# **Evaluation of taurine status in cats consuming diets containing different amounts of taurine by determination of plasma and whole blood taurine concentrations**

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We report the whole blood and plasma taurine concentrations in cats and kittens fed only a taurinesupplemented or a taurine-free diet for various lengths of time up to several years. The results are reported and discussed as a function of age (time consuming the diet), as whole blood concentration as a function of plasma concentration, and as whole blood/plasma ratio as a function of age. The most striking changes were a decrease in plasma taurine concentrations and an increase in whole blood/plasma ratio in taurinedeficient cats and kittens as a function of age. We conclude that measurement of both parameters provides the best assessment of taurine status of felines.

Keywords: taurine; plasma; whole blood; whole blood-plasma ratio; cats

## Introduction

Interest in the nutritional importance of taurine has greatly increased over the last two decades. It has been shown beyond all doubt that taurine is an essential nutrient for the cat, and without an adequate dietary supply, cats become taurine-depleted and suffer from a wide range of abnormalities, including retinal degeneration, dilated cardiomyopathy, and poor reproductive performance.<sup>1,2</sup> There is an increasing body of evidence that suggests primates, including humans, may have a dietary requirement for taurine under certain circumstances, particularly during development.<sup>1,2</sup> We have previously reported taurine concentrations in a wide variety of tissues from cats consuming diets containing different amounts of taurine and from infant monkeys fed from birth with a human infant formula (taurine-free) with and without taurine supplementation.<sup>3-6</sup> However, such measurements were only possible when animals were killed, and monitoring of taurine status was performed by measuring plasma taurine concentrations. Recent studies in humans have indicated that plasma taurine concentrations can vary greatly and concluded that measurement of whole blood and plasma taurine concentrations provide the best estimate of taurine status.<sup>7,8</sup> For the past 2 years we have been following this advice for monitoring taurine status in cats from our colony consuming diets containing different amounts of taurine. We report our findings in this article.

#### Methods and materials

Female domestic cats raised in the colony of the Institute for Basic Research in Developmental Disabilities and vaccinated against rhinotracheitis, panleukopenia, calici virus [FVR-C-P (MLV) Pitman-Moore, Washington Crossing, NJ USA] were fed a completely defined, taurine-free purified diet<sup>3</sup> (Bioserve, Frenchtown, NJ USA) alone or containing 0.05% taurine. Kittens remained with their mothers until they were killed, or until 8 weeks after birth when they were weaned onto the same diet as their mother. Only female kittens were retained beyond weaning. Blood samples were obtained from the jugular vein under light ketamine anes-

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thesia (Ketaset, Aveco Company, Fort Dodge, IA USA) with a plastic syringe containing heparin (LyphoMed, Melrose Park, IL USA). Other samples were obtained by direct heart puncture when cats or kittens were killed for other experimental protocols. Plasma was separated by centrifugation at 1800  $\times$  g for 15 min at room temperature. Portions of whole blood and plasma were deproteinized using 5 vols 20% trifluoroacetic acid (TFA) and centrifuging at 20,000  $\times$  g for 30 min. The clear supernatant was passed through a 0.45 µm µStar filter (Costar, Cambridge, MA USA) and stored at -70° C until used for taurine measurements. Taurine analysis was accomplished by derivatizing with phenylisothiocvanate and separated by reverse-phase high performance liquid chromatography (HPLC).9 The apparatus consisted of a Spectra Physics 8800 Ternary HPLC pump (Piscataway, NJ USA) and a 4.6 mm  $\times$  25 cm Baker Bond C-18 column (Baker, Phillipsburg, NJ USA) maintained at 34° C. The taurine derivative was detected at 254 nm with LDC SpectroMonitor D (Milton Roy, Riviera Beach, FL USA) and quantified using Nelson Analytical 2600 chromatography software (Cupertino, CA USA) with an IBM PC-AT (New York, NY USA). A refrigerated Waters 712 WISP (Milford, MA USA) maintained at 5° C allowed the automatic analysis of up to 96 samples.

