Clinical and clinicopathological changes in 6 healthy ponies following intramuscular administration of multiple doses of imidocarb dipropionate

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ABSTRACT

Haematological variables and selected serum indices, particularly those affected by changes in renal and hepatic function, were examined in 6 healthy ponies following 4 intramuscular doses of 4 mg/kg imidocarb dipropionate administered every 72 hours. This treatment regime has been reported to sterilise experimental *Babesia equi* infections in horses and may have value in preventing the spread of this disease during exportation of possible carrier horses to non-endemic countries. Serum bile acids and serum gamma glutamyltransferase activity were measured to evaluate the effect of this treatment regime on hepatic function. Owing to the absence of any increase in these variables it was concluded that this treatment regime had no clinically detectable deleterious effect on hepatic function in healthy ponies. Urinary gamma glutamyltransferase : creatinine ratios (IU/g), serum creatinine and fractional clearance of sodium, potassium and phosphate (%) were calculated as a measure of renal function. Urinary GGT and urinary GGT : creatinine ratios were significantly elevated on Day 5 of the trial, with 2 of the trial animals also exhibiting mild azotaemia indicative of changes in renal function. The changes in urine GGT : urine creatinine ratios observed in this study also provides evidence of the value of this ratio for the early detection of renal toxicity, following exposure to nephrotoxic agents.

Key words: bile acids, creatinine, equine, fractional clearance, gamma glutamyl transpeptidase, imidocarb dipropionate.

Meyer C, Guthrie A J, Stevens K B **Clinical and clinicopathological changes in 6 healthy ponies following intramuscular administration of multiple doses of imidocarb dipropionate**. *Journal of the South African Veterinary Association* (2005) 76(1): 26–32 (En.). Equis Veterinary Practice, PO Box 2155, Hillcrest, 3650 South Africa.

INTRODUCTION

Equine piroplasmosis is a tick-transmitted, disease caused by the intra-erythrocytic protozoa *Babesia equi* and *Babesia caballi.* The geographical distribution of this disease is limited to regions in which the tick vectors occur[14,37,42]. To prevent the spread of infection to unexposed, susceptible equids, endemic countries are subject to rigid exportation restrictions with substantial economic implications for equine industries. The spread of infection to non-endemic regions could be contained if a suitable and safe sterilisation protocol became instituted to eliminate infection in equids destined for exportation. Imidocarb dipropionate [3, 3′ bis-(2-imidazolin-2-yl) carbanilide dipropionate] and the less readily available dihydrochloride derivative are aromatic diamidines used as chemotherapeutic agents for the treatment of *Babesia equi* and the less patho-genic *Babesia caballi* infections in equids[12,19,29,30]. A study performed by Frerichs *et al.* imidocarb dihydrochloride was used experimentally to sterilise *Babesia equi* infections in horses and donkeys[19,20]. They showed that only those animals treated with 4 doses of 4 mg/kg imidocarb dihydrochloride at 72-hour dosage intervals were successfully sterilised. Further, animals treated with the same dose and number of treatments, but at different dosage intervals, failed to eliminate the infection, which demonstrated that timing of administration of imidocarb is crucial when attempting to sterilise piroplasmosis.

Dose-dependent nephrotoxic and hepatotoxic effects of imidocarb have been documented in sheep, goats, cattle, dogs and horses[2]. In a study by Adams, increasing levels of imidocarb were correlated with increasing rates of morbidity and mortality attributed to acute renal cortical tubular necrosis and acute periportal hepatic necrosis[2]. Although in the study by Frerichs *et al.* the anticholinesterase effects of imidocarb, including salivation, restlessness, moderate colic and hypermotility of the gastro-intestinal tract as well as local reactions at the injection sites were described, clinicopathological tests addressing the potential nephro- and hepatotoxic effects of the drug were not performed[20].

The primary aim of this study was to examine the clinical and clinicopathological effects of a multiple dose treatment regime of imidocarb dipropionate in healthy ponies with particular emphasis on evaluation of renal and hepatic function.

MATERIALS AND METHODS

Experimental animals

Three healthy Nooitgedacht ponies and 3 part-bred Welsh Mountain ponies aged 2–4 years and weighing 244–294 kg (mean 272.5 kg) were used for the trial. The 3 Nooitgedacht ponies and 2 of the part-bred Welsh Mountain ponies were mares. The remaining part-bred Welsh Mountain pony was a gelding. The animals were maintained in a tick-free isolation unit at the Faculty of Veterinary Science, University of Pretoria, Onderstepoort, for the duration of the trial.

