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# Rectal Motility and Bioavailability

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**Abstract:** The contractile activity of the canine rectal wall exhibits a positive influence on the behaviour of fatty suppositories *in vivo* with respect to both spreading abilities and rate and extent of release of the readily water-soluble compound phenazone. This influence on bioavailability was marked when the drug was suspended in a large particle size (100–125 $\mu$m). When used in small particles ($< 35\mu$m), far less influence of contractile activity was found. Small particles were equivalent to coarse particles with respect to the bioavailability. The addition of colloidal silicium oxide has a marked influence on spreading and bioavailability. Enhanced rectal motility exhibits an influence on the absorption only when a coarse fraction of the drug is suspended. It was concluded that rectal motility might be a cause of variation in bioavailability of drugs from rectal suppositories. For this reason only well-trained animals should be used when bioavailability of drugs from suppositories is tested in an animal model.

The bioavailability of a drug administered rectally in fatty suspension suppositories might be influenced by various factors related to the suppository base, the drug incorporated and the rectal environment (1). Among these factors the properties of the physiological environment may play an important role. However, until now this role is of a yet unknown extent.

Especially the forces exerted on the suppository in the rectal cavity may have a marked impact on the spreading characteristics of the suppository mass. These forces result from 1) the pressure caused by the weight of the intra-abdominal organs, which may be further increased by respiratory activity and exercise, and 2) the contractions of the muscular layers in the rectal wall.

The influence of pressure caused by the weight of the abdominal organs has been recognized for valproic acid/cocoa butter suppositories (2): in sitting volunteers the maximum plasma concentration reached significantly higher levels than in supine persons. Furthermore, administration of freshly prepared and aged aminophylline suppositories to sitting volunteers caused no significant difference in bioavailability (3). Yet, the viscosity of the aged suppositories was increased considerably, probably due to the formation of high melting diamides (4).

These results lead to the hypothesis that the intraluminal rectal pressure, caused by the weight of the abdominal organs and exerted on the molten suppository mass might promote spreading of this mass, thus enhancing the bioavailability of the suspended drug.

Another factor that increases the intraluminal pressure in the rectal cavity is the contractile activity of the rectal wall. Until now, rectal pressure, caused by motility of the rectal wall, has not been studied extensively, in contrast to the numerous studies on the motility of other parts of the gastrointestinal tract (5). Most dynamic changes in the anorectum are believed to be due to the change in intra-abdominal pressure and the entry of material from the colon into the rectal cavity (6).

In order to elucidate the possible impact of rectal motility on the spreading and subsequently on the bioavailability of drugs from fatty rectal suppositories, we investigated the rectal motility pattern in dogs and the influence of this activity on the performance of suppositories.

The dog was chosen as an experimental animal, since other laboratory animals, like rats, are more difficult to train for experiments like the ones presented here. We found a considerable influence on rectal intraluminal pressure in rats, due to manipulation or even to the approach of the researcher. For this reason we concluded that the rat is not useful for experiments on rectal bioavailability.

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Dogs can be rightly used for studies on spreading and absorption, since they can be trained without great difficulty and since the canine rectum is not essentially different from the human rectum, except for the absence of the rectal valves.

## Materials and Methods

*Measurement of Rectal Motility*

Strain gauge force transducers (Micro-Measurements, type SA-03-090DH-350) were constructed for implantation (7) and were calibrated *ex vivo*. Three female beagle dogs, weighing approximately 11 kg, were trained to lie down for at least four hours, without concomitant sedative medication. They were implanted under aseptic conditions with six force transducers (8).

The transducers and lead wires were drawn through a stab wound between the scapulas, pulled through a subcutaneous tunnel and drawn into the abdominal cavity through a stab wound in the left abdominal wall. The transducers were sutured in transverse direction to the serosal side of the colon/rectum at regular intervals between the ileocolic junction and the anus. The most cranial transducer was fixed 4 cm caudally from the ileocolic junction, the most caudal one 4 cm from the anal verge. The contractility of the circular muscle was expressed as the amplitude of the contractions, i. e. the distance between the maximum of the motility curve and the baseline.

The dogs wore a canvas jacket in order to prevent scratching of the connectors, lead wires and wound. The connectors were packed in a pocket inside the jacket.