Standard deviations and significance of difference between groups by using the Student t test were performed using a standard computer program (STATA, Computing Resource Center, Santa Monica, CA USA). Linear regression plots and correlation coefficients were generated using SigmaPlot (Jandel Scientific, San Rafael, CA USA).

#### **Results and discussion**

The data from the taurine-supplemented cats and kittens are presented separately from the data from the taurine-deficient cats and kittens for comparison. Our results show a wide range of whole blood taurine concentrations in taurine-supplemented cats and kittens (*Figure 1A*) accompanied by an even wider range of values for plasma taurine concentration (*Figure 1C*). Trautwein and Hayes<sup>10</sup> reported that the range of whole

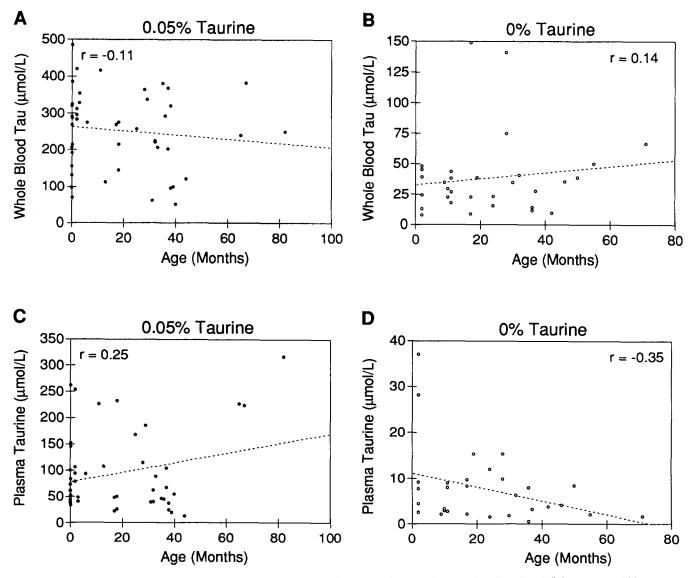


Figure 1 Whole blood (A,B) and plasma (C,D) taurine concentrations in taurine-supplemented and taurine-deficient cats and kittens as a function of age.

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blood taurine concentrations in adult cats, males and females, is 300-400 µmol/L, and that it declined only after prolonged dietary taurine deprivation, whereas plasma taurine concentrations fluctuated widely depending on dietary availability. The mean whole blood taurine concentration of the taurine-supplemented cats and kittens in this study (Table 1) is somewhat lower than reported by Trautwein and Hayes,10 but the dietary histories of the cats are different; those in this study had only been fed the purified diet, whereas those in the other study had spent a substantial time consuming a commercial nonpurified diet that contained 0.12% taurine. There is no significant correlation between whole blood or plasma taurine concentrations and age (which corresponds to the time spent consuming the diet) (Figure 1A, C).

The whole blood taurine concentrations in the taurine-deficient cats and kittens (Figure 1B) are much lower than observed in the taurine-supplemented cats and kittens (15%), as are the plasma taurine concentrations (8%) (Figure 1D). Whole blood concentrations do not correlate significantly with age, i.e., time consuming a taurine-free diet, but plasma taurine concentrations tend to decrease with age. A previous study<sup>10</sup> reported that after 100 days consuming a taurine-free diet, female cats have a whole blood taurine concentration of 102  $\pm$  58  $\mu$ mol/L compared to 32  $\pm$  12  $\mu$ mol/L for adult male cats. Why adult female cats resist taurine depletion more than adult male cats is not known, but presumably the same lower values would be reached after a more prolonged dietary deprivation of taurine. This study does not address rate of decrease of taurine concentrations because the cats always consume the same diet, either taurine-supplemented or taurine-free, and would be expected to be in equilibrium with the amount of taurine biosynthesized, plus the dietary taurine in the taurine-supplemented group, and the taurine lost via excretion or bacterial degradation in the gut.