The 6 ponies used in the trial were introduced into their new environment approximately 1 week prior to the commencement of treatment. Two of the ponies were stabled individually and the remaining ponies were housed in pairs in large $4 \times 5\,m^2$ box stalls. The stalls were in an insect-proof, artificially ventilated building with continuous air filtration. The ponies were maintained in these stalls for most of the day and at night. They were turned out into a sand enclosure for a short period during the day to allow them to exercise. Diets were standardised and a routine was established to eliminate the potential effects of nutrition and stress on clinicopathological testing. The animals received mixed grass hay and water ad libitum, as well as 500 g of a 12 % protein pelleted ration (Epol Rider Cubes, Epol (Pty) Ltd, Pretoria, South Africa) twice daily. Clean pine shavings were used for bedding.

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serum and urine indices established that all of the ponies were healthy at the onset of the trial.

Experimental design

The 6 ponies were given 4 intramuscular injections of 4 mg/kg imidocarb dipropionate [3, 3′-bis-(2-imidazol-2 yl) carbanilide dipropionate] (Forray 65, Schering-Plough, Isando, South Africa) at 72-hour dosage intervals. The drug was administered alternately into the left and right cervical muscles with 2 treatments on each side at different sites. Immediately prior to each treatment the ponies were administered 0.02 mg/kg atropine sulphate (Atropine 10, Centaur Laboratories, Bryanston, South Africa) intravenously, to minimise adverse reactions caused by the anticholinesterase effects of imidocarb. Injection sites were examined daily by the same investigator and local reactions were subjectively classified as mild, moderate, severe or not observed. A reaction was classified as mild if the affected area was less than 5 cm in diameter, required firm palpation to elicit pain or resulted in a barely discernible elevation in skin temperature over the affected area. A reaction was termed moderate if the affected area was between 5 and 10 cm in diameter, if pain was evident with gentle palpation or if heat was easily detected over the affected area. A reaction was classified as severe if the local reaction was larger than 10 cm in diameter, the animal resisted or avoided palpation or heat was evident with the hand raised above, but almost touching, the affected area. Oedema was recorded if these areas were seen to 'pit on pressure' while more persistent swellings were firm and were noted as such. The moderate to severe local reactions observed following the 1st intramuscular injection, prompted the investigators to divide the remaining doses in half and administer each half at a different site.

At the onset of the trial and 24 hours after each treatment on Days 2, 5, 7 and 11 as well as on Days 18 and 38, blood samples were collected from the jugular vein of each animal into a serum and an EDTA tube. At the same time a urine sample was collected via sterile urinary catheterisation. To facilitate urinary catheterisation of the gelding, 15 mg acepromazine maleate (Aceprom, Centaur Laboratories, Bryanston, South Africa) was administered intravenously half an hour prior to collection. All parameters were measured on samples collected on Days 0, 2, 5, 7 and 11 with the exception of bile acids, which owing to financial constraints were only measured on Days 0, 5 and 11. In addition, due to its long half-life serum AST was

Table 1: Summary of the incidence and duration of systemic signs observed following intramuscular administration of 4 doses of 4 mg/kg imidocarb dipropionate every 72 hours to 6 healthy ponies.

Systemic signs	Treatment				Duration
	1	2	3	4	
Mild colic	3/6	3/6	3/6	3/6	±30 minutes
Depression	2/6	2/6	2/6	0/6	2–6 hours
Anorexia	0/6	2/6	2/6	0/6	2–6 hours

Table 2: Summary of the incidence and nature of the local reactions that were noted at the injection site following 4 intramuscular treatments of 4 mg/kg imidocarb dipropionate to 6 healthy ponies.

Day	Number of animals	Size (cm)	Heat*	Pain	Oedema	Firm swelling
1	6/6	10 × 10	+ + +	+ +	–	–
2	6/6	15 × 10	+ +	+ + +	+ + +	–
3	5/6	5 × 8	+	+	+ +	–
5	2/6	5 × 5	–	+	–	–
15	2/6	3 × 5	–	–	–	+
30	2/6	2 × 3	–	–	–	+
38	0/6	–	–	–	–	–

* + mild, ++ moderate, +++ severe, – not observed.

also determined on Day 38. All samples were stored at 2–8 °C and were analysed within 24 hours of collection.

Analytical techniques

Blood cell counts were measured using an automated cell counter (Cell-Dyn 3500, Abbott Diagnostics, Abbott Park, USA). Blood smear examinations for babesia parasites were done manually using thin blood smears stained with Cam's Quick-Stain (Milsch, Krugersdorp, South Africa). Urinalyses consisted of macroscopic examination of the urine, urine dipstick analysis (Combur 9 Test, Boehringer, Mannheim, Germany), measurement of urine specific gravity and microscopic evaluation of urine sediment.