Motility patterns were recorded on an eight-channel carbon paper writer (U8000, Schwarzer München). Motor activity of rectum and colon was measured on a number of days, between 14 and 60 days after implantation. The transversally sutured transducers measured the contractile activity of the circular muscle.

*Stimulation of Rectal Motility*

Rectal motility was stimulated by neostigmine (20 $\mu g \cdot kg^{-1}$ s.c.), given to the fasting dogs. In an absorption experiment neostigmine was given 10 min before the insertion of the suppository.

*Suppositories*

The suppositories were prepared with Witepsol H15 (Dynamit Nobel, Witten, FRG) using the fusion method. The suppositories had a volume of 2 ml and contained 200 mg phenazone. Particle size fractions with two mean diameters were used, obtained by sieving (100–125 $\mu$m and <35 $\mu$m, respectively).

To study the influence of viscosity, 5 % m/m hydrophobic colloidal silicium oxide (Aerosil R972, Degussa Frankfurt, FRG) was added to the suppositories with phenazone in either particle size, resulting in four series of suppositories. The hydrophobic form of colloidal silicium oxide was chosen in order to prevent wetting and passage of the silica to the rectal mucus.

The content of the suppositories was found to be within acceptable limits (c.v. < 1 %, n = 3).

*Spreading of the Suppositories in vivo*

In a stainless steel mortar an aqueous colloidal suspension of approximately 30 MBq $^{99m}$Tc-Sn colloid (Amerscan, Amersham, UK) was heated under a gentle stream of nitrogen until dryness. A few ml acetone was added and mixed well with a pestle until the acetone had evaporated, thus preventing large agglomerates of Tc-colloid. A few ml molten suppository base was added and mixed well with the Tc-colloid. After addition of the phenazone, and if needed the silicium oxide, the remainder of the suppository base was added and mixed thoroughly. The mass was then poured in 2 ml suppository moulds. After preparation each suppository contained 3–5 MBq $^{99m}$Tc.

The $^{99m}$Tc-Sn colloid was not extractable from the molten suppository mass with water at 40 °C, and spread along with the suppository mixture, when the suppository was melted in a canine colon *ex vivo,* immersed in a waterbath at 36 °C. These results confirm that the spreading distance as measured outside the body indicates the real spreading.

The spreading of the suppositories in situ was measured in the dog, supine on a table, using a gamma-camera (General Electric Portacamera IIb) equipped with a parallel hole collimator. The range of spreading was calculated in comparison to a 15 cm standard. With an on-line computer the activity-distance curves were recorded in frames of 1 min.

*Bioavailability of Phenazone*

The dog was fasted for 40 h before each absorption experiment in order to achieve an almost complete emptying of the bowel; water was available ad libitum. Suppositories of each series were administered to each dog in a randomized sequence (n = 2–5). As a result of the training of the dog, no special precautions like sedation were needed. The dogs were supine on a table in horizontal position.

Before each experiment a teflon cannula (Abbocath-T 18-G, Abbott Ireland Ltd.) was brought into a fore-paw vein of the dog, and a three-way stopcock (Connecta, Viggo, Sweden) was connected. Blood samples were taken at regular intervals, and after each sampling the stopcock and cannula were flushed with sterile saline.

Since there is a constant ratio between the phenazone concentrations in plasma and whole blood (9), the latter was used for reasons of simplicity.

The blood samples were collected in heparinized tubes, and 0.5 ml was diluted with 1.0 ml water to obtain hemolysis. To each blood sample 2 $\mu$g phenacetine in ethyl alcohol was added as an internal standard. After addition of 100 $\mu$l 5M NaOH the mixture was extracted on a whirlmixer for 15 sec with 5 ml of a mixture containing equal amounts of dichloromethane and pentane. After separation from the aqueous phase the organic solvent was evaporated and the residue was dissolved in 100 $\mu$l ethyl alcohol. An aliquot was gas-chromatographed on a 2 % carbowax 20M column using N-P FI-detection (Hewlett-Packard 5710 A) (9).

Phenazone blood levels were calculated by comparing the ratio between the phenazone and phenacetine peak heights in the samples with the ratio from a standard, containing a known amount of phenazone and treated in a similar way to the blood samples.

The bioavailability was estimated by calculating the maximum concentration obtained (Cmax) and the absorbed fraction of administered phenazone. The fraction absorbed was estimated from the ratio of the total areas under curve (AUC) after rectal and intravenous administration, after extrapolation to infinity.

