STANDARD ARTICLE

Journal of Veterinary Internal Medicine

ACVIM
American College of Veterinary Internal Medicine

Open Access

Water requirements of canine athletes during multi-day exercise

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Background: Exercise increases water requirements, but there is little information regarding water loss in dogs performing multi-day exercise

Objectives: Quantify the daily water turnover of working dogs during multi-day exercise and establish the suitability of SC administration of tracer to determine water turnover.

Animals: Fifteen privately owned Labrador retrievers trained for explosive detection duties and 16 privately owned Alaskan Huskies conditioned for mid-distance racing.

Methods: All dogs received 0.3 g D_2O/kg body weight by IV infusion, gavage, or SC injection before the start of a multi-day exercise challenge. Explosive detection dogs conducted 5 days of simulated off-leash explosive detection activity. Alaskan sled dogs completed a mid-distance stage race totaling 222 km in 2 days. Total body water (TBW) and daily water turnover were calculated using both indicator dilution and elimination regression techniques.

Results: Total body water (% of body weight) varied from 60% ± 8.6% in minimally conditioned Labrador retrievers to 74% ± 4.5% in highly conditioned Labrador retrievers. Daily water turnover was as high as 45% of TBW during exercise in cold conditions. There was no effect of sex or speed on daily water turnover. There was good agreement between results calculated using the indicator dilution approach and those calculated using a semilog linear regression approach when indicator isotope was administered IV or SC.

Conclusions and Clinical Importance: Water requirements are influenced primarily by the amount of work done. SC administration of isotope-labeled water offers a simple and accurate alternative method for metabolic studies.

KEYWORDS
hydration, sled dog, thermoregulation, water turnover, working dogs

1 | INTRODUCTION

Thermal homeostasis requires the dissipation of metabolic heat through various processes, including conduction, convection, radiation, and evaporation.[1,2] Of these processes, evaporation is unique in that it consumes a body resource (water) and thus places an additional homeostatic requirement in the form of hydration on the athelete.[3]

Because thermal homeostasis is achieved by evaporation of water from the skin and respiratory tract, water intake requirements increase. It follows that conditions that increase daily energy expenditure and metabolic heat production, such as exercise, not only increase dietary energy requirements but also water intake requirements to dissipate the increased heat accumulation.[4] The exact relationship between a change in energy expenditure and water intake requirements is difficult to predict because of the variable dependence on evaporation for thermal homeostasis.[1,5] Given the concurrent reliance on hydration for cardiovascular performance in support of exercise, failure to meet water

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requirements by water intake can lead to dehydration, poor performance, and hyperthermia.[6]

Exercising dogs can expend 6,000-12,000 kcal of energy per day during prolonged submaximal exercise,[7,8] potentially increasing daily water requirements depending on how much of this heat is dissipated by evaporation. Water consumption has been studied in resting dogs,[9] hunting dogs in cold climates,[10] working sled dogs,[11] and ultramarathon distance sled dogs,[8] with all exercise studies describing increased water requirements of dogs exercising under cold conditions (−6 to −20°C). No published studies describe the water requirements of dogs exercising in more temperate or hot conditions. Furthermore, although published studies cover a range of daily exercise amounts and energy expenditures (19.4 km/day and 1,170 kJ/kg^{-75}/day for hunting dogs,[10] 240 km/day and 4,400 kJ/kg^{-75}/day for ultramarathon sled dogs[8]), gaps remain in published data about the water requirements of dogs travelling 40–80 km/day and generating proportionately less metabolic heat. Knowledge of the water expenditure of dogs under these conditions, which are typical of a range of activities from explosive detection work to mid-distance racing, is vital to ensure dogs are hydrated appropriately for athletic performance. Therefore, the goal of our study was to quantify water requirements for dogs performing moderate amounts of endurance exercise and determine whether these requirements are affected by conditioning, sex, or exercise intensity.

The double-labeled water technique for estimation of energy and water demands has been used since the 1950s and has been validated and utilized in many studies in dogs and other species.[12–14] This technique historically has required IV catheter placement for administration of the isotope tracers. This approach is cumbersome, expensive, and requires additional facilities and trained operators for studies in the field. In addition, private animal owners might be averse to participating in studies in which increased risk of infection or complication is associated with placement of IV catheters. The option to administer isotopes by alternative routes could allow other research avenues in the future. Therefore, a second goal of our project was to evaluate the bias and precision of total body water (TBW) and daily water turnover results determined using alternative methods of isotope administration including IV, PO gavage, and SC routes.