Serum and urine creatinine analysis was based on the Jaffe alkaline picrate reaction as described by Rossignol using a Technicon RA-1000 analyser[36] (Technicon Instrument Corporation, Tarrytown, USA). Sodium and potassium were measured using a NOVA Biomedical (Nova Biomedical, Waltham, USA) electrolyte analyser 1 with sodium and potassium ion selective electrodes[16]. Serum and urine inorganic phosphate was measured using the phospho-molybdate reaction as described by Amador and Urban using a Technicon RA-1000 analyser[3]. Serum and urine GGT were analysed by the optimised, kinetic method of Szasz using a Technicon RA-1000 analyser[40].

Total serum protein analysis was based on the biuret reaction of Weichselbaum as described by Skeggs and Hochstrasser using a Technicon RA-1000 analyser[38]. AST analyses was performed using an

optimised, kinetic assay based on the Karmen method described by Bergmeyer et al. using a Technicon RA-1000 analyser[5,7]. A Sigma Diagnostics bile acid reagent kit was used to measure serum bile acid concentrations enzymatically using the method described by Mashige et al. on a Technicon RA-1000 analyser[32]. Glucose concentrations were measured using the hexokinase procedure for glucose determination as described by Léon et al. using an automated analyser (Technicon RA-1000)[31].

Fractional clearances of creatinine, sodium, potassium and phosphate (%) as well as urine GGT : urine creatinine ratios (IU/g) were calculated. The fractional clearance of electrolytes were calculated using the following equation:

$$\% \text{ Clearance Ratio } [x] = \frac{\text{urine } [x] \times \text{plasma (creatinine)}}{\text{plasma } [x] \times \text{urine (creatinine)}} \times 100$$

Statistical methods

The data in this study were analysed using Sigma Stat 2.0 statistical software (Jandel Scientific Software, San Rafael, USA). Significant changes with time within the treatment group were analysed using 1-way analysis of variance (ANOVA) for repeated measures followed by Dunnett's test to examine deviation from pre-treatment values. Where the data were either not normally distributed or the equal variance test failed, the data were analysed using Friedman repeated measures analysis of variance (ANOVA) on ranks followed by Dunnett's method to examine deviations from pre-treatment

values. $P < 0.05$ was considered significant. Data are reported as mean ± standard error; $\alpha = 0.050$.

RESULTS

Clinical manifestations and observations

Mild colic, depression and anorexia were seen in some animals after each treatment. The type and duration of these systemic signs are summarised in Table 1. One of the ponies became severely depressed and anorectic following the administration of the 3rd intramuscular injection. This behaviour coincided with a serum glucose and elevated serum creatinine level of 5.4 mmol/ℓ and 195 μmol/ℓ, respectively (reference ranges 3.5–6.3 μmol/ℓ; 76.8–174.5 μmol/ℓ) [18]. These variables had returned to normal by the following day. The incidence of the systemic reactions following the 1st injection did not differ from the incidence of reactions following subsequent injections. Local reactions were noted at the injection site in all of the animals following the 1st treatment. Distribution of the drug across various sites, as well as the administration of a smaller amount per site, resulted in a marked reduction in the local tissue reactions seen. The characteristics of the local reactions are summarised in Table 2.