The elimination rate constants were determined by least-square regression analysis of the log-linear terminal parts of

the blood concentration-time curves. The AUC's were calculated by the trapezoidal rule from t = 0 the last sampling time. The AUC from the last sampling point to infinity was determined by dividing the concentrations at the last sampling time by the elimination rate constant.

Differences in pharmacokinetic and bioavailability parameters were tested using Student's t-test ($p < 0.05$).

## Results

### *Rectal Motility*

Immediately after a meal frequent contractions occurred on the rectal wall with a frequency of 5–6 $min^{-1}$, and a mean amplitude of 30–70 mN, depending on the dog. After some time (30–70 min) the amplitude of these contractions diminished, and the contractions faded away. During the rest of the day rectal contractions occurred simultaneously with the interdigestive colonic motor complexes (10), which occurred at intervals of 20–30 min and lasted 3–7 min. The appearance of complexes on the canine colonic wall will be discussed in more detail elsewhere (11).

In the fasting condition contractions of the rectal wall rarely occurred (Fig. 1).

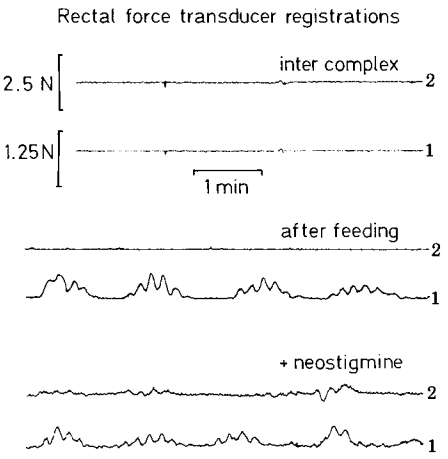


**Fig. 1** Rectal motility measured with force transducers, implanted 3 and 7 cm (1 and 2, resp.) from the anus.

Rectal contractions can be stimulated by an acetylcholine-esterase inhibitor like neostigmine: after administration of neostigmine (20 $\mu g \cdot kg^{-1}$) to the fasting dogs a motor activity of the rectal wall similar to the muscular activity after feeding occurred for a period longer than 60 min (Fig. 1). Thus in the spreading and absorption experiments neostigmine was administered to simulate the physiological rectal motor activity.

### *Spreading of the Suppository*

The spreading of the suppository mass was examined in quadruplicate in one fasted dog after administration of a fat/phenazone suppository without colloidal silicium oxide. Two conditions were selected: without drug and after administration of neostigmine. An example of a sequence of fluorescent screen photographs is shown in Figure 2. There was a marked influence of enhanced rectal motility, induced by neostigmine,

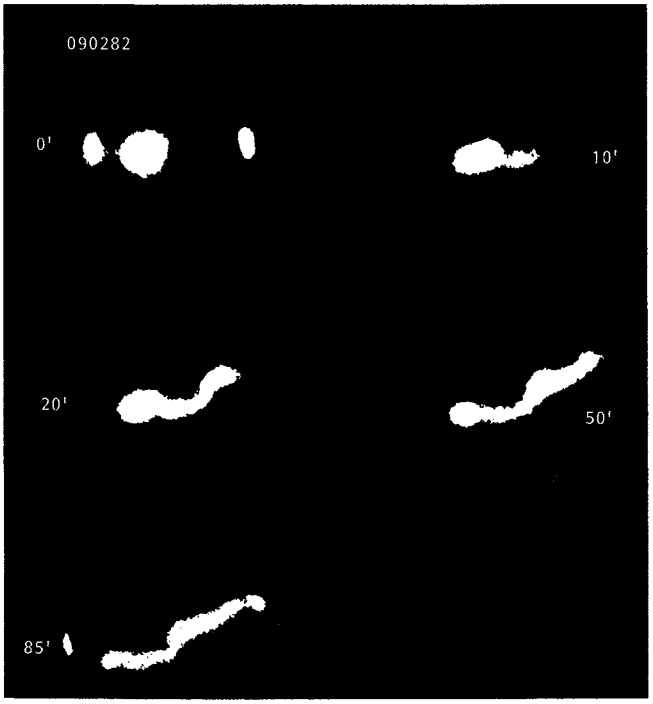


**Fig. 2** Polaroid photographs from the gamma-camera fluorescent screen at the indicated time after insertion of the suppository; on the first picture (0') the comparative 15 cm-standard is seen.

on the spreading distance of the molten fat (Fig. 3). As a measure of the spreading distance, the distance between the upper and lower limits of detected radioactivity was taken. Reproduction of the activity-distance frames from the on-line computer shows clearly the distribution of the incorporated activity in the rectal cavity (Fig. 4 a and b). Calculation of the activity in each frame after correction for Tc-decay, shows that the total activity did not decrease, indicating that the Tc was not absorbed.

