The absorption of carrageenans

SIR,—An understanding of the manner of gastrointestinal absorption of carrageenans would be useful in two ways. First, it would add to knowledge of the absorption of sulphated polysaccharides, including heparin, which is present on both sides of the gastrointestinal wall, but appears not to be absorbed in active form or quantity. Second, it would help to explain the actions of carrageenan in reducing histamine-induced parietal cell hyperplasia (Anderson & Soman, 1965a), in reducing secretion (Anderson, Marcus & Watt, 1962), and in preventing histamine-induced gastric ulceration in the pylorus-ligated guinea-pig when administered intraduodenally (Anderson & Soman, 1963). The last-named action indicated for the first time that the gastric effects of carrageenans (principally, anti-ulcer and anti-secretory) could be accomplished either after absorption or by one of several possible humoral effects (Giertz, Hahn, Schmutzler & Seseke, 1964; Anderson & Soman, 1966).

Carrageenans. Degraded κ-carrageenan from *Eucheuma spinosum* (C 16), degraded λ-carrageenan from *Gigartina pistillata* (GP-λ-D2), and undegraded λ-carrageenans from *Chondrus crispus* and *G. Pistillata* (CY-λ and GP-λ respectively) were used. Weight-average molecular weights (light-scattering) for the degraded carrageenans are 20,000–30,000; for the undegraded carrageenans they are 800,000–1,000,000.

Administration of carrageenans. Single intravenous injections of saline solutions of C 16 were given to male guinea-pigs, average weight 400 g, of P strain (Anderson & Soman, 1966) food, but not water, was withheld for 18 hr and at the end of this period urine was manually expelled from the bladder (or removed immediately after killing) and examined for sulphated polysaccharide. Qualitative examination was by staining with toluidine blue on filter paper; quantitative examination was by MacIntosh's (1941) method, suitably modified.

Oral administration was by inclusion in the drinking water, the initial and residual volumes over 18 hr being recorded. Food was withheld during the period. Oral consumption was also measured during experiments designed to demonstrate the anti-duodenal ulcer activity of carrageenan. Duodenal ulceration was produced by histamine acid phosphate (10 mg/kg in a wax-oil base, intramuscularly) (Anderson & Soman, 1965b).

Intraduodenal administration in 2 ml of saline was effected during anaesthesia (pentobarbitone sodium, 30 mg/kg, intraperitoneally) in the pylorus-ligated guinea-pig in an experiment designed (Anderson & Soman, 1963), to demonstrate the anti-gastric ulcer effect of carrageenan.

The results of intravenous injection of C16 are in Table 1. Increase in dose of

TABLE 1. URINARY CONTENT OF DEGRADED CARRAGEENAN AFTER A SINGLE INTRAVENOUS INJECTION TO GUINEA-PIGS

Group	Number of animals	Dose of degraded carrageenan mg/kg i.v.	Degraded carrageenan in urine mg/ml
1	4	2·5	0 (but present qualitatively)
2	3	4	0 03
3	4	5	0·05
4	3	5	0·03
5	4	7·5	0·13
6	5	7·5	0·11
7	3	10	0·07
8	3	12·5	0·06
9	3	15	0·33

C16 corresponds roughly to increase in urinary content of the polysaccharide. Assuming that it is degraded carrageenan which appears in the urine, the results show that between 2·5 and 5 mg/kg C16 given intravenously to the guinea-pig results in the apppearance of urinary degraded carrageenan, indicating that there is no total renal barrier to its excretion. A method depending on metachromasia indicates that the polyanionic nature of the carrageenan is unchanged during its passage through the animal body.

C16 was also administered orally to guinea-pigs in drinking water (1%), over a period of 18 hr. When 298 mg of degraded carrageenan was consumed, the urine showed the qualitative presence of C16 only; using a 5% solution (1850 mg consumed) the urine contained about 0·3 mg/ml at the end of the experiment. Intraduodenal administration of 400 mg C16 in an anti-gastric ulcer experiment in the pylorus-ligated guinea-pig gave detectable, but not measurable, urinary metachromasia, indicating intestinal absorption. Gastric ulceration was reduced by 60%. GP-λ-D2 gave similar results. The experiment with GP-λ-D2 was conducted in association with an anti-duodenal ulcer test of this substance and the consumption of 675 mg of GP-λ-D2 was accompanied by 73% reduction in duodenal ulceration and demonstrable, but not measurable, urinary metachromasia. In an anti-gastric ulcer experiment the intraduodenal administration of 150 mg of GP-λ-D2 gave no demonstrable urinary metachromasia, but resulted in 47% reduction in gastric ulceration. Absorption of GP-λ-D2, if indeed it occurred, therefore appeared to be less than about 2·5 mg/kg (equivalent to about 1 mg total per animal) on the basis of the results in the Table. The minimum dose of GP-λ-D2 showing anti-gastric ulcer activity is 50 mg (Anderson & Soman, to be published); of this, it appears that about 1 mg or less can be absorbed. This suggests that the effect of carrageenan in anti-ulcer experiments is to stimulate some protective mechanism. When 400 mg of C16 was administered intraduodenally in addition to an intravenous injection (groups 4 and 6) the measurable urinary carrageenan did not increase, supporting the conclusion that intraduodenal carrageenan is absorbed in very small amounts.