Clinicopathological results

The red cell count and haematocrit were significantly lower than their respective pre-treatment values on Days 2, 5 and 18 of the study. No significant changes were observed in the haemoglobin concentration. No significant changes were observed in the total white cell count or in the mature and immature neutrophil counts at any time during the trial. The lymphocyte count was significantly lower than the pre-treatment value on Day 5 while the eosinophil count was significantly reduced on Day 18. No significant changes were observed in monocyte counts. The total serum protein concentration was significantly lower than the pre-treatment values on Day 11 and 18. Serum aspartate aminotransferase activity was significantly elevated above pre-treatment values on Days 2, 5, 8, 11 and 18, but had returned to pre-treatment levels by Day 38 of the trial (Fig. 1). Serum and urine creatinine concentrations did not differ significantly from the pre-treatment value at any time during the trial although there appeared to be a trend towards an elevated creatinine concentration and 2 of the 6 treated animals developed mild transient azotaemia (Fig. 2). The serum sodium
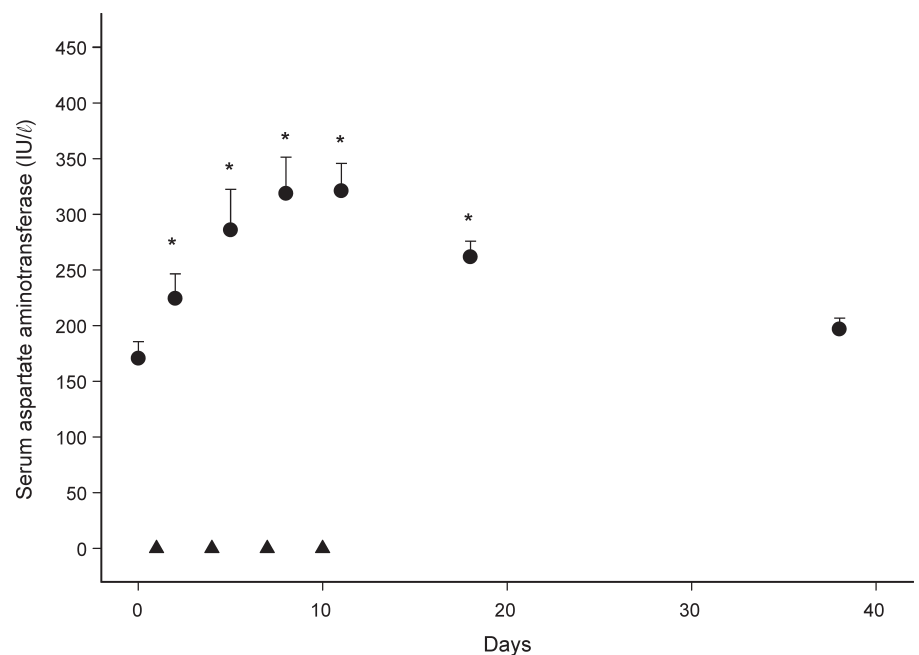

Fig. 1: **Temporal changes of mean and standard error of aspartate aminotransferase activity** (IU/ℓ) **in 6 ponies following intramuscular administration of 4 mg/kg imidocarb dipropionate on Days 1, 4, 7 and 10. ▲ indicates days on which imidocarb was administered. * = mean at time indicated was significantly different** ($P < 0.05$) **from the pre-treatment value.**
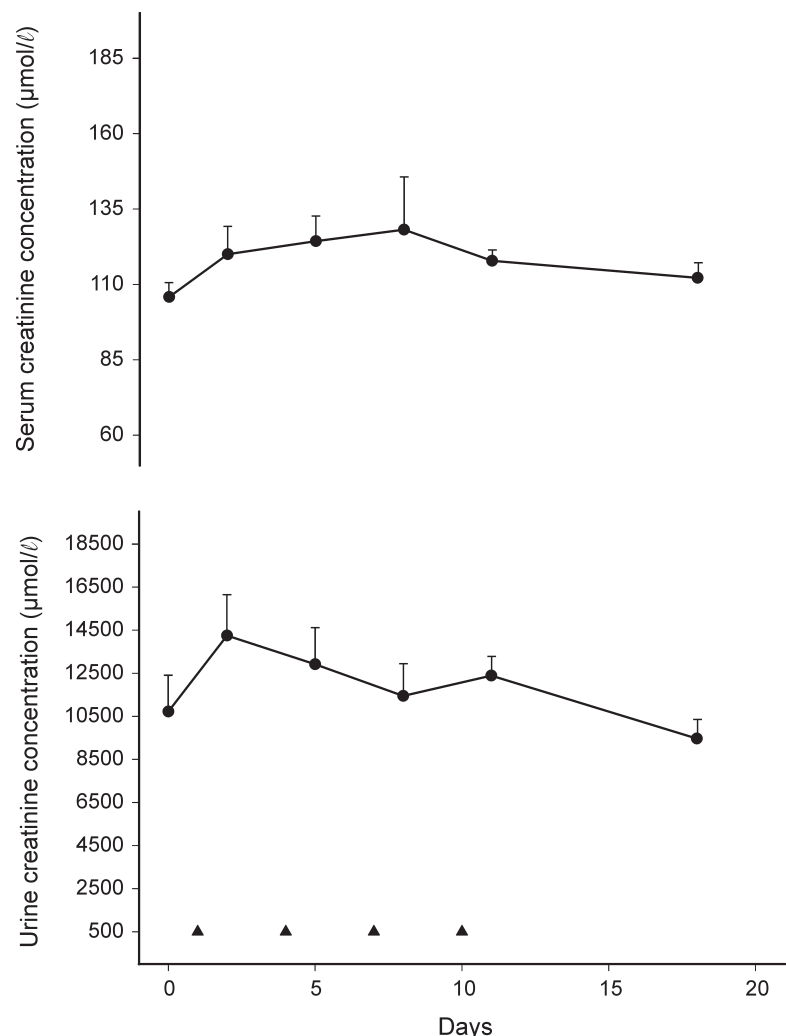