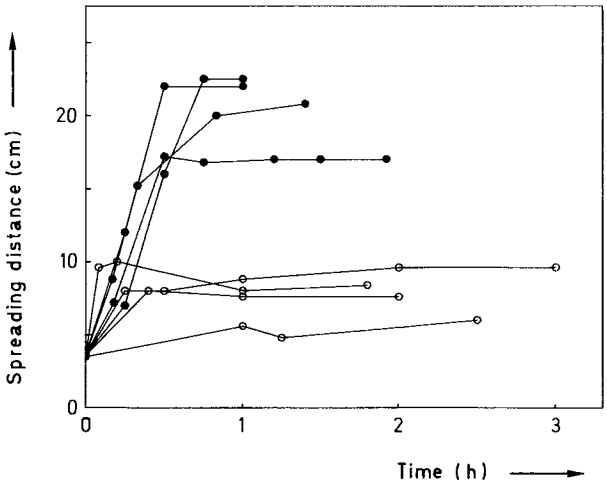


**Fig. 3** Spreading distances of the Witepsol/phenazone suppositories in situ in the canine rectum; the closed symbols indicate the spreading after neostigmine stimulation.

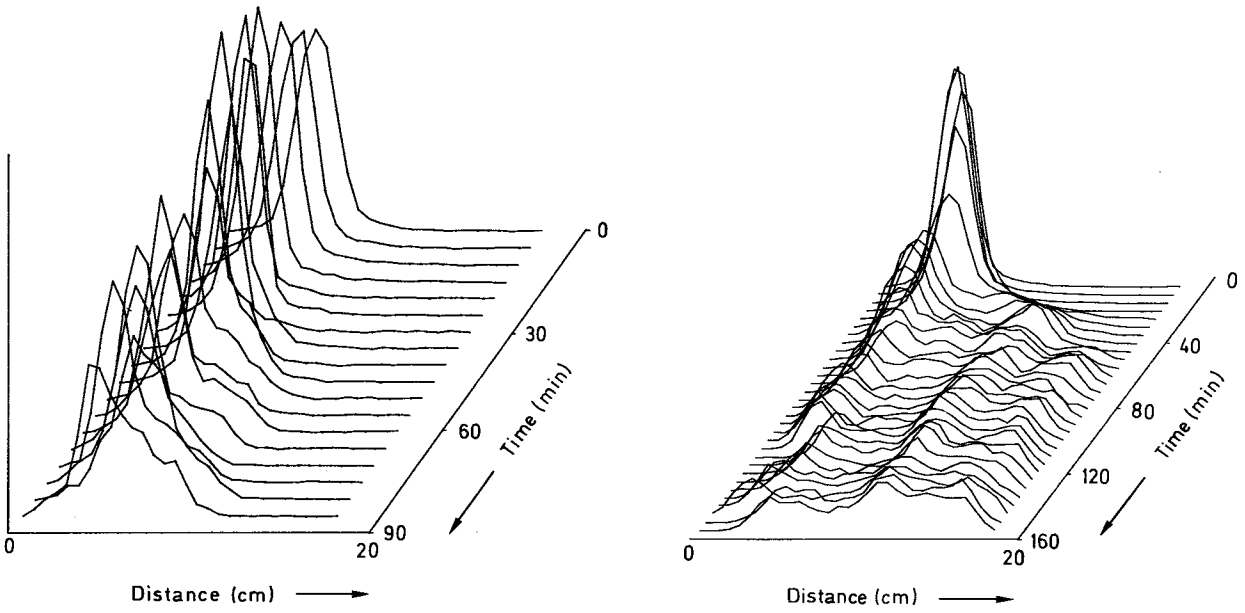


**Fig. 4a+b** Activity-distance curves in one minute frames from the on-line computer; in 4b after stimulation with neostigmine.

