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Plasma pharmacokinetics, faecal excretion and efficacy of pyrantel pamoate paste and granule formulations following *per os* administration in donkeys naturally infected with intestinal strongylidae

Cengiz Gokbulut[a,*], Dilek Aksit[b], Giorgio Smaldone[c], Ugo Mariani[d], Vincenzo Veneziano[c]

[a] Department of Medical Pharmacology, Faculty of Medicine, Balikesir University, Balikesir, Turkey
[b] Department of Pharmacology and Toxicology, Faculty of Veterinary Medicine, Balikesir University, Balikesir, Turkey
[c] Department of Veterinary Medicine and Animal Production, University of Naples Federico II, Naples, Italy
[d] Istituto Zooprofilattico Sperimentale del Mezzogiorno, Benevento Unit, Portici, Italy

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ABSTRACT

The plasma disposition, faecal excretion and efficacy of two formulations of pyrantel pamoate in donkeys were examined in a controlled trial. Three groups of seven donkeys received either no medication (control) or pyrantel paste or granule formulations at horse dosage of 20 mg/kg B.W. (equals 6.94 mg/kg PYR base) of body weight. Heparinized blood and faecal samples were collected at various times between 1 and 144 h after treatment. The samples were analysed by high-performance liquid chromatography. The last detectable plasma concentration (t_{max}) of paste formulation was significantly earlier (36.00 h) compared with granule formulation (46.29 h). Although, there was no significant difference on terminal half lives ($t_{1/2}$: 12.39 h *vs.* 14.86 h), t_{max} (14.86 h *vs.* 14.00) and MRT (24.80 h *vs.* 25.44 h) values; the C_{max} (0.09 μg/ml) AUC (2.65 μg h/ml) values of paste formulation were significantly lower and smaller compared with those of granule formulation (0.21 μg/ml and 5.60 μg h/ml), respectively. The highest dry faecal concentrations were 710.46 μg/g and 537.21 μg/g and were determined at 48 h for both paste and granule formulation of PYR in donkeys, respectively. Pre-treatment EPG of 1104, 1061 and 1139 were observed for the control, PYR paste and PYR granule groups, respectively. Pre-treatment EPG were not significantly different ($P > 0.1$) between groups. Post-treatment EPG for both PYR treatment groups were significantly different ($P < 0.001$) from the control group until day 35. Following treatments the PYR formulations were efficient (>95% efficacy) until day 28. In all studied donkeys, coprocultures performed at day−3 revealed the presence of Cyathostomes, *S. vulgaris*. Faecal cultures performed on different days from C-group confirmed the presence of the same genera. Coprocultures from treated animals revealed the presence of few larvae of Cyathostomes.

1. Introduction

Pyrantel (PYR) is an imidazothiazole derivative, which belongs to the tetrahydropyrimidine group of anthelmintics. PYR is available as tartrate and pamoate

* Corresponding author. Tel.: +90 266 612 14 61; fax: +90 266 612 14 48.
E-mail addresses: cengizgokbulut@yahoo.com, cgokbulut@gmail.com (C. Gokbulut).

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(syn. embonate) salts. Different salts of PYR have different pharmacokinetic properties and consequently different toxicities to the host. The pamoate salt is almost insoluble in water, poorly absorbed from the gastrointestinal tract, and most passes unchanged in the faeces (Arundel, 1983). Reduced systemic absorption of the pamoate form potentially increases availability in the lumen of the intestine (Bjorn et al., 1996). The tartrate salt of PYR is soluble in water and absorbed rapidly and extensively from the intestine of monogastric animals (Faulkner et al., 1972).

The daily administration of PYR tartrate to horses at a continuous low level was highly effective against common equine parasites, including adult large strongyles (*Strongylus vulgaris*, *Strongylus edentatus*, and *Triodontophorus* spp.), adult small strongyles (*Cyathostomum* spp., *Cylicocyclus* spp., and *Cylicostephanus* spp.) and adult and fourth-stage *Parascaris equorum* (Valdez et al., 1995). It was reported that the recommended dose level of PYR (13.2 mg/kg, bodyweight) pamoate paste formulation was effective (95–98%) against infections of *Anoplocephala perfoliata* (FDA, 2005). Moreover, a recent investigation by Reinemeyer et al., 2010 indicated that PYR pamoate paste formulation was highly effective against adult (91.2%) and fourth-stage *Oxyrus equi* larvae (>99%).

