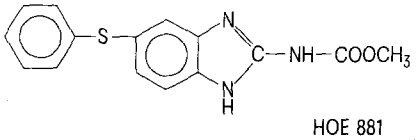
Fenbendazole: A New, Highly Effective Anthelmintic

During the past few years, several reports have been published on investigations with various anthelmintics which have a good efficacy against gastrointestinal nematodes in domestic animals. In our investigations with benzimidazole carbamates, we found substances which exhibited extremely high tolerance and very good efficacy against various nematodes of laboratory and domestic animals.

Some time ago we noted that the addition of a phenoxy group to a chemotherapeutically active molecule can considerably increase its efficacy[1]. The excellent action against liver flukes of rafoxanide, also a molecule with an added phenoxy group[2], was a further indication to us of the usefulness of our working hypothesis. The molecule from which it is derived belongs to a series of compounds, the salicylanilides, which are of interest because of their anthelmintic action. Mebendazole (methyl 5 benzoylbenzimidazole-2-carbamate), the anthelmintic newly developed at JANSSEN[3] as an enlarged molecule of the already known, highly effective nematodal remedies 2-benzimidazolyl-carbamic-acid esters[4], appeared to fit this same pattern. That gave us cause to extend our working hypothesis to this class of compounds. We found highly effective preparations, and were even surprised at the high degree of anthelmintic activity exhibited especially by methyl 5-(phenoxy)-2-benzimidazolecarbamate, and even more by methyl 5-(phenylthio)-2-benzimidazole-carbamate[5]. The latter compound



HOE 881
Fenbendazole

($C_{15}H_{13}N_3O_2S$, mol. wt. 299.2) is an almost colourless powder, melting at 233° with decomposition; it is freely soluble only in dimethylsulfoxide. Its UV-spectrum measured in methanol shows the maximum adsorption at 295 nm.

The first investigations on the efficacy of fenbendazole were carried out with *Nippostrongylus muris* and *Heterakis spumosa* in rats, and also with *Nematospiroides dubius* and *Aspiculuris tetraptera* in mice. At the end of the prepatent period of the different nematode species, the compound was given once to 3 times orally or subcutaneously on consecutive days to the experimentally infected laboratory animals. The curative dose for the individual species is

Nippostrongylus muris	3 × 75 mg/kg p.o.; 3 × 500 mg/kg s.c.
Heterakis spumosa	1 × 25 mg/kg p.o.; 1 × 50 mg/kg s.c.
Aspiculuris tetraptera	3 × 10 mg/kg p.o.; 3 × 12,5 mg/kg s.c.

Up to a dose of 3 × 1000 mg/kg given orally or subcutaneously, fenbendazole is not adequately effective against *Nematospiroides dubius*. The development of larval stages in the host animal is interrupted if during the prepatent period the following dosages are applied against the individual nematode species:

Nippostrongylus muris (3 days p.i.)	< 1 × 250 mg/kg p.o.
Heterakis spumosa (21 days p.i.)	< 1 × 250 mg/kg p.o.

The anthelmintic action of fenbendazol in domestic animals was investigated by the 'controlled test' and the results were confirmed in field trials. Consequently a single treatment with 5 mg/kg, given orally, to experimentally or naturally infected sheep causes a reduction in the worm burden of about 99–100% in respect of Haemonchus, Ostertagia in the abomasum, of Cooperia, Trichostrongylus, Nematodirus, Bunostomum, Oesophagostomum, and Chabertia in the small and large intestine, respectively. A dose of 5–10 mg/kg by mouth in experimentally or naturally infected cattle is 95–100% effective against Haemonchus, Ostertagia in the abomasum, against Trichostrongylus, Cooperia in the small intestine, and against *Dictyocaulus viviparus* in the lung. An oral dose of 5 mg/kg body weight in experimentally or naturally infected pigs results in a 100% elimination of *Hyostrongylus rubidus* (stomach) and also of the intestinal nematodes *Ascaris suis* and Oesophagostomum spec. A dose of 5 mg/kg by mouth is also highly effective in horses against small strongylids (Trichonema spec.), large strongylids (Strongylus spec.), as well as against ascarides. According to the results available so far, 20 mg/kg is required in dogs to bring about a 90–100% reduction in the worm burden in respect of ancylostomes, ascarides and trichurids. In so far as it has been possible to conduct such investigations, the burden of immature stages of the afore-mentioned worm species could also be largely reduced with the doses stated.

Fenbendazole in the acute experiment is extremely well tolerated. It was not possible in smaller laboratory animals to cause fatalities due to poisoning with the maximum quantities that could be administered, either orally or parenterally, so that the mean lethal dose (LD_{50}) could not be established. The LD_{50} is certainly higher than

10,000 mg/kg in rats orally
1,250 mg/kg in rats intraperitoneally
2,000 mg/kg in rats subcutaneously
10,000 mg/kg in mice orally.

Domestic animals: a single oral dose of 500 mg/kg caused no clinical changes in the dog. A dose of 1 × 5,000 mg/kg was tolerated by sheep also without clinical symptoms.

Studies of the oral subacute toxicity were carried out in rats by applying doses up to a maximum of 2,500 mg/kg body weight, in each case on 30 consecutive days. Even this dose was tolerated without any toxic reaction. Application of 30 × 25, 30 × 80 or 30 × 250 mg/kg to dogs caused no clinically detectable impairment of health; nor did 30 × 5, 15 and 45 mg/kg, respectively, administered to sheep, bring about either clinico-chemical or biochemical deviations from the physiological norm. The pathologic-histological findings were not prohibitive.