*Bioavailability*

The absorption of phenazone from the rectal suppositories was measured under two conditions in three fasted dogs: without and after administration of neostigmine. In all experiments the rectum was empty before administration of the suppositories. The insertion of the suppositories did not stimulate the muscular activity of the rectal wall. According to the blood concentration-time data after intravenous administration of 200 mg phenazone this compound exhibits two-compartment kinetics with a rather short distribution phase and an elimination half-life of approximately 1 h. Administration of neostigmine prior to the dose of phenazone did not change the distribution and elimination parameters significantly ($t\ ½ = 0.91\ h \pm 0.10$ without vs. $0.83\ h \pm 0.04$ after neostigmine, NS; Figure 5 for one dog, n = 3).

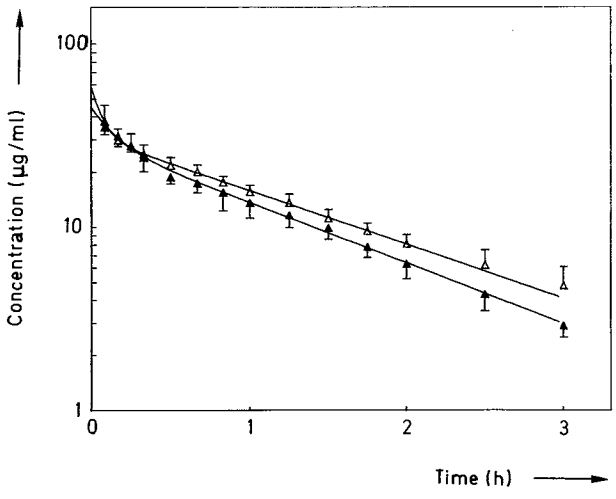


**Fig. 5** Mean blood concentration-time curves after intravenous administration of phenazone (n = 3); closed symbols after neostigmine stimulation.

The blood concentration-time curves after administration of the various suppositories are given in Fig. 6. In each figure the mean and standard deviations of several experiments are given; all data are the mean of 3–5 experiments, except for the silicium oxide suppositories in dog R, which were given only twice. The estimated maximum concentrations and the fractions absorbed are given in Table I, with standard deviations.

When phenazone was used in the large particle size, a significant increase in bioavailability is seen during enhanced rectal motility. The maximum concentration is significantly higher, and the extent of absorption is also higher, although still far below 100 % (Fig. 6 and Table I). It is rather surprising that the bioavailability of this readily water-soluble drug is never complete; to investigate the reason for this incomplete absorption, we washed the rectum of one dog with warm water at the end of six experiments. We recovered 37 ± 6 mg phenazone, indicating that the incomplete bioavailability is at least partly caused by incomplete release and/or absorption.

After administration under not-stimulated conditions of suppositories with phenazone suspended as smaller particles ($< 35\mu m$) the rate and extent of absorption was slightly improved compared to the suspended coarse fraction (Fig. 6). Stimulation of the rectal contractions did not significantly increase the rate or extent of absorption when the small particles were used. The absorption with this particle size was also far below 100 %.

Despite the hydrophobic character of the colloidal silicium oxide there was a marked increase in viscosity after addition of this adjuvant to phenazone/fat mixtures, which indicates that there is still an interparticular bonding that may be caused by hydrogen bonding (12). This increased viscosity significantly reduced spreading and release characteristics *in vitro* as well as *in vivo* (12, 13). Since the absorption was not completed before the last blood sample, the bioavailability could not be estimated using the AUC extrapolated to infinity. In the experiments with the silicium oxide suppositories, the fraction absorbed was calculated until the end of the particular experiment. In all experiments the maximum blood concentrations