Only limited data are available on the pharmacokinetics and efficacy of anthelmintic drugs used in donkeys because donkeys are often a neglected species for studies in domestic animals. Most of drugs in different classes used in horses and ruminants are commonly extrapolated for use in donkeys without optimization of dosing regimens and determination of pharmacokinetic properties (Veneziano et al., 2011). Because of the lack of drugs approved for use in donkeys, anthelmintics licensed for use in horses or ruminants are used at the same dosages for treatment of parasitic infections in donkeys. It has been reported that donkeys have a greater capacity to metabolize certain drugs, compared with the capacity for horses; thus, higher dosages or shorter intervals could be required to maintain effective drug concentrations in donkeys (Welfare et al., 1996; Matthews et al., 1997; Coakley et al., 1999; Peck et al., 2002; Lizarraga et al., 2004; Grosenbaugh et al., 2011). Hence, in the present study, the pharmacokinetic disposition, faecal excretion and anthelmintic efficacy of two different formulations (paste and granule) of PYR pamoate are reported in donkeys naturally infected with intestinal strongylidae after oral administration.

2. Materials and methods

2.1. Study Animals

Twenty-one female crossbreed donkeys (*Equus asinus*) weighing 200–280 kg were used in this study. The bodyweight (BW) of each animal was estimated 1 day prior to treatment (day−1) using the nomogram proposed by The Donkey Sanctuary (2003). The animals had a mean age of 9.8 (±1.5) years and they had a history of grazing pasture contaminated with equine nematode parasites and have not been treated with any anthelmintics during the previous 9 months. Faecal examinations (individual Faecal Egg Counts and pooled coproculture) performed before the beginning of the study (day−3) showed individual counts >150 eggs per gram (EPG) and a high prevalence of intestinal nematodes (Cyathostomes, *S. vulgaris*) in all studied donkeys. The study animals were tagged for identification and housed communally in an indoor pen until the day 0 of the trial. The animals were kept indoors and fed hay-based diet. Water was provided *ad libitum* throughout the course of the study. This investigation was approved by the Animal Ethic Committee of University of Naples Federico II.

2.2. Experimental groups

On day−3, 21 of the experimental donkeys had an average of 1101 ± 583 EPG. The animals were ranked from lowest to highest EPG counts. Based on increasing EPG counts, replicates of 3 animals were formed. Within each replicate, animals were randomly assigned to treatment. The 21 selected donkeys were assigned consecutively to the following treatment groups of 7 animals each: PYR paste treated group (PYR-P group), PYR granule treated group (PYR-G-group), and untreated control group (C-group).

2.3. Drug administration

Commercially available equine formulations of PYR paste (Strike PYR paste pamoate 38%, Acme, Italy) and PYR pamoate granulate (Strike, PYR pamoate 20%, Acme, Italy) licensed for horses were administered orally to donkeys at a recommended dosage rate of 20 mg/kg B.W. which equals 6.94 mg/kg PYR base.

2.4. Sampling procedure

Heparinized blood samples were collected by jugular venipuncture prior to drug administration and 1, 2, 4, 8, 12, 16, 24, 30, 36, 48, 56, 72, 96, 120 and 144 h thereafter. Faecal samples (>10 g) were collected per rectum throughout the blood-sampling period, before drug administration and then at 4, 8, 12, 16, 24, 30, 36, 48, 56, 72, 96 and 120 h in order to determine faecal excretion of PYR under study. Blood samples were centrifuged at $1825 \times g$ for 30 min and plasma was transferred to plastic tubes. All the plasma and faecal samples were stored at −20 °C until estimation of drug concentration.

2.5. Analytical procedure

The parent compound of PYR was analysed by high performance liquid chromatography (HPLC). The liquid–liquid phase extraction procedure used for PYR was adapted from that described by McKellar et al. (1993a). Briefly, 1 ml drug-free plasma samples were fortified with PYR standard to reach the following final concentrations: 0.01, 0.05, 0.1, 0.5, 1 and 5 μg/ml. Morantel citrate was used as an internal standard. Sodium hydroxide (NaOH) (0.5 ml, 0.4 M) was added to tubes. After vortex for 15 s, 6 ml chloroform was added. The tubes were shaken for 2 min. After centrifugation at $2000 \times g$ for 15 min, 4 ml of the organic phase was transferred to the glass tube and evaporated to dryness at 43 (C in a sample concentrator—Maxi-dry plus, Heto Lab. Equipment, Denmark). The dry residue was dissolved in

300 μl of the mobile phase and 100 μl of this solution was injected into the chromatography system.