Fig. 2: **Temporal changes of mean and standard error of serum creatinine** (mg/dℓ) **and urine creatinine concentration** (mg/dℓ) **in 6 ponies following intramuscular administration of 4 mg/kg imidocarb dipropionate on Days 1, 4, 7 and 10. ▲ indicates days on which imidocarb was administered.**

concentration was significantly lower than the pre-treatment value on Day 5 of the trial. There were no significant differences between the fractional clearance of sodium (reference range 0.032–0.52)[10], the serum potassium concentration, the fractional clearance of potassium (reference ranges 23.3–48.1)[10], the serum inorganic phosphate or the fractional clearance of phosphate (reference ranges 0–20)[10] at any time during the trial and their respective pre-treatment values. The serum and urine GGT activities and urine GGT : urine creatinine ratios were all significantly elevated above pre-treatment values on Day 5. The serum GGT activity was also elevated on Day 18 of the trial (Fig. 3). The total bile acid concentration was significantly lower than the pre-treatment values on Day 5 of the trial (Fig. 4). No significant changes were observed in urine specific gravity during this trial. In contrast, urine pH was significantly lower than the pre-treatment value on Day 18 of the trial (Table 3). No abnormalities or changes were detected on macro- or microscopic examination of the urine and urine sediment. Glucosuria was present in 2 of the animals following the 3rd intramuscular injection. The concurrent serum glucose and serum creatinine levels in these animals were 5.2 μmol/ℓ and 5.4 μmol/ℓ, and 168 μmol/ℓ and 195 μmol/ℓ, respectively (reference ranges 3.5–6.3 μmol/ℓ; 76.8–174.5 μmol/ℓ)[18].

DISCUSSION

The use of enzymuria in screening for renal disease has been actively pursued in human medicine and is considered a non-invasive, sensitive indicator of early renal dysfunction[5,21,24]. Elevations in urine GGT activity are considered to be renal in origin, as the high molecular weight of this enzyme prevents its filtration from the blood by the normal glomerulus[24]. Therefore no correlation exists between increased serum GGT activity and increased urine GGT activity originating from the brush border of the proximal renal tubular epithelium[24].

In horses, urine GGT : urine creatinine ratios less than 25 IU/g are considered normal, but the small sample sizes on which these estimates are based may not be representative of a larger population and there is some indication that ratios between 25 and 100 IU/g should be interpreted with care[24,35]. In this study, the serial determination of this variable allowed comparison with individual baseline activity with the significant elevation observed on Day 5, confirming renal involvement. However, the rapid return to previous baseline values supported reported observations that
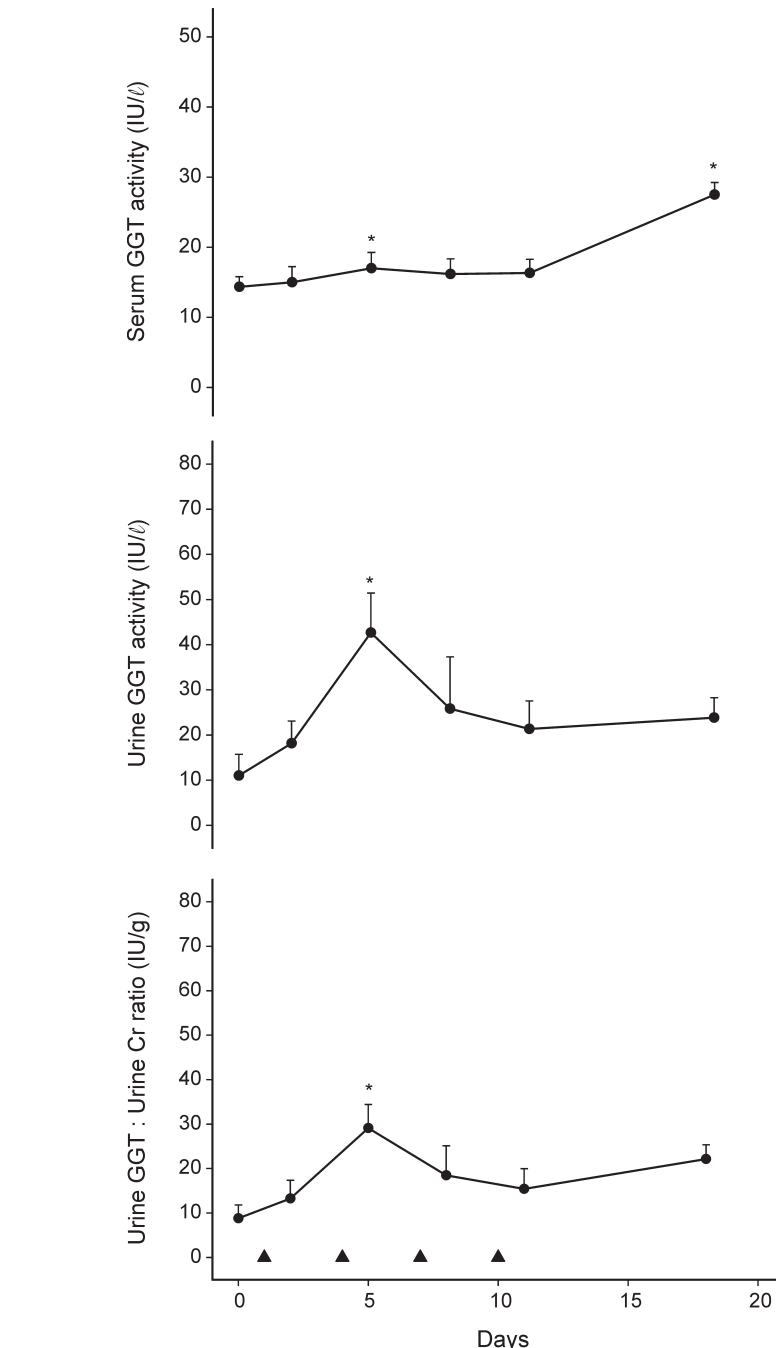