2 | MATERIALS AND METHODS

All studies were approved by the Institutional Animal Care and Use Committee of Oklahoma State University and studies were conducted in accordance with the principles outlined in the NIH Guide for the Care and Use of Laboratory Animals. In all studies, TBW and water turnover were calculated by measuring blood concentrations of deuterium oxide (D_2O). In general, dogs received 0.3 g D_2O/kg body weight, with the actual delivered dose calculated as the difference in mass between the filled syringe and needle and the postinjection syringe and needle as measured on a scale accurate to 10 mg. Except where indicated in the description of the specific studies, venous blood samples (6 mL) were obtained from each individual dog immediately before isotope dosing, 4 hours after administration of D_2O, and at various time

points during the exercise challenge. The dogs were allowed to drink water ad libitum throughout the study (except for the 4 hours period immediately after isotope administration) to help ensure normal hydration at the time of blood sample collection.

Study #1 was conducted using 6 unconditioned explosive detection dogs (3 intact males, 3 intact females; 26 ± 2 months old; 30.7 ± 4.5 kg body mass, body condition score [BCS], 5–6/9) from a single kennel while conducting a 5-day simulated deployment exercise on the kennel premises as previously described.[15] Dogs received 0.3 g D_2O/kg body weight (30% atomic excess) delivered by gavage during gastric endoscopy. Serum enrichment with D_2O was measured in blood samples obtained immediately before isotope administration, 4 hours after isotope administration, and each morning before deployment exercise for 5 consecutive days, including the morning after the last exercise day.

Study #2 was conducted using 9 explosive detection dogs (2 unconditioned and 7 highly conditioned) from a single kennel while conducting a 5-day simulated deployment exercise on the kennel premises as previously described.[7] Unconditioned dogs (1 intact male, 1 intact female; 46 ± 3 months old) were 34.5 ± 3.3 kg body mass (BCS, 7/9) and highly conditioned dogs (3 intact males, 4 intact females; 44 ± 2 months old) were 28.5 ± 3.4 kg body mass (BCS, 4/9). Dogs received 0.3 g D_2O/kg body weight (99% atomic excess prepared as pharmaceutical-grade ultraviolet light [UV]-sterilized 0.9% NaCl solution filtered through a 0.2 μm filter) IV. Serum enrichment with D_2O was measured in blood samples obtained immediately before isotope administration, 4 hours after isotope administration, and each morning before deployment exercise for 5 consecutive days, as well as the morning after the last exercise day.

Study #3 was conducted using 16 mature Alaskan Husky sled dogs (10 intact males, 3 intact females; 4.5 ± 1.8 years old; 24.6 ± 1.8 kg body mass; BCS, 3–4.5/9) comprising 2 racing teams of 8 competing in the CopperDog 150 sled dog race, a 3 × approximately 46 mile (74 km) stage race held during the last weekend of February in the upper peninsula of Michigan, starting in Calumet, Michigan. Each stage is separated by approximately 9 hours. Each team was owned and cared for by separate mushers. Team A consisted of 8 intact males whereas Team B consisted of 3 intact females and 5 intact males. Both sets of dogs lived year-round within 50 miles of the race start. Dogs underwent physical examinations, including body weight, 8 hours before the start of the race and received 0.3 g D_2O/kg body weight (99% atomic excess prepared as pharmaceutical-grade UV-sterilized 0.9% NaCl solution filtered through a 0.2 μm filter) SC injected between the shoulder blades (approximately 7.5 mL for the average 25 kg sled dog). To decrease cost, blood samples to determine baseline (predosing) concentrations of the isotope only were collected from 4 randomly selected dogs from each team before injection of the isotope to correct for background abundance of D_2O in the dogs. Because all dogs from a single team had a common drinking water source before the study, the average background enrichment of D_2O in the selected blood samples was presumed to represent the background enrichment for the entire kennel. Water samples were collected from tap sources at each checkpoint to measure local enrichment of D_2O and rule out

the possibility of skewing data if abnormally high amounts of D_2O were consumed along the race.

