 days	41 days	90 days
		Final body	Testes	Seminal vesicles	Prostate	Epididymal fat pad (g)					
Sch 9122 HCl (7)	5.0	476 (13.0)	3.54 (0.248)	0.66 (0.077)	1.67 (0.233)	5.91[b] (0.36)	66[b] (8.2)	100	88	88	75
Conjugated equine estrogen (5)	0.5	394[b] (26.4)	1.29[c] (0.428)	0.17[c] (0.044)	0.42[b] (0.100)	4.57[c] (0.62)	65[b] (8.7)	25	40	13	20
Controls (9)	—	554 (18.3)	3.79 (0.117)	0.67 (0.055)	2.05 (0.138)	8.48 (0.78)	86 (4.4)	78	67	78	67

[a] Experimental details in Methods section.
[b] Significantly different from control value, $p<0.05$; [c] $p<0.01$.

trogenic than standard compounds. In short-term experiments, the ED_{30} cholesterol (5 mg/kg) produced no estrogenic stimulation of the uterus or vagina and had no effect on seminal vesicle or prostate weights. In the present experiments with Sch 9122 HCl, we have demonstrated that the reduction in serum cholesterol is maintained on prolonged administration and that there is no cumulative estrogenic effect over the relatively long time periods studied (75 and 100 days). Male rats treated with Sch 9122 HCl mated as frequently as control animals (*i.e.*, there was no apparent effect on libido) and were fertile. Furthermore, there was no evidence of estrogenic stimulation of the mammary gland. We have shown, therefore, that it is possible to obtain prolonged reductions in serum cholesterol levels without evidence of estrogenicity. We have observed, in earlier short-term experiments as well as now, that it is not possible to obtain hypocholesterolemic activity with any of the standard estrogens, including conjugated equine estrogen, without potent estrogenic side-effects. To this extent, Sch 9122 HCl may represent a significant advance in drug therapy.

Although the mechanism of the hypocholesterolemia is not known, Sch 9122 HCl has been observed (unpublished observations) to have a marked effect on fat synthesis as indicated by its ability to decrease the incorporation of glucose into the epididymal fat pad. This inhibition of lipogenesis may explain the reductions in epididymal fat pad and body weights that were observed and will be studied further.

Summary. Sch 9122 HCl (2-(*p*-anisyl)-3-(2-pyridyl)-pentane hydrochloride), a compound structurally related to diethylstilbestrol, had significant hypocholesterolemic activity on prolonged administration to male rats. At oral doses which lowered serum cholesterol by as much as 30%, Sch 9122 HCl did not produce estrogenic effects on secondary sex structures, stimulate mammary tissue, or have adverse effects on mating behavior and fertility. Standard estrogenic substances had significant effects on these parameters at comparable hypocholesterolemic doses.

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