Fig. 3: **Temporal variation of mean and standard error of serum gamma glutamyl transpeptidase** (GGT) (IU/ℓ), **urine GGT** (IU/ℓ) **and the urine GGT : urine creatinine ratios** (IU/g) **in 6 ponies following intramuscular administration of 4 mg/kg imidocarb dipropionate on Days 1, 4, 7 and 10. ▲ indicates days on which imidocarb was administered. * = mean at time indicated was significantly different** ($P < 0.05$) **from the pre-treatment value.**

changes between 25 and 100 IU/g may be a function of drug excretion and are not necessarily indicative of significant nephrotoxicity[5,21,24,35]. The lack of evidence of significant renal damage may, however, in part be due to the extensive regenerative capacity of the kidney[11]. Consequently, in response to these changes, reduction of dose or frequency of drug administration may prevent manifestation of clinical nephrotoxicity. This conclusion can be supported by Adams' study which showed that increasing levels of imidocarb were correlated

with increasing local and systemic reactions, increasing levels of blood urea nitrogen, serum aspartate amino transferase, serum sorbitol dehydrogenase, serum creatine phosphokinase, neutrophilia and increasing severity of renal, hepatic and pulmonary lesions. Mortalities were attributed to acute renal cortical tubular necrosis and acute periportal hepatic necrosis induced by 2 injections of 16 or 32 mg/kg of imidocarb dipropionate[2].

In concurrence with previous studies, the urine GGT : urine creatinine ratio
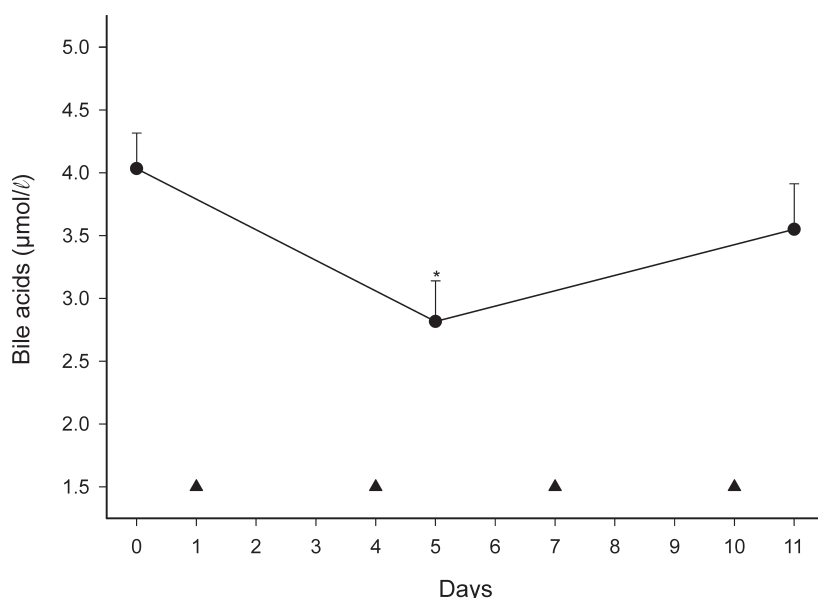
Fig. 4: **Temporal changes in mean and standard error of serum bile acids (μmol/ℓ) in 6 ponies following intramuscular administration of 4 mg/kg imidocarb dipropionate on Days 1, 4, 7 and 10. ▲ indicates days on which imidocarb was administered. * = mean at time indicated was significantly different** ($P < 0.05$) **from the pre-treatment value.**