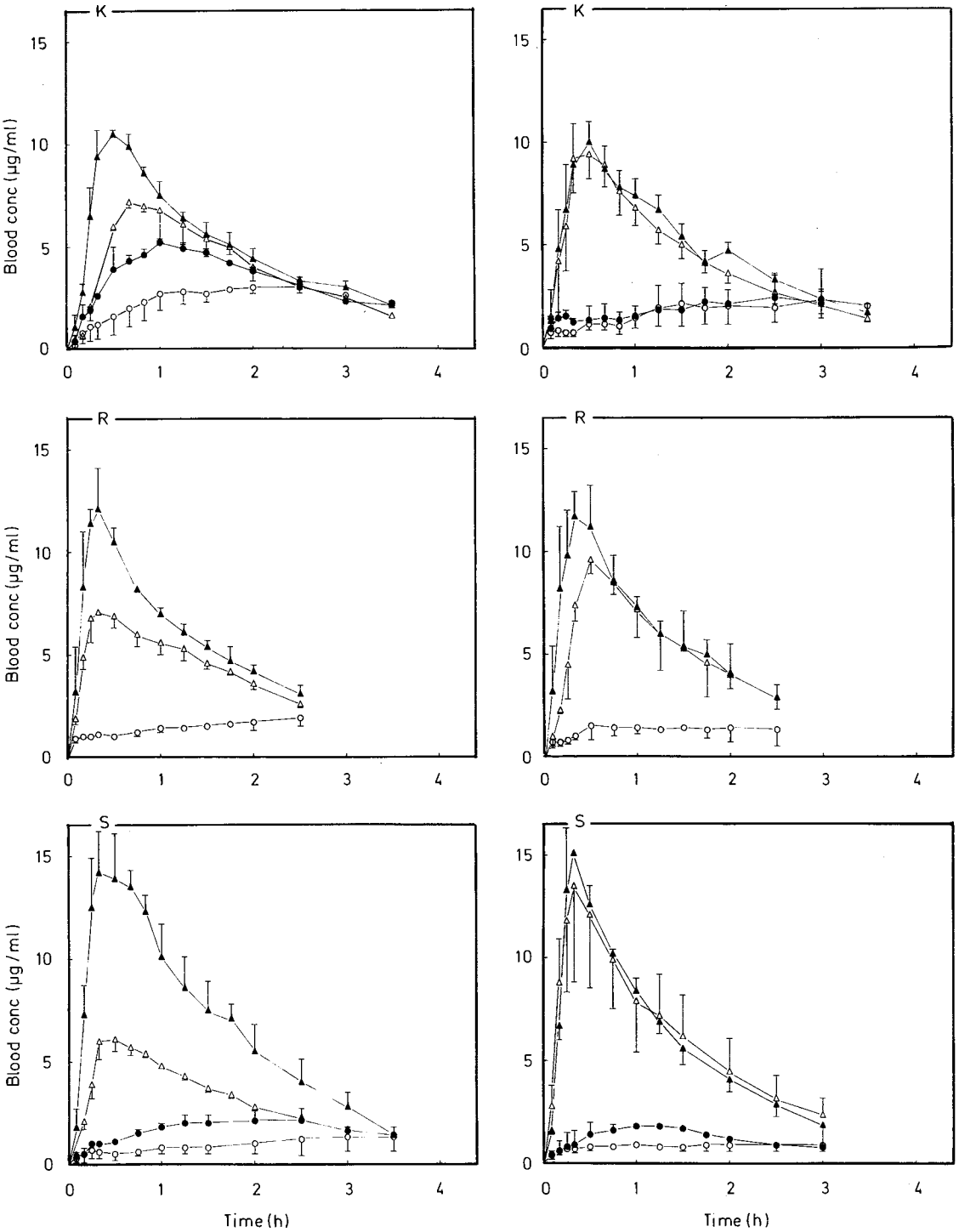


**Fig. 6** Mean blood concentration-time curves after administration of suppositories with phenazone to three dogs (± s.d.).
Δ: without, O: with 5 % Aerosil R972.
Left charts represent particle size 100–125 µm, right < 35µm.
Closed symbols: after s.c. administration of neostigmine.

were very low compared to the experiments with non-viscous suppositories (Table I).

The influence of enhanced rectal motility on bioavailability in the case of these more viscous suppositories was significant, when the large particle size was used. Nevertheless, the total amount absorbed and the maximum concentration achieved was still less than under not-stimulated conditions with non-viscous suppositories.

When phenazone was suspended as small particles in the suppositories with hydrophobic colloidal silicium oxide, no significant influence of the increased motility of the rectal wall was noticed. The higher viscosity of these suppositories that is caused by the particle size reduction of the phenazone, may have prevented the improved absorption rate in comparison to the coarse fraction, in contrast to the non-viscous suppositories.

