this plant is non-preferred or indeed untouched in the field by such insects. Its potential as an applied antifeedant in crop protection requires further work on other insect species, on possible systemic uptake by plants and on its stability in field conditions. It is possible that other iridoids exhibit similar activity as they are structurally very similar.

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Observations on salicyl hydroxamic acid, an experimental trypanocide

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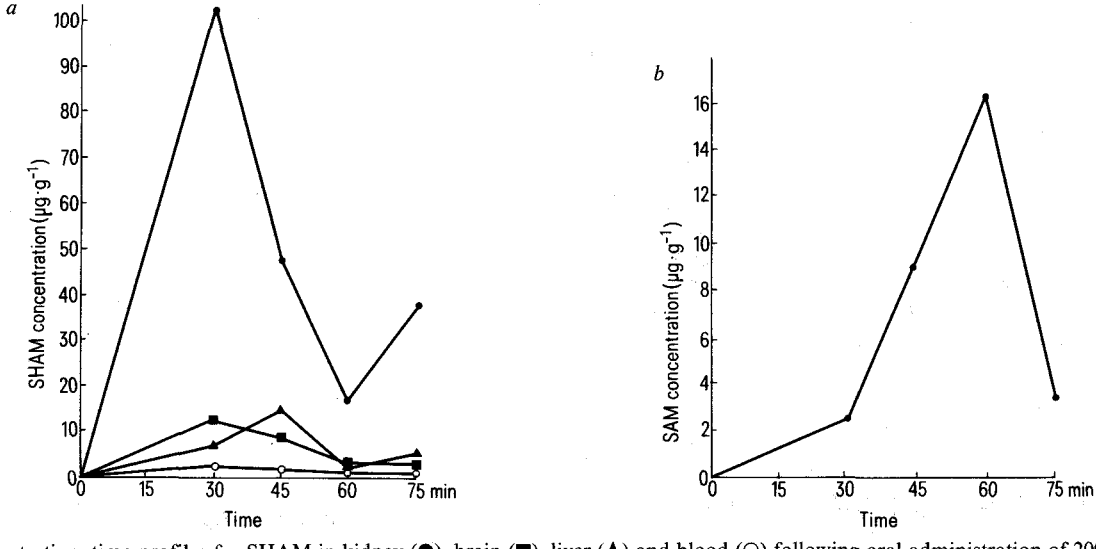
Summary. The inferior in vivo efficacy of salicyl hydroxamic acid against trypanosomiasis may be explained by its short half-life, high degree of protein binding and low tissue levels.

Salicyl hydroxamic acid (2-hydroxybenzhydroxamic acid; SHAM) has been used as an experimental trypanocide since it is a potent inhibitor of the respiration of bloodstream forms of *Trypanosoma brucei* in vitro[1]. Its therapeutic effect in infected rats is less marked[2], both as a single agent and in combination with glycerol, another potent in vitro inhibitor of anaerobic glycolysis in *T. brucei*. Permanent cures in rats infected with *T. brucei* were obtained using a dosage regime which was just sublethal[3]. The plasma concentration of SHAM has been shown to fall rapidly after stopping the administration of the drug[2,3]. We describe here some aspects of the pharmacokinetics and tissue levels of SHAM in mice.

SHAM (a gift from Prof. T. Urbanski, Technical University, Warsaw, Poland) was administered to mice in a dose of 200 mg/kg as a solution (in isotonic saline, pH 7.5) by gavage tube. 5 female, SAS/ICI mice were killed at each of the time intervals 30, 45, 60 and 75 min after administration and samples of blood, liver, kidney and brain taken. Tissue metabolism was halted with 10% trichloroacetic acid and the tissues weighed and homogenized by ultrasonication forming a homogenate at a concentration of 1:4 w/v. 0.3-ml aliquots of these homogenates were extracted and analyzed by the method of Barnicoat et al.[4]. This method of assaying SHAM and its major metabolite, salicylamide (SAM), uses high pressure liquid chromatography and UV-absorbance detection and is very sensitive, accurate and reproducible with a coefficient of variation of 4–6.6% over the concentration range 1–50 µg/ml and a minimum level of detection of 0.1 µg/ml from a 0.1-ml sample for both SHAM and SAM.

Tissue levels of SHAM following oral administration are shown in the figure. Observations from the organs of control animals treated with saline only showed that in the absence of SHAM there are no interfering peaks in the chromatograms. SAM was detected in all samples of tissues



a Concentration, time profiles for SHAM in kidney (●), brain (■), liver (▲) and blood (○) following oral administration of 200 mg/kg to mice (each point is the mean of 5 individual animals). *b* Concentration, time profile for SAM in kidney following oral administration of 200 mg/kg SHAM to mice (each points is the mean of 5 individual animals).

but the levels were only measurable in samples of kidney homogenate. These results show that after oral administration of a solution of SHAM, its absorption and distribution into the tissues is rapid. The concentrations of SHAM in the kidney, which is responsible for the excretion of unchanged SHAM as well as the glucuronide and sulphate conjugated metabolites of SHAM[5], are noticeably higher than in the other tissues. The levels of SHAM determined in the liver are relatively low (they are of the same order as those found in the brain). Although salicylamide was detected in all sampled tissues, the levels were only measurable in the kidney.