therefore appears to be one of the earliest practical indicators of renal involvement and may thus have clinical value in the therapeutic monitoring of potentially nephrotoxic substances, including imidocarb derivatives[5]. Serial monitoring of renal function may be particularly significant in diseased animals as it has previously been shown that significant alterations in the disposition kinetics of imidocarb were evident in diseased goats.[1] In these goats, changes in the volume of distribution and drug clearance, related to the pathophysiology and febrile reactions of various experimentally induced disease states, were reported[1]. Altered drug disposition increases the susceptibility of treated animals to the potential toxic effects of various substances[1,39].

The effect of this treatment regime on liver function was examined by sequential analysis of serum bile acid and serum GGT concentrations. Bile consists of several components of which bile acids make up 90 % of the organic portion[4]. These bile acids act as detergents, which facilitate the uptake of lipid soluble compounds from the gastrointestinal tract as well as excretion of cholesterol and phospholipids from the liver[4,34]. Bile acids are produced solely by the liver and elevation of total plasma bile acid concentration is considered a highly sensitive and very specific indicator of hepatic disease and provides a measure of hepatobiliary transport function[33,39].

The majority of bile acids are restricted to the enterohepatic circulation resulting in normally low plasma bile acid concentrations[43]. A study by Hoffman *et al.* determined reference values for serum bile acid concentrations from 15 fasted horses to be 6.06 ± 2.56 μmol/ℓ, from 5 ponies to be 5.64 ± 3.09 μmol/ℓ, and for all equids, 5.94 ± 2.27 μmol/ℓ[25]. There was no significant difference between the mean bile acids of ponies compared with values for horses[25]. It has been estimated that bile acids are circulated approximately 38 times per day between the gastrointestinal tract and the liver in healthy ponies[25,34]. The age and sex of animals does not influence bile acid concentrations, nor is there a significant diurnal variation in these concentra-

tions[43]. There is also no significant difference in results of bile acid concentrations obtained from serum or plasma samples and examination of either blood component is acceptable[43]. An additional analytical advantage is that plasma bile acids remain stable during storage at –20 °C for at least 1 month, enhancing the practicality of this test[43].

Serum gamma glutamyl transpeptidase occurs as a membrane-bound enzyme on the canalicular surface of hepatocytes and on the biliary epithelial cell membrane. The only other potential source of serum GGT is the pancreas, but pancreatic disease in equids is rare and an elevated serum GGT is more likely to be of hepatic origin[8,14,20,34]. There is no correlation between serum bile acid concentrations and serum GGT activity[34]. In contrast to serum bile acids, which may increase significantly within 24–48 hours of acute hepatic damage, increases in serum GGT are more consistent in chronic disease[8,20,34]. Similarly, fasting for 2 to 4 days results in elevations of serum bile acids to approximately 3 times base-line concentrations, with little or no effect on serum GGT activity[14]. Thus, although not as sensitive as serum bile acids, serum GGT activity may be used in combination with measurement of bile acids and a particular diagnosis is made only if all the tests performed are positive[22]. This method of testing increases the specificity or number of animals without a disease that test negative. The positive predictive value, or probability of a disease being present in an animal that tests positive, is also increased[22]. Normal serum GGT activity may thus rule out suspected hepatic damage in animals with elevated serum bile acid concentrations which have fasted for longer than 3 days[20,25]. The significant increase in serum GGT in conjunction with decrease in serum bile acid concentrations observed on Day 5 may have been due to drug-induced hepatic enzyme induction. Clearly, further studies would be required to elucidate such speculation. Total plasma bile acids have been shown to be the most sensitive indicator of hepatic disease compared with other liver function tests[17,22,33,43]. Owing to the absence of any increase in this variable, it was concluded that this treatment regime had no detectable deleterious effect on hepatic function in healthy ponies.

Fractional clearance of phosphate (FC_P) is reportedly a sensitive indicator of early renal dysfunction in horses and elevations in this variable precede increases in fractional clearance of sodium (FC_{Na}) in this species[6,9,15,27,28,41]. The absence of significant increases above pre-treatment con-

Table 3: **Mean and standard error of the urine specific gravity and urine pH and incidence of glucosuria measured on Days 0, 2, 5, 8, 11 and 18.**

Day	Urine specific gravity (Mean ± standard error)	Urine pH (Mean ± standard error)	Glucosuria
0	1.030 ± 0.00258	8.17 ± 0.31	0/6
2	1.030 ± 0.00258	8.83 ± 0.17	0/6
5	1.030 ± 0.00307	8.83 ± 0.21	0/6
8	1.040 ± 0.00333	7.83 ± 0.17	2/6
11	1.040 ± 0.00211	7.67 ± 0.33	0/6
18	1.040 ± 0.00167	7.33 ± 0.33*	0/6

*Mean at time indicated was significantly different ($P < 0.05$) from the pre-treatment value.

centrations for either variable suggests that the nephrotoxic effects of imidocarb dipropionate may be limited at this dose and dosage interval.