An alternative way to examine the results is to compare whole blood taurine concentration to plasma taurine concentration. As might be expected, in both groups higher whole blood values are generally accompanied by higher plasma values, although the correlations are weak (*Figure 2 A, B*). A systematic study of whole blood and plasma taurine concentrations in human subjects also concluded that these parameters are not correlated.<sup>7.8</sup> The ratio of whole blood to plasma taurine in taurine-supplemented cats and kittens showed no variation with age (*Figure 2C*), although taurinedeficient cats and kittens showed an increase with age (Figure 2D). The explanation for this observation is not clear but may be related to the continued growth of the cats accompanied by no increase in taurine synthesizing capability, which would show up in reduced plasma concentrations more quickly than in whole blood or tissue concentrations. There is also some evidence that blood components undergo alterations in proportions during prolonged taurine deprivation,<sup>11</sup> although no obvious changes in whole blood concentration with age were noted (Figure 1B). Whole blood taurine stores take much longer to deplete than plasma in the absence of dietary taurine and appear to be more truly representative of whole body taurine stores, which for slowly turning over pools in tissues such as muscle and brain take many months to reach equilibrium.

We conclude from these observations that assessment of taurine status should preferably include measurement of whole-blood and plasma taurine concentrations, as suggested by Trautwein and Hayes.<sup>7,8</sup> Certainly the use of whole blood avoids any possibility of error that can occur in plasma measurements if any hemolysis occurs or if there is any contamination with platelets and appears to give the best assessment of whole body taurine status. On the other hand, it is clear from the taurine depletion studies of Trautwein and Hayes<sup>10</sup> that the effect shows up more rapidly in plasma than whole blood and therefore plasma taurine levels will give the best indication of a recent dietary taurine insufficiency.

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Table 1 Concentrations of taurine in whole blood and plasma and whole blood/plasma ratios from taurine-supplemented and taurine-deficient cats and kittens

	Taurine-supplemented(47)	Taurine-deficient(31)	Р
Whole blood (µM/L)	252.3±105.1	38.5±32.5	< 0.00001
Plasma (µM/L)	95.6±76.1	$7.7 \pm 7.8$	< 0.00001
Whole blood/plasma	$3.89 \pm 2.59$	$8.98 \pm 8.68$	0.004

Each value represents the mean  $\pm$  S.D. from the number of cats and kittens in parentheses.

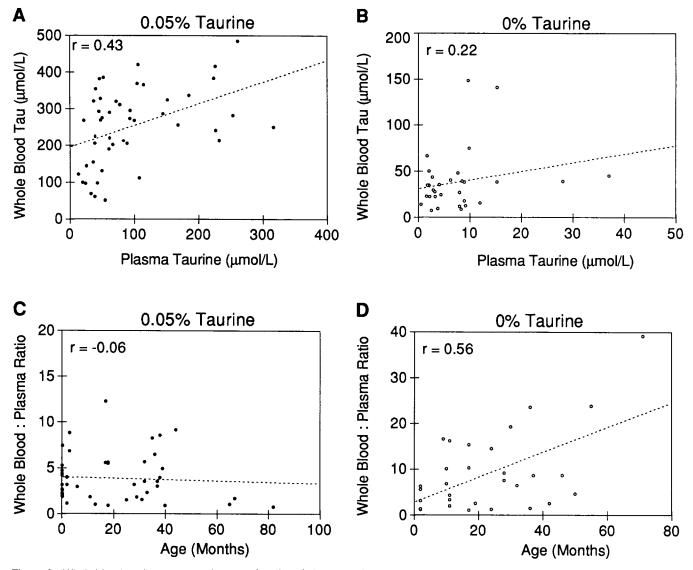


Figure 2 Whole blood taurine concentrations as a function of plasma taurine concentrations in taurine-supplemented (A) and taurine-deficient (B) cats and kittens. Whole blood/plasma taurine ratio as a function of age in taurine-supplemented (C) and taurine-deficient (D) cats and kittens.

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