Samples were analyzed by a commercial laboratory (Metabolic Solutions, Inc; Nashua, New Hampshire) to determine the enrichment of the samples with the isotope. Total body water (expressed as both total mass and percentage of body mass) was calculated using 2 different formulas: the indicator dilution approach, in which the difference in enrichment between the plasma samples obtained 4 hours after D_2O injection and the preinjection enrichment is used to calculate the enrichment pool using standard formulas (Technical Bulletin: Deuterium Analysis and TBW Calculations, Metabolic Solutions, Inc, Nashua, New Hampshire) and the regression intercept method in which the time 0 enrichment and thus the enrichment pool is extrapolated from linear regression of the plasma enrichment results (corrected for background enrichment) over the course of the study (Technical Bulletin: Calculation of Energy Expenditure, Metabolic Solutions, Inc, Nashua, New Hampshire). Water turnover rate was calculated using both results for TBW as the starting pool size and expressed as both an absolute rate as well as a percentage of TBW. Within each study, agreement between the 2 approaches for calculating TBW was expressed using Bland-Altman analysis (GraphPad Prism 6.01, Graphpad Software, Inc, La Jolla, California). Bias was calculated as the average of the differences and the standard deviation of those differences between paired measurements. Unpaired Student's t-tests were used in Study #2 to assess the effects of conditioning on TBW and water turnover, and in Study #3 to determine the effect of team and sex on TBW and water turnover, with $P < .05$ considered statistically significant. All data were expressed as mean ± standard deviation unless otherwise noted.

3 | RESULTS

3.1 | Study #1

Dogs were examined at each sample time point to assess relative hydration and were normally hydrated throughout the study. Total body water was not calculated for 2 dogs because of observed losses of isotope-labeled water during dosing. For the remaining 4 dogs, TBW before the start of the first deployment simulation was 18.79 ± 1.60 kg (63.0% ± 8.4% body weight) when calculated using the indicator dilution method and 17.98 ± 1.32 kg (60.3% ± 8.6% body weight) when calculated using the regression method. Dogs averaged a total of 161 km over 5 days of exercise in temperatures between 21°C and 29°C,[15] with average daily water turnover of 4.62 ± 0.42 L/day (25.6% ± 1.3% TBW/day). Bland-Altman analysis demonstrated bias (0.8125 ± 0.3835) towards higher results for TBW when calculated using the indicator dilution method compared with the regression method.

3.2 | Study #2

Dogs were examined at each sample time point to assess relative hydration and were normally hydrated throughout the study. Total body water before the start of the second deployment simulation was 21.08 ± 2.63 kg (74.00% ± 4.48% body weight) in highly conditioned dogs and 21.92 ± 3.87 kg (63.30% ± 5.26% body weight) in unconditioned dogs when calculated using the indicator dilution method, and 20.94 ± 3.17 kg (73.53% ± 7.90% body weight) in highly conditioned dogs and 20.86 ± 2.92 kg (60.33% ± 2.77% body weight) in unconditioned dogs when calculated using the regression method. Unconditioned dogs had significantly lower TBW as a % of bodyweight using both methods (indicator dilution, $P = .01$; regression, $P = .03$). Highly conditioned dogs averaged 178.8 ± 4.7 km over 5 days of exercise in temperatures between 21°C and 29°C,[7] with average daily water turnover of 4.88 ± 0.95 L/day (23.3% ± 4.4% TBW/day), whereas unconditioned dogs covered significantly less distance (155.8 ± 2.4 km) during the same period, but had a similar rate of water turnover (4.93 ± 0.33 L/day; 22.7% ± 2.5% TBW/day; $P = .44$). Bland-Altman analysis showed low bias and good agreement between the 2 methods for calculating TBW. Bias for TBW (kg) was 0.3433 ± 1.107 and for TBW (% of body weight) was 1.023 ± 3.703.

3.3 | Study #3

Both teams finished the race, placing 12th and 14th out of 20 teams. Eleven dogs raced all 3 stages of the race, 2 dogs did not race all stages, and 3 dogs were dropped from the study because of injection error. Dogs were examined at each sample time point to assess relative hydration and were normally hydrated throughout the study. Background enrichment of the dogs averaged −87 delta/mil, and SC dosing of D_2O resulted in an average enrichment of >3200 delta/mil. Tap water collected at each checkpoint ranged from −95 to −99 delta/mil. At the completion of the study period, average enrichment remained >1000 delta/mil in all dogs. Total body water before the start of the race was 14.80 ± 1.42 kg (65.6% ± 4.27% body weight) when calculated using the indicator dilution method and 14.77 ± 1.36 kg (65.48% ± 4.33% body weight) when calculated using the regression method. A significant effect of team on TBW was found when expressed as a % of body weight (Table 1). Alaskan Huskies in a stage race situation required 6.56 ± 0.83 L (44.43% ± 3.54% TBW per day). No significant difference was found between males and females in water turnover ($P = .64$). The difference in water turnover per day was not significant between the 2 teams ($P = .77$). Bland-Altman analysis showed low bias and excellent agreement between the 2 methods for calculating TBW. Bias for TBW (kg) was −0.01133 ± 0.4168, and for TBW (% of body weight) was −0.08667 ± 1.846.