Tests for any teratogenic effect in rats after oral application from the 7th–16th day of pregnancy in daily doses of up to 2,500 mg/kg did not reveal any damage to the intrauterine development of the fetuses. They developed normally.

Preliminary investigations in the rat, dog, rabbit and sheep showed that elimination is virtually complete

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within 3–7 days. The maximum concentrations detected 14 days after treatment were 0.3 µg/g in the liver (sheep). In the other organs and tissues, particularly in the musculature, the concentrations were considerably lower.

Publications giving further details are in preparation.

Zusammenfassung. Fenbendazol wirkt bei einer Dosis von 5 bis 10 mg/kg p.o. auf alle bedeutenden Magen-, Darm-Nematoden inkl. einiger Organnematoden von Schwein, Schaf, Rind und Pferd, wobei nicht nur die Adulten, sondern auch die chemotherapeutisch schwer zu beeinflussenden Entwicklungsstadien (*O. ostertagi*) praktisch vollständig eliminiert werden. Bei extrem guter Verträglichkeit (bis zu 1000fach therapeutischer Dosis) und fehlender teratogener Wirkung konnte hier ein vielversprechendes Anthelminthikum entwickelt werden.

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Effects of High Gravity on Amoebae (*Pelomyxa carolinensis*). I. Division Rates

Amoebae have the distinction of being a life form which has undergone centrifugal experimentation more often than most other cell types or organisms[1–4]. In general, the experimental gravitational stresses experienced by these organisms has ranged from $27.5 \times g^5$ to $6{,}000 \times g^6$. The immediate effect of these forces is the density-dependent stratification of cytoplasmic inclusions of the amoebae in well-documented zonal patterns[7–9]. Following centrifugation, the amoebae appear to recover without any apparent ill effects and cytoplasmic streaming begins almost immediately; shortly thereafter, organelle redistribution begins[5,8,9]. According to HOLTER[10], microscopic appearance, motility, and oxygen consumption appear normal 30 to 60 min after centrifugation. However, no long range effect of gravitational stress on this organism has been reported. Therefore, we report here an extended study of the division rates of amoebae subjected to gravitation stresses below $27.5 \times g$ for various exposure periods.

Stock cultures of the amoebæ, *Pelomyxa carolinensis*, were maintained in PACE and MCCASHLAND[11] medium under subdued light at 22 °C and fed *Paramecia sp* on alternate days. A specially designed centrifuge supplied 20, 10, 5, 3.5, and $2.0 \times g$ for 1, 6 and 18 h. A 10% solution of gum arabic was used as the suspensory medium with amoebae medium as the solvent. Randomly selected organisms and 10 ml of medium were placed in a 50 ml centrifuge tube previously half-filled with suspensory medium. After transfer, the tubes were gently rotated in the palms of the hand causing the amoebae to become monopodal and establishing a suspensory gradient. The tubes were plugged with gauze and subjected to stress. Control organisms were exposed to the suspensory medium

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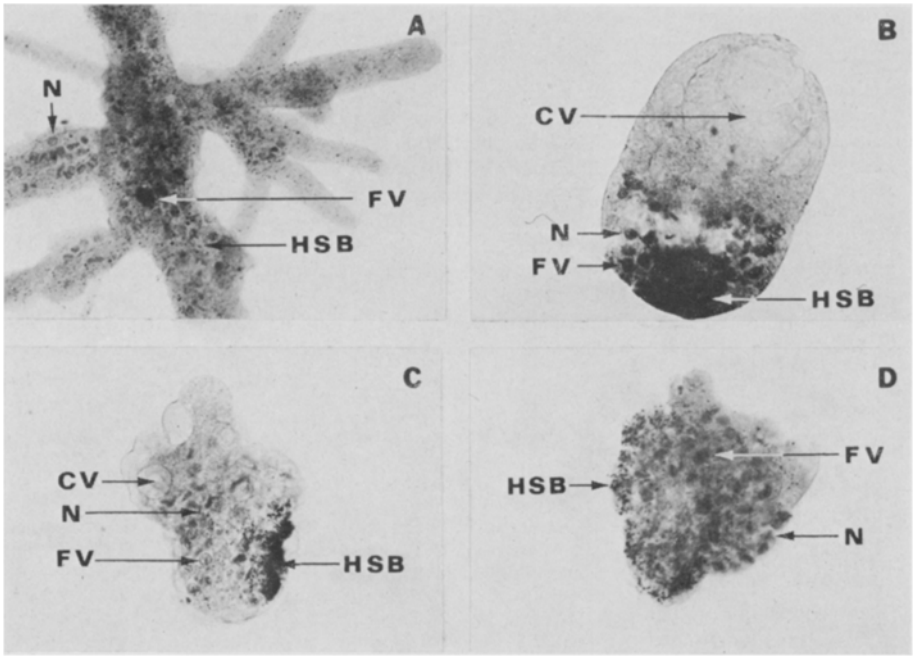

Fig. 1. Representative photographs of amoebae subjected to gravitational stress. Specimens are Lavdowsky fixed and Ehrlich's hematoxylin stained. Whole mounted × 600. A), B), C) and D) represent controls and amoebae centrifuged at 20, 3.5, $2.0 \times g$ for 24 h respectively. Symbols: HSB, heavy spherical bodies; N, nuclei; FV, food vacuoles; CV, contractile vacuoles.