Faecal material was mixed with a spatula to obtain a homogeneous sample. Drug-free faecal samples (0.5 g) were fortified with PYR standard to reach the following final concentrations: 1, 5, 50, 100, and 300 μg/g. Morantel citrate was used as an internal standard. Acetonitrile (2 ml) was added to tubes containing 0.5 g fortified blank samples and experimental samples. After vortex for 15 s, 6 ml chloroform was added. The tubes were shaken for 2 min. After centrifugation at 2000 × *g* for 15 min, 4 ml of the organic phase was transferred to a glass tube and evaporated to dryness at 43 (C in the sample concentrator. The dry residue was dissolved in 300 μl or 5 ml of the mobile phase and 50 μl of this solution was injected into the chromatography system. Because of the photosensitivity of PYR all preparative processes were conducted in covered containers. To determine the dry weight of wet faecal samples, 1.0 g of wet faeces from each sample was weighed exactly into an evaporating bowl and heated in an oven at 70 °C for 10 h. The weight of each sample was determined and the percentage of each dry sample was calculated.

In the HPLC system, a mobile phase of acetonitrile:water (30:70) with 0.6% (v/v) triflouro acetic acid (TFA) pumped (1100 Series QuatPump, Agilent, Waldron, Germany) at flow rate 1 ml/min was used for plasma samples and acetonitrile:water (15:85) with 0.6% (v/v) TFA pumped at flow rate 1.4 ml/min was used for faecal samples. A nucleosil C_{18} analytical column (Luna, 4 μm, 150 mm × 4.6 mm, Phenomenex, Macclesfield, Cheshire, UK) with nucleosil C_{18} guard column (Phenomenex, Macclesfield, Cheshire, UK) was used with ultraviolet detection (1100 Series PDA detector, Agilent, Waldron, Germany) at a wavelength of 322 nm.

The analytical methods used for PYR in plasma and faecal samples were validated prior to the start of the studies. The analyte was identified with the retention times of pure reference standard. Recoveries of the molecule under study were measured by comparison of the peak areas of spiked plasma and faecal samples with the areas resulting from direct injections of standards prepared in acetonitrile. The inter- and intra-assay precision of the extraction and chromatography procedures were evaluated by processing replicate aliquots of drug-free donkey plasma and faecal samples containing known amounts of the drugs on different days.

Calibration graph for PYR was prepared (linear range 0.01–5 μg/ml in plasma and 1–300 μg/ml faecal analysis). The slope of the lines between peak areas and drug concentration was determined by least squares linear regression and correlation coefficient (*r*) and coefficient of variations (CV) calculated. Linearity was established to determine the PYR concentration/detector response relationship. The detection limit of PYR was established with HPLC analysis of blank plasma fortified with the standard, measuring the baseline noise at the retention time of the peak. The mean baseline noise at the peak retention time, plus three standard deviations was defined as the detection limit (LOD). The mean baseline noise plus six standard deviations was defined as the limit of quantification (LOQ).

2.6. Coprological examinations and anthelmintic efficacy

According to general recommendations proposed by Nielsen et al. (2010) faecal samples were taken from the rectum from each study animal, were stored in a refrigerator (4 °C) and individual faecal egg counts (FEC) were performed, within 12 h before the start of the trial (day−3), at days 0, 7, 14, 21, 28, 35, 42 and 56 after treatment. Individual faecal egg counts were determined using a modified McMaster technique with a detection limit of 10 EPG, using a Sheather's saturated sugar solution with a specific gravity of 1.260 (MAFF, 1986).

On each sampling day, individual faecal samples were incubated at 27 °C for 7–10 days for larval identification. Only before the start of the trial (day–3) were performed pooled coprocultures. Third stage larvae were identified using the morphological keys proposed by MAFF (1986). When a coproculture had 100 or fewer third stage larvae, all were identified; when a coproculture had more than 100 larvae, only 100 were identified.