4 | DISCUSSION

The accuracy of any particular technique or administration protocol for the measurement of TBW depends on the timing of the postindicator sample, with the optimal timing striking a balance between the time required for complete equilibration of the isotopic indicator whereas minimizing the loss of indicator through normal physiological processes. Furthermore, the type of error (overestimation or underestimation) of TBW can depend on the specific protocol. When sampling from the central compartment (blood), a sample that is obtained too long after isotope administration will result in overestimation of TBW because of

TABLE 1 TBW and water turnover during multi-day exercise in working dogs

	TBW (%BM)	rH$_2$O (l/day)	Water turnover (%TBW/day)
Study #1: Unconditioned Retrievers (n = 6)	63.0 ± 8.4	4.6 ± 0.4	25.6 ± 1.3
Study #2: Unconditioned Retrievers (n = 2)	60.3 ± 2.8	4.9 ± 0.3	22.7 ± 2.5
Conditioned Retrievers (n = 7)	74.0 ± 4.5	4.9 ± 0.9	23.3 ± 4.4
Study #3: Sled dogs (n = 13)	65.6 ± 4.3	6.5 ± 0.8	44.2 ± 3.9
Male (n = 10)	65.8 ± 4.3	6.6 ± 0.7	44.6 ± 3.9
Female (n = 3)	65.8 ± 2.8	6.8 ± 1.3	45.8 ± 2.6
Team 1 (n = 7)	64.2 ± 4.4	6.3 ± 0.7	44.5 ± 3.9
Team 2 (n = 6)	67.1 ± 3.9[a]	6.7 ± 0.7	43.9 ± 3.1

Abbreviations: TBW, total body water expressed as % of body mass; rH$_2$O: daily water turnover in liters/day.
[a]Significantly different from team 1, $P = .02$.
Study 1: isotope delivered by intragastric gavage. Study 2: isotope delivered IV. Study 3: isotope delivered SC. Studies 1 and 2 were performed in North Carolina in warm weather. Study 3 was performed in Michigan in winter.

physiological loss of isotope. Some loss of isotope through physiological processes is unavoidable because these processes occur continuously, but can be decreased by minimizing those losses (ie, keeping the subject in a cool environment and minimizing activity to decrease isotope loss through respiratory evaporation) and preventing the subject from replacing lost water between the time of isotope administration and postisotope sampling. The effect of sampling too quickly is dependent upon the route of isotope administration. When isotope is administered directly into the central compartment, sampling too quickly will result in underestimation of TBW because of incomplete equilibration with secondary water spaces. When isotope is administered outside of the central compartment, such as via the gastrointestinal tract or by SC injection, the effect of sampling before complete equilibration is somewhat unpredictable because of the offsetting rates of equilibration between the site of administration and the central compartment, and between the central compartment and the secondary water spaces. Two studies[12,16] showed that from 2 to 4 hours after IV administration of isotope, serum enrichment was relatively constant and suggested that, during this period, equilibration of the isotope within the body water could be considered complete. However, data from another study[9] showed a distinct negative slope to plasma enrichment of D$_2$O between 1 and 3 hours postisotope administration by IV infusion in some, but not all, dogs. Based on these studies and with consideration of the fact that complete equilibration of isotope administered by routes other than IV infusion would require additional time for the administered isotope to diffuse into the central compartment, we chose to obtain samples 4 hour post-administration.

An additional technique for calculating TBW is to obtain multiple postisotope samples at known time points, and construct a regression curve of time (x-axis) versus enrichment (expressed as the natural logarithm) on the y-axis.[8,12] With this technique, calculation of the enrichment at the y-intercept (time 0) can provide a basis for the prediction of TBW. The advantage of this approach is that it eliminates the need to minimize physiological water losses during the measurement process. Instead, it only requires that the appropriate rates of physiological

water loss remain relatively constant between sample points. In addition, use of this approach provides additional data on fractional water loss over time as the slope of the regression line. The disadvantage of this technique is that it requires multiple samples, thus increasing the cost and duration of the procedure. In addition, it is dependent upon the subjects being in a comparable hydration state at each sampling point, which from a practical standpoint means that the subjects must be normally hydrated. In these studies, we met this latter requirement by scheduling the sampling times for when the dogs had been provided ad libitum access to water for at least 2 hours in the case of the racing sled dogs and overnight in the case of the explosive detection dogs.