In horses, elevations above normal, or an increase in serum creatinine within the normal reference range of greater than 26.5 μmol/ℓ in the absence of an alternative underlying cause of renal damage, is considered an indicator of nephrotoxicity[24]. Nephrotoxicity in man has similarly been associated with elevations in serum creatinine concentration greater than 44.0 μmol/ℓ[24]. Elevated serum creatinine levels outside the normal reference range were observed in 2 of the ponies on Day 11 of the trial. Changes in concentrations of this variable from pre-treatment values for the group were, however, not significant and supported the observation that the renal effects of this treatment regime were limited, although individual variation may occur. Glucosuria has also been documented as a non-specific indicator of renal disease[5].

There should be no glucose present in normal urine, although in the presence of serum hyperglycaemia the renal threshold for glucose is exceeded resulting in glucosuria in the absence of underlying renal disease[13]. In this study the glucosuria, in the presence of normal serum glucose levels and elevated serum creatinine levels, was probably a consequence of the drug-induced renal changes. As previously reported, the poor sensitivity and specificity of these 2 variables render them inappropriate as individual spot tests for detection of early renal dysfunction[13,23,24].

Although in the trial performed by Frerichs *et al.*, imidocarb dihydrochloride was used, the locally available aromatic diamidine, imidocarb dipropionate was substituted for the less readily available dihydrochloride derivative. This may, however, be preferable, because while these 2 compounds have the same active ingredient, the dipropionate salt [3, 3'-bis-(2-imidazol-2 yl) carbanilide dipropionate] is reported to be less irritating[20,29]. All ponies in this study showed uniform moderate to severe local responses at the site of injection following the administration of multiple doses of imidocarb dipropionate. Focal myonecrosis induced by repeated intramuscular injections of imidocarb dipropionate may explain the transient elevation in aspartate aminotransferase (AST) activity seen in this trial. AST is also present in liver and heart tissue and examination of sera for AST enzyme activity in the absence of additional serum biochemistry, is not a specific indicator of tissue source[6]. The plasma half-life of AST in the horse is 2–4 days with persistent eleva-

tions indicative of ongoing damage and a continuous release of isoenzymes[6]. The absence of an ongoing myonecrosis in this study could be confirmed by the return of AST levels to within normal limits (reference range: 10–240 U/ℓ)[18] approximately 2 weeks after administration of the final intramuscular injection.

Although this drug is routinely associated with adverse effects such as salivation, restlessness, moderate colic and hypermotility of the gastro-intestinal tract the results of this trial, in concurrence with previous reports, showed that administration of parasympatholytics such as atropine sulphate significantly reduced these effects[12,29,30]. In addition, varying injection sites and administering smaller amounts of drug per site substantially reduced the mild to moderate local reactions previously reported with this drug[12].

The haematological results obtained in this study indicated that the dosage regime of imidocarb dipropionate used had minimal effect on cellular blood components. The changes in haematological variables, including the mild leukocytosis and concurrent lymphocytosis observed were considered a normal response for healthy, young and excited animals and often occur when a struggle develops during restraint for sample collection[26]. This physiological leukocytosis occurs in response to adrenaline release during which the marginal pools of neutrophils and/or lymphocytes are mobilised into the general circulation raising the absolute neutrophil and/or lymphocyte counts[26]. However, the increase in white blood cell parameters may also have been due in part to a response to the transient myonecrosis occurring at the injection sites[26]. The decrease in total eosinophils over time was interesting and may have been due to the limited exposure of the animals to insects or allergens while housed in an insect-proof, artificially ventilated building with continuous air filtration. This observation may be supported by a concurrent significant decrease in total serum proteins, which in turn may have resulted from a decrease in antigenically stimulated immunoglobulin production[26].

In conclusion, mild changes in indicators of renal function observed in this study were indicative of renal involvement. The nephrotoxic properties of imidocarb have been reported in previous studies and although the renal changes were mild and transient, the potentially harmful effects of imidocarb dipropionate should not be disregarded as individual variation may occur[24]. These effects may also be of particular signifi-

cance when prior renal or generalised cardiovascular compromise is evident. The severe hepatotoxic effects of imidocarb previously reported were not evident at the dose and dosage interval used in this study.

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