To determine the efficacy of PYR against intestinal strongyles at each faecal sampling time, the arithmetic mean of EPG was calculated. Following the American Association of Equine Practitioners (AAEP) Parasite Control Guidelines (Nielsen et al., 2013a), for each animals percent efficacy (%) was calculated in terms of faecal Egg Count Reduction (FECR) at the different days according to the formula:

$$\mathrm{FECR}(\%) = \frac{\text{Mean EPG (before treatment)} - \text{Mean EPG (after treatment)}}{\text{Mean EPG (before treatment)}} \times 100$$

2.7. Pharmacokinetics and statistical analysis of data

The plasma concentration *vs.* time curves obtained after each treatment in individual animals, were fitted with the WinNonlin software program (Version 5.2, Pharsight Corporation, Mountain View, CA, USA). The pharmacokinetics parameters for each animal were analysed using non-compartmental model analysis. The maximum plasma concentration (C_{max}) and time to reach maximum concentration (t_{max}) were obtained from the plotted concentration-time curve of each drug in each animal. The trapezoidal rule was used to calculate the area under the plasma concentration time curve (AUC):

$$\mathrm{AUC}_{0\to\infty} = \sum_{i=1}^{n} \frac{C_i + C_{i-1}}{2} \times (t_i - t_{i-1}) + \frac{C_n}{\lambda_z}$$

Where C represents the plasma concentration, i-1 and i are adjacent data point times. The area under the first

movement curve (AUMC) was calculated using the equation:

$$AUMC_{0\to\infty} = \sum_{i=1}^{n} \frac{C_i t_i + C_{i-1} t_{i-1}}{2} \times (t_i - t_{i-1}) + \left(\frac{C_n}{\lambda_z^2} + \frac{t_n C_n}{\lambda_z} \right)$$

Thus, the mean residence time (MRT) was calculated as:

$$MRT_{0\to\infty} = AUMC_{0\to\infty} / AUC_{0\to\infty}$$

Terminal half-life ($t_{1/2\lambda z}$) was calculated as: $t_{1/2\lambda z} = -\ln(2)/\lambda_z$

Where λz represent the first order rate constant associated with the terminal (log linear) portion of the curve.

The pharmacokinetic parameters are reported as mean ± SD. Mean pharmacokinetic parameters were statistically compared by one-way analysis of variance (ANOVA). All statistical analyses were performed by using MINITAB for Windows (release 12.1, Minitab Inc., State College, PA, USA). Mean values were considered significantly different at $P < 0.05$.

For comparison of the anthelmintic efficacy of both formulations, statistical analysis of data was performed on arithmetic mean (AM) EPG counts using the parametric *t*-test to compare differences between treatment groups for significance at the $P < 0.01$ level.

3. Results

The analytical procedures and HPLC analysis of PYR were validated for plasma and faecal analysis. The linear ranges were 0.01–5 μg/ml for plasma and 1–300 μg/ml faecal analysis and showed correlation coefficients of 0.999 and 0.998, respectively. The mean recovery of PYR from plasma and faecal samples were 72.72% and 83.56%, respectively. The detection limit of the analytical technique was 0.002 μg/ml and 0.25 μg/ml; the quantification limit was 0.01 μg/ml and 1.00 μg/ml for plasma and faecal samples, respectively. The inter and intra-assay precisions of the analytical procedure obtained after HPLC analysis of spiked standards of PYR (0.01–5 μg/ml) showed a CV of 4.22% and 6.54% for plasma and 6.12% and 7.55% for faecal samples, respectively.

Mean pharmacokinetic parameters of PYR, administered orally as paste and granule, are shown in Table 1, with the plasma concentration *vs.* time curves in Fig. 1. PYR was detected in plasma samples between 2 h and 36 h after paste administration and 1 h and 48 h after granule administration. The last detectable plasma concentration of paste formulation was significantly earlier (36.00 h) compared with granule formulation (46.29 h). Although, there was no significant difference on terminal half lives ($t_{1/2}$: 12.39 h *vs.* 14.86 h), t_{max} (14.86 h *vs.* 14.00 h) and MRT (24.80 h *vs.* 25.44 h) values; the C_{max} (0.09 μg/ml *vs.* 0.21 μg/ml) and AUC values (2.65 μg h/ml *vs.* 5.60 μg h/ml) were significantly different in donkeys received paste formulation compared with the animals received granule formulation, respectively. Mean kinetic parameters of dry-faeces concentrations after paste and granule formulations are shown in Table 2 with the dry-faecal concentration *vs.* time curves (Fig. 2). PYR was detected in faecal samples between 16 h and 120 h. Both PYR formulations displayed a similar faecal excretion profiles following oral administration. The highest dry faecal concentrations were 710.46 μg/g and 537.21 μg/g and were determined at 48 h

Table 1
Mean (±SD) pharmacokinetic parameters of pyrantel (PYR) pamoate paste and granule following *per os* administration to donkeys at 20 mg/kg B.W. (equals 6.94 mg/kg PYR base) bodyweight.