Total body water was measured using both approaches, allowing us to compare the results of the different routes of indicator administration for precision and bias as well as the 2 calculation methods. In studies of human subjects, in which isotope is rarely administered by IV infusion and enrichment of body water is often performed by analysis of urine samples, it is generally concluded that both techniques provide comparable results. However, a previous study[12] found that in sedentary dogs that received isotope by IV infusion with body water enrichment determined in serum samples, the indicator dilution method produced higher values than the regression method and concluded that the indicator dilution method was the more accurate of the 2 techniques. In Study #1 (gastrointestinal administration of isotope), there was a bias towards higher results for TBW using the indicator dilution method compared to the regression method, suggesting that either the initial postisotope sampling time was too long (resulting in excessive loss of isotope through physiological processes) or that the sampling time was too short (resulting in insufficient equilibration of isotope between the gastrointestinal tract and the central compartment). In contrast, the bias for Study #2 (IV administration of isotope) was low and for Study #3 (SC administration of isotope) was quite low, with the range of a single standard deviation including zero (no bias). The agreement between the indicator dilution and regression calculations provides some reassurance for the reliability of these approaches to isotope administration, with the very low bias and comparative ease of

administration of the SC injection perhaps being the preferred approach for studies such as these.

In Study #3, to account for the possibility of erroneously altered background enrichment affecting results, samples were examined to rule out both the normal kennel environment of the dogs and water sources along the race. Isotope enrichment was measured from pooled blood samples from dogs before isotope administration and samples from water sources taken along the race route. The difference between the tap water enrichment at the checkpoints versus the kennels (represented by pre-race blood enrichment) was miniscule compared to the postdosing enrichment and should not affect the calculation of water turnover. The average pre-race enrichment was −87 delta/mL and the tap water enrichment was −99 delta/mL, which is more than 1,000 times lower than the postenrichment samples.

Total body water results for domestic dogs, as measured using D_2O dilution, have been published in numerous studies, with mean values ranging from 56% of body mass in healthy sedentary 2-year-old Beagles[9] to 72% of body mass in healthy sedentary random-source adult dogs.[16] A study of racing sled dogs found an average TBW of 66% of body mass.[8] The values measured in our various studies fall within this range, with TBW in retrievers ranging from 60% to 73% of body weight and in sled dogs virtually the same as previously published.[8] One important factor that can result in shifts in TBW as a percentage of body mass is the amount of body fat in the subject. Because body fat contains very little water, dogs with higher BCS will tend to have lower TBW as a percentage of body mass. We believe this is the primary explanation for the differences in TBW% in the retrievers of Studies #1 and #2. All dogs in Study 1 and the unconditioned dogs in Study 2 participated in a minimal exercise program and had higher BCS compared with the conditioned dogs in Study #2 that had undergone an intensive exercise conditioning program. As a result, the conditioned dogs of Study #2 likely had less body fat, resulting in higher TBW%.

The data from Study #3 are consistent with the premise that water usage is influenced primarily by the amount of work done (distance covered) and thus the amount of heat generated, and not by speed or sex. The similarity of water turnover between teams indicates that those physical principles are likely present in all dogs engaged in this activity. There was no team effect on water turnover, further indicating that metabolic requirements for water are not impacted by team-based variables such as diet, racing speed, athletic conditioning, and breeding. Rather, the most important factor in meeting the metabolic needs of canine athletes appears to be the amount of metabolic heat that needs to be dissipated to remain within physiologic temperatures. Within the speeds and racing distances observed in this study, water turnover appears to be more of a function of distance than speed. Meeting hydration requirements by replacing water lost is important for athletic performance and normal cardiovascular function.