The plasma protein binding of SHAM was assessed by equilibrium dialysis using Visking membrane using human plasma from 1 individual spiked at concentrations over the range between 12 µg/ml and 10 mg/ml. Dialysis was made against pH 7.4 phosphate buffer (0.067 M) at 37 °C. Over this concentration range the overall binding of SHAM was 89.7 (SD 2.2)% for 31 determinations. There was no evidence of saturation of binding at the highest concentrations since the mean binding was 95.7% at 10 mg/ml, 98.6% at 1000 µg/ml and 87.7% at 12 µg/ml. This binding is probably tight and there was no displacement of SHAM at a concentration of 50 µg/ml from its binding by concentrations of salicylate ranging from 5-30 µg/ml. These observations would suggest that the relatively low tissue concentrations of SHAM may reflect its high degree of protein binding. Furthermore, they also indicate that renal elimination may be mediated mainly via tubular secretion rather than by glomerular filtration.

Half-maximal inhibition of oxygen uptake by trypanosomes in vitro requires 15 µM SHAM (approximately 2.3 µg/ml) and 100 µM (approximately 15.3 µg/ml) inhibits over 90% of the trypanosome respiration[2]. Figure a shows that this latter concentration is barely attained in any of the tissues studied, apart from the kidney, and that because of the rapid half-life of SHAM in the tissues, effective levels are not long maintained. These factors presumably explain why even a dose of 500 mg/kg SHAM given to rats infected with *Trypanosoma brucei* does not prolong their survival time[2].

Thus the short half-life (found to be 30 min in the mouse) and the high degree of protein binding may explain the relative lack of in vivo efficacy of SHAM[2,3,6] despite its promising in vitro profile. Although its elimination is probably predominantly renal, interference with this route of excretion is unlikely to prove beneficial in improving the therapeutic ratio.

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Spontaneous rhythmic contractions in isolated human coronary arteries[1]

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Summary. Isolated post mortem human coronary arteries developed rhythmic contractions in physiological saline solution without being exposed to vasoactive agents.

In recent years, coronary vasospasms have been attracting attention as possible trigger mechanisms in the onset of various forms of ischemic heart diseases such as Prinzmetal's variant form of angina pectoris[2,3] or myocardial infarction[4,5]. However, the etiology of such spasms is still unclear. We noticed that isolated post mortem human coronary arteries developed phasic, rhythmic contractions in a period of several min in a nutrient solution without being exposed to vasoactive agents.

Materials and methods. 4 coronary artery segments each were taken from 29 post mortem human hearts (19 males, 10 females, aged from 25 to 80, average 59 years), each segment measuring about 1.5 cm in length. The hearts were extirpated within 18 h after death. The segments were from the proximal portion of the right coronary and left anterior descending arteries and the mid portion and distal portion of the left anterior descending artery (116 coronary artery segments). Immediately after death, the cadavers were placed in a room kept at a constant temperature of 8-9 °C. The time that elapsed from the autopsy to the start of the experiments was no more than 5 h. During this period, the isolated arteries were preserved in a nutrient solution saturated with a mixture of 95% O_2 and 5% CO_2 at a temperature of 4 °C. The arteries were helically cut at an angle of approximately 45° to the longitudinal axis into strips according to the method of Furchgott and Bhadrakom[6]. The helical strips were fixed vertically between hooks in a 50-ml bath containing a nutrient solution. The upper end of the strip was connected to the lever of an isometric tension transducer (TB611T Nihonkoden Kogyo Co., Tokyo, Japan). The resting tension was adjusted to 1.5 g (proximal portion and mid portion) and 0.5 g (distal portion). The bathing solution was bubbled with a mixture of 95% O_2 and 5% CO_2 and was maintained at 37 ± 0.5 °C. The pH of the solution was 7.4. The composition of the bathing medium was as follows (in mmolar concentrations): Na^+, 162.1; K^+, 5.4; Ca^{++}, 2.5; Mg^{++}, 0.76; Cl^-, 157.0; $H_2PO_4^-$, 1.7; HCO_3^-, 14.9; dextrose, 5.6. The sam-

Incidence of rhythmic contractions and developed tension classified by portion of the coronary arteries

	Outer diameter (mm)*	Incidence N	%	Developed tension (g)**
RCA (proximal)	3.95 ± 0.74	14/24	58	1.00 ± 0.49
LAD (proximal)	4.15 ± 0.71	10/24	42	0.66 ± 0.19
LAD (mid)	3.55 ± 0.53	12/24	50	0.77 ± 0.24
LAD (distal)	1.81 ± 0.60	4/24	17	0.88 ± 0.39

N, number of preparations developed rhythmic contractions/number of preparations examined. RCA, right coronary artery. LAD, left anterior descending artery. * Mean ± SD, ** Mean ± SE.