Parameters	PYR groups	
	Paste	Granule
$t_{1/2\lambda z}$ (h)	12.39 ± 5.35	14.86 ± 5.59
C_{max} (μg/ml)	0.09 ± 0.02*	0.21 ± 0.07
t_{max} (h)	14.86 ± 5.52	14.00 ± 9.45
t_{last} (h)	36.00 ± 0.00*	46.29 ± 4.53
AUC_{last} (μg h/ml)	2.65 ± 0.81*	5.60 ± 0.59
$AUMC_{last}$ (μg h^2/ml)	68.30 ± 36.04*	143.68 ± 64.72
MRT_{last} (h)	24.80 ± 5.54	25.44 ± 10.68

C_{max}: peak plasma concentration; t_{max}: time to reach peak plasma concentration; AUC_{last}: area under the (zero moment) curve from time 0 to the last detectable concentration, $AUMC_{last}$: area under the moment curve from time 0 to *t* last detectable concentration; MRT_{last}: mean residence time; $t_{1/2\lambda z}$: terminal half-life.
Mean kinetic parameters obtained following PYR paste administration significantly different (* $P < 0.01$) from those obtained following PYR granule administration.

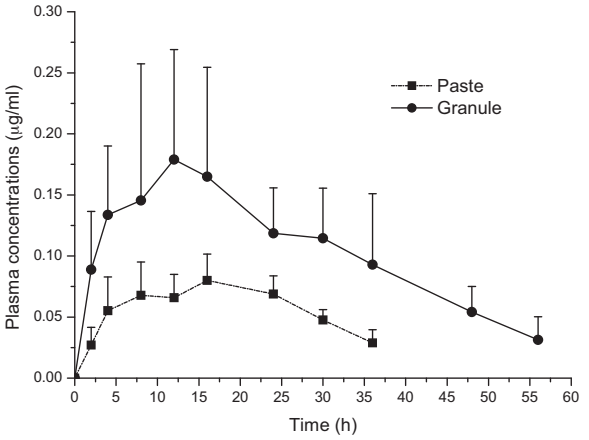

Fig. 1. Mean (±SD) plasma concentration (μg/g) of pyrantel (PYR) pamoate paste and granule following oral administration to donkeys at a dose rate of 20 mg/kg B.W. (equals 6.94 mg/kg PYR base) bodyweight.

Table 2
Mean (±SD) faecal kinetic parameters of pyrantel (PYR) pamoate paste and granule following *per os* administration to donkeys at 20 mg/kg B.W. (equals 6.94 mg/kg PYR base) bodyweight.

Parameters	PYR groups	
	Paste	Granule
t_{max} (h)	46.00 ± 9.73	47.43 ± 5.86
C_{max} (μg/g)	839.13 ± 212.98	650.39 ± 264.00
t_{last} (h)	102.86 ± 11.71	109.71 ± 12.83
AUC_{last} (μg h^2/g)	29,765.56 ± 4687.66	26,414.17 ± 8184.14
MRT_{last} (h)	48.43 ± 5.89	53.68 ± 8.18

C_{max}: peak faecal concentration; t_{max}: time to reach peak faecal concentration; AUC_{last}: area under the (zero moment) curve from time 0 to the last detectable concentration; t_{last}: time to last detectable faecal concentration; MRT_{last}: mean residence time;

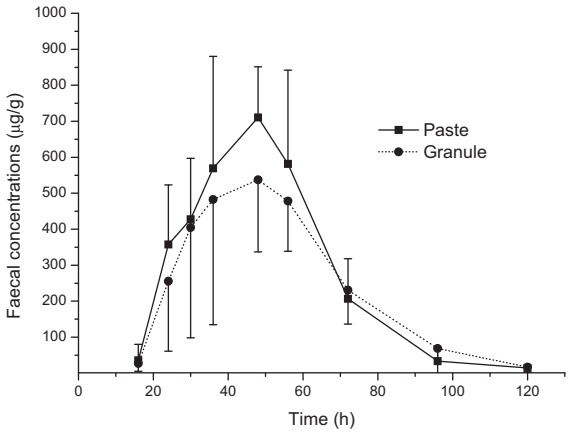

Fig. 2. Mean (±SD) dry-faecal concentration (µg/g) of pyrantel (PYR) pamoate paste and granule following oral administration to donkeys at a dose rate of 20 mg/kg B.W. (equals 6.94 mg/kg PYR base) bodyweight.

for both paste and granule formulation of PYR in donkeys, respectively.