**Table I.** Absorption parameters with standard deviations after administration of rectal suppositories to three dogs

| Particle size | Dog: / Stimulus | K F % | K Cmax $\mu g \cdot ml^{-1}$ | S F % | S Cmax $\mu g \cdot ml^{-1}$ | R F % | R Cmax $\mu g \cdot ml^{-1}$ |
|---|---|---|---|---|---|---|---|
| | – | 53±4 | 9.9±1.0 | 46±13 | 13.6±4.6 | 60±11 | 9.6±0.7 |
| < 35μm | – | Aerosil | | | | | |
| | + | 61±6 | 10.5±3.0 | 44±6 | 15.8±1.4 | 70±3 | 12.0±1.3 |
| | – | 53±4 | 8.0±1.1 | 31±2 | 7.9±2.0 | 53±4 | 7.5±3.0 |
| 100–125 μm | * – | Aerosil * | * | * | * | * | * |
| | + | 67±3 | 10.4±0.2 | 58±9 | 15.3±2.1 | 66±2 | 12.3±1.8 |
| | – | 19±8 | 2.9±1.0 | 6±1 | 1.1±0.2 | 11±2 | 1.9±0.5 |
| < 35 μm | + | Aerosil | | | | | |
| | + | 20±1 | 3.0±0.8 | 9±1 | 1.9±0.1 | n.d. | |
| | – | 27±3 | 3.0±0.3 | 6±3 | 1.4±0.7 | 10.2/12.5 | 1/1.2 |
| 100–125 μm | * + | Aerosil * | | | | | |
| | + | 36±1 | 5.2±0.2 | 11±2 | 2.1±0.5 | n.d. | |

*significantly different after stimulus; n.d. not done

## Discussion

In dogs, the rectal muscular wall exhibits a motor activity in recurring periods of the day. The contractions in the muscular layer of the rectum have a pronounced influence on the range of spreading of a molten fatty suppository in the rectal cavity. The impact of enhanced rectal motility on the bioavailability of a water soluble compound like phenazone is also quite pronounced, but only when the drug in the suppository is suspended in relatively large particles (100–125 μm).

When hydrophobic silicium oxide was added to the suppositories with the large particles in order to enhance the viscosity, a decrease in rate and extent of absorption was observed. Stimulation of rectal contractions also improved the rate and extent of absorption, which were however, far inferior to the plain fat/drug suppositories. These results lead to the rejection of the use of high concentrations of silicium oxide in suppository preparation without further examination.

In suppository preparation it is usually recommended to use a particle size < 150 μm for of readily water-soluble drugs to avoid rapid sedimentation during the manufacturing process and to improve the content uniformity (1). A very small particle size, however, influences the viscosity negatively, especially in high-dose suppositories.

In the suppositories with the drug suspended in small particles only a slight, not-significant influence was found of enhanced rectal contractile forces on the bioavailability, in contrast to suppositories with the coarse fraction. Rather surprisingly the rate of absorption after stimulation is equivalent in either particle size. This equivalence suggests that the release *in vivo* is no longer the rate limiting step in the overall process, but the diffusion through the rectal mucus and/or wall might become rate limiting. This suggestion is supported by the rate of absorption of phenazone from an aqueous solution: the maximum levels are reached in a time comparable to the results presented here.

From the experiments discussed here, it can be concluded that rectal contractile activity may influence the bioavailability of a drug from a suppository. The influence of rectal motility on bioavailability might produce misleading results when suppositories are tested in animals not accustomed to experimental manipulation. To exclude the influence of rectal contractions, bioavailability testing of fatty suppositories should be performed in well trained laboratory animals under strictly controlled conditions. For this reason, the use of rodents in suppository testing should be rejected unless they can be trained for this purpose.

It was shown that in dogs the bioavailability of this particular water-soluble drug from fatty suspension suppositories was far from complete. A certain amount of phenazone could be recovered from the rectal cavity after several hours. A bioavailability of 100 % was found after administration to upright human volunteers (13). In this trial the experiments were undertaken in an animal model in order to examine both rectal motility and bioavailability, since the investigation of these two parameters simultaneously is not possible in humans with current techniques. It should be emphasized, however, that the comparison of the results presented here and the mentioned results in literature might indicate that the dog in supine position is not a good substitute for man in bioavailability trials for suppositories, and bioavailability studies of drugs administered rectally should be performed in man.

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