The impact of environmental conditions on water requirements during exercise is complex. High environmental temperatures (such as Studies #1 and #2) would be expected to decrease heat dissipation by conduction and thus indirectly increase reliance on evaporation and water loss for dissipation of metabolic heat. However, cold environmental temperatures (such as in Study #3) will increase obligatory water loss through the respiratory tract because of the passive humidification of inspired air with very low absolute water content. The data in our study provides some examples of the relative impact of environmental conditions on routes of heat dissipation and, by extension, the impact of metabolic heat dissipation on daily water requirements in working dogs. For the sake of illustration, we will assume that 2 L of daily water turnover was lost through a nonevaporative route (ie, urination and fecal water loss),[4] and that the balance of the daily water turnover was lost to evaporation, resulting in dissipation of metabolic heat. In Study #1, dogs expended approximately 3,446 kcal/day of energy,[7] and assuming that 2.6 L of water were lost each day by evaporation, these dogs dissipated 1,404 kcal of metabolic heat by evaporative water loss, or approximately 41% of their daily production of metabolic heat. In Study #2, the unconditioned and conditioned dogs produced 5,271 and 6,298 kcal of metabolic heat each day, respectively.[7] Both groups of dogs had similar rates of daily water turnover, and based on the assumption of 2.9 L of water lost by evaporation (1,566 kcal of metabolic heat dissipated), unconditioned and conditioned dogs dissipated approximately 30% and 25% of their daily metabolic heat production through water evaporation. In Study #3, dogs are estimated to have produced 6,875 kcal/day (based on estimates derived from a previous study[8] of approximately 2,000 kcal/day for baseline maintenance plus 65 kcal/mile of exercise and 75 miles/day). With an approximate daily evaporative water loss of 4.5 L, racing sled dogs shed 2,430 kcal or 35% of their daily metabolic heat production by water evaporation. The dogs in the previous study[8] performed twice the work of the sled dogs in Study #3 but in much colder conditions. Thus, the turnover of approximately the same amount of water per day represents a much smaller fraction (18%) of daily metabolic heat production dissipated by evaporation. These estimates support the expected pattern that relative reliance on evaporation (and resulting increased rate of body water turnover) increases with increasing environmental temperature, with the data from the stage racing sled dogs (Study #3) deviating from this expected pattern. Differences in feeding strategy (such as mixing water into kibble as is common in sled dog kennels versus offering free choice water in Studies #1 and 2) may impact water turnover. Sled dog feeding strategies including mixing water into kibble may be overhydrating the dogs and falsely increasing TBW. The reasons for the unexpectedly large daily water requirement in these dogs are unknown and will require further study.

Conditioning-induced increases in TBW can improve exercise capacity in dogs by 2 physiological mechanisms: expanding plasma volume and increasing the availability of water for thermoregulation.[4] Increasing plasma volume, and thus cardiac output, improves exercise capacity and has been reported previously in other athletic dogs,[17] but because dogs in these studies were unlikely to be exercising at or near maximum cardiac output, the increase in TBW may not improve exercise capacity in this manner. Rather, it is possible that in these dogs (and dogs participating in similar activities), increased TBW facilitates exercise by expanding the water available in the body for thermoregulation. The potential importance of this role of body water in exercise performance in dogs, particularly in dogs performing prolonged submaximal exercise, should not be underestimated.

In conclusion, our results show that exercising dogs may have remarkably high water requirements when exercising during hot conditions, but that these high water requirements are not necessarily restricted to warm weather exercise. No statistical difference was found between male and female dogs, although there were only 3 females on which to base this trend. Despite differences in training, feeding programs, fitness levels, breeding, and race strategy, overall water turnover remained statistically similar. We obtained values for TBW in Study #3 that were similar in value to previous studies using IV administration of D_2O,[8,9,16] supporting the SC administration of isotope-labeled water in dogs in a field environment. Further work characterizing water turnover and caloric requirements in the various situations under which working dogs perform and race using this newly established SC administration method may provide more information for handlers, mushers, and veterinarians to best care for these canine athletes. Our data indicate that maintenance of hydration may require replacement of > 40% of a dog's TBW each day during periods of high rates of metabolic heat generation, and underscores the potential for rapid dehydration if these losses are not met during athletic activities. Finally, we have outlined the utility of an alternative method for administration of stable isotopes used for measurement of TBW that was easily performed, well accepted by the dogs and their owners, and resulted in useful data on the water requirements of dogs in a nonlaboratory environment.

ACKNOWLEDGMENTS

CONFLICT OF INTEREST DECLARATION

Authors declare no conflict of interest.

OFF-LABEL ANTIMICROBIAL DECLARATION

Authors declare no off-label use of antimicrobials.

INSTITUTIONAL ANIMAL CARE AND USE COMMITTEE (IACUC) OR OTHER APPROVAL DECLARATION

All procedures were reviewed and approved by the Oklahoma State University IACUC.

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