Clinically, no adverse reaction was observed in any of the donkeys treated with PYR during the study. Pretreatment EPG of 1104, 1061 and 1139 were observed for the control, PYR paste and PYR granule groups, respectively. At the start of the study the pretreatment EPG were not significantly different ($P > 0.1$) between groups. Total EPG counts and efficacy values for both treated groups compared with the control untreated group (C-group) at each time study points are shown in Table 3. The results of the post-treatment EPG for both PYR treatment groups were significantly different ($P < 0.001$) from the control group until day 35 following treatments. The PYR formulations were efficient (>95% efficacy) on days 7, 14, 21 until day 28. There was no significant difference in anthelmintic efficacy between the two PYR formulations.

In all studied donkeys, coprocultures performed at day−3 revealed the presence of Cyathostomes and *S. vulgaris*. Faecal cultures performed on different days from C-group confirmed the presence of the same genera. Coprocultures from treated animals revealed the presence of few larvae of Cyathostomes in both PYR groups.

4. Discussion

Although, PYR has been used as an anthelmintic drug for almost a half century, there is a paucity of data available in the literature on the pharmacokinetics and comparative efficacy of different formulations in domestic animals including equid species. Therapeutic equivalence was demonstrated for PYR pamoate oral formulations paste and granule based on FECR test. In this study, both PYR formulations were highly effective against the intestinal strongylidae of donkeys until day 28 post treatment. The cutoff values (mean percent reduction in FEC) for interpreting results of strongyle FECRT suggested in the guidelines of the AAEP for pyrantel, reported a range of 94–99% effectiveness expected in the absence of resistance in horses. The cyathostomin and strongylid nematodes present in the studied donkeys were apparently susceptible to pyrantel pamoate, as confirmed by FECR values. The percentage reductions in faecal egg counts for the PYR paste group, compared to the C-group, were 98.3% on day 7; 98.5% on day 14; 98.2% on day 21 and 96.6% on day 28.

The percentage reductions in faecal egg counts for the PYR granule group, compared to the C-group, were 97.1% on day 7; 97.3% on day 14; 96.3% on day 21 and 96.4% on day 28. The values of efficacy in the present study performed on the donkey are close to those suggested (92%) by Nielsen et al., 2013b to evaluate the expected efficacy of pyrantel against strongyle parasites in horses. The Egg Reappearance Period (ERP) of PYR in the horses is not expected to exceed 4 weeks (Reinemeyer and Nielsen, 2013); the ERPs of PYR pamoate paste and granule formulations following *per os* administration in donkeys are fairly similar to that noted in horses with PYR-susceptible populations of nematodes and treated at similar dosages.

The results obtained in both treated groups are consistent with the currently approved label claim for PYR oral formulations against intestinal strongyle parasites of horses. The mode of administration of PYR would not seem to alter the anthelmintic efficacy in donkeys.

Even though, the pharmaceutical formulation of anthelmintic drugs may affect its bioavailability, which depends on the rate and extent of absorption of the drug from the gastrointestinal tract into the bloodstream. The pharmacokinetic results of the present study obtained after *per os* administration of the paste formulation differ substantially from the granule formulation of PYR in donkeys at the same dose rate. The plasma level of PYR is much lower following administration of paste formulation than those observed following granule administration. After oral administration, C_{max} (0.09 µg/ml) was significantly lower ($P < 0.01$) and AUC (2.65 µg h/kg) smaller ($P < 0.01$) for PYR paste than PYR granule (C_{max}: 0.21 µg/ml, AUC: 5.60 µg h/kg) given at same dose rates (6.94 mg/kg B.W.). It is likely that the poorer dissolution of paste formulation reduces its absorption compared with granule formulation in donkeys.