With the degraded carrageenans it therefore appears that the anti-ulcer effect can be achieved with amounts of carrageenan less than that necessary to give detectable urinary excretion.

Turning to undegraded carrageenan (GP-λ), we have never detected it in urine, although the maximum total dose consumed was 158 mg. GP-λ does have an anti-ulcer effect (Anderson & Soman, to be published), but its high viscosity in solution precludes larger oral or intraduodenal dosage, which the experiments with degraded carrageenan show are necessary to obtain urinary excretion. Thus with the undegraded carrageenans also, doses giving anti-ulcer effect are lower than those which would be required to show urinary excretion. The absorption of undegraded carrageenan, although generally held to be unlikely, is still an open question on the basis of its anti-gastric ulcer effect after intraduodenal administration in the pylorus-ligated guinea-pig (Anderson & Soman, to be published). There are two further points about undegraded carrageenan: first, its molecular size (assuming absence of excessive polydispersity) may well prohibit gastrointestinal absorption; second, intravenous injections of higher doses of undegraded carrageenans in this type of experiment are prevented by extreme toxicity both in rabbits (Anderson & Duncan, 1965) and in guinea-pigs at 1 mg/kg, the animals dying, probably from pulmonary embolism, within half an hour.

That gastrointestinal absorption of degraded carrageenan is possible, helps to explain the more efficient protection (Anderson & Soman, 1965b) against experimental histamine duodenal ulceration the carrageenan gives when administered in drinking water before the histamine rather than after it.

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Effect of amphetamine and reserpine on the pressor response to tyramine in the rabbit and cat

SIR,—The potentiation of the pressor response to tyramine after amphetamine has been shown in the dog (Eble & Rudzik, 1965) and in the rat (Eble & Rudzik, 1966a). However, combinations of amphetamine and reserpine antagonise the pressor response to tyramine in the dog (Eble & Rudzik, 1966b). In these studies of the interactions of amphetamine and tyramine in other species it was found that in the rabbit, tyramine was a relatively weak pressor agent and that its effect was not potentiated by amphetamine, but was blocked by combinations of amphetamine and reserpine. The pressor responses of the cat to tyramine and the interaction with amphetamine and reserpine were similar to those found in the dog but not like those found in the rabbit.

Rabbits (3-5 kg) and cats (1·8–3·6 kg) of either sex were anaesthetised with sodium pentobarbitone (32 mg/kg, i.v.), with supplements of 3·2 mg/kg as required. Blood pressures were recorded from the carotid artery and drug injections made through a polyethylene tube passed 3 to 5 cm into a femoral vein.

(+)-Amphetamine (250 or 500 μg/kg, i.v.) failed to potentiate the pressor response to tyramine in rabbits (Table 1). In two additional experiments larger

TABLE 1. EFFECT OF AMPHETAMINE AND RESERPINE ON THE PRESSOR RESPONSE TO TYRAMINE IN THE RABBIT

Treatment	No. of Determinations	Mean b.p. response to tyramine (250–500 μg/kg, i.v.)	P value
Control	5	31 ± 5·9	
After amphetamine (250 μg/kg, i.v.)		36 ± 4·9	> 0·1
After amphetamine (250 μg/kg, i.v.) + reserpine (1 mg/kg, i.v.)		51 ± 4·5	< 0·02
Control	7	21 ± 3·0	
After amphetamine (500 μg/kg, i.v.)		28 ± 4·2	> 0·1
After amphetamine (500 μg/kg, i.v.) + reserpine (1 mg/kg, i.v.)		45 ± 4·6	< 0·01
Control	5	41 ± 5·4	
After reserpine (1 mg/kg, i.v.)		56 ± 10·1	> 0·05
After reserpine (1 mg/kg, i.v.) + amphetamine (250 μg/kg, i.v.)		28 ± 3·4	< 0·05
Control	5	30 ● 5·0	
After reserpine (2 mg/kg, i.v.)		62 ± 7·6	< 0·02
After reserpine (2 mg/kg, i.v.) + amphetamine (250 μg/kg, i.v.)		30 ± 5·0	< 0·001