The results of PYR in the present study differ substantially from those previously reported by Gokbulut et al. (2001) in horses at same formulation and administration route. In the previous study, PYR paste was administered at 13.3 mg/kg in horses, whereas in this study, it was administered at 6.94 mg/kg in donkeys. The lower concentrations and shorter MRT of PYR in the plasma of horses reflect lower bioavailability and shorter persistence compared with donkeys. After correction for dose assuming proportionality, C_{max} (0.045 µg/ml), AUC (0.53 µg h/ml) and MRT (11.99 h) values in horses much lower than those observed in the present study (C_{max}: 0.09 µg/ml, AUC: 2.65 µg h/ml and MRT: 24.80 h). The gastrointestinal passage rate of digesta is affected by alteration in the quality and quantity of the feed consumed and this could confer variable absorption time and therefore bioavailability of drugs administered orally. It was indicated that variations in the quality (McKellar et al., 1993b; Knox and Steel, 1997; Oukessou and Souhaili, 1998; Gokbulut et al., 2007) and quantity (Ali and Hennessy, 1995; Lifschitz et al., 1997; McKellar et al., 2002; Gokbulut et al., 2010) of diet

Table 3
Strongyle egg counts in eggs per gram (EPG) and percentage reductions in faecal egg counts (FECR) for donkeys treated with pyrantel pamoate paste (PYR P-group) and for donkeys treated with pyrantel pamoate granule (PYR G-group), compared with control untreated group (C-group) at each time study points.

Day	C group	PYR Paste group			FECR (%)	*P* value	PYR Granule group		FECR (%)	*P* value
	EPG AM	EPG Range	EPG AM	EPG Range			EPG AM	EPG Range		
3	1104	450–1470	1061	160–1770	–	0.8636	1139	170–2440	–	0.9220
7	849	450–1170	14	0–30	98.3	0.0000	24	0–80	97.1	0.0000
14	936	500–1340	14	0–30	98.5	0.0000	26	0–10	97.3	0.0000
21	1036	140–1450	19	0–30	98.2	0.0001	39	0–110	96.3	0.0001
28	1334	170–2030	46	0–110	96.6	0.0002	49	20–80	96.4	0.0002
35	1163	610–2000	213	20–740	81.7	0.0003	124	40–290	89.3	0.0001
42	854	90–1370	520	40–1790	39.1	0.2657	303	70–540	64.5	0.0095
49	823	90–1680	616	10–1950	25.2	0.5055	533	150–1090	35.2	0.1925
56	663	210–1130	737	80–1410	−11.2	0.7371	651	240–1410	1.7	0.9536

*Parametric *t*-test (PYR-groups *vs.* C-group) values are significantly different $P < 0.001$—arithmetic means (AM).

could affect the bioavailability of anthelmintics in different animal species. Differences among the studies may be associated with diet type in donkeys compared with horses. Since in the present study, donkeys were kept indoors and fed hay-based diet. The horses were yarded for the period during and immediately (4 h) after drug administration and were then returned to a grass paddock (Gokbulut et al., 2001). Grass-based diet probably decreased gut transit time of food, thus the rapid passage of food in the gut causes a decrease in the bioavailability of PYR in horses compared with those in donkeys in the present study. Moreover, anatomic features influence the passage of digesta, and the bioavailability and pharmacokinetics of anthelmintics may be affected by different gut transit time in animal species (McKellar and Scott, 1990).

Parasitism could have had an effect since the donkey used in this study were naturally infected with intestinal parasites and such infections may have modest effects on absorption of anthelmintics (McKellar et al., 1991), unfortunately the parasitological status of the horses or the history of anthelmintic treatment in the previous study was unknown.

PYR is not known to have any substantial effect on faecal invertebrates, however, it is apparent that faeces from treated donkeys will have high concentrations of PYR for at least 120 h for both formulations. This period of faecal excretion of PYR is considerably longer in donkeys compared to in the horses (48 h) (Gokbulut et al., 2001).

5. Conclusion

The present study demonstrates relatively limited absorption of PYR pamoate following *per os* administration as paste or granule formulations in donkeys. Although the higher level and longer persistence of PYR in the gastrointestinal tract where the adult stages of most parasitic nematodes reside after both formulations may provide higher and prolong efficacy, low plasma levels could result in subtherapeutic plasma concentrations. Moreover, the donkey differs from the horse and requires species specific pharmacological and parasitological studies. Therefore, further research should be carried out to determine the optimal dosage and dose scheme for antiparasitic compounds in donkeys.

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