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Loss of body weight and lean mass in long-stay, hospitalized canine patients

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Abstract

A high prevalence of malnutrition occurs in human hospitals and has been associated with detrimental consequences. By comparison, much less is known in hospitalized veterinary patients. The aims of this study were to evaluate the prevalence of malnutrition and body composition changes in long-stay hospitalised patients using an isotopic dilution technique. An additional objective was to compare the changes in composition with commonly used methods measuring body fat and lean mass. The dogs consumed on average 77.5% of their estimated resting energy requirements during their stay. The majority (78.3%) of dogs lost body weight, of which a greater proportion was lean mass (61.8%) than fat mass (FM) (38.2%). There was a moderate correlation between body condition score and percentage FM measured at admission (Kendall's $\tau = 0.51$; $p = 0.002$), and at discharge (Kendall's $\tau = 0.55$; $p = 0.001$). However, there was no correlation between muscle condition score and fat-free mass at either admission or discharge ($p > 0.1$). Duration of stay was positively associated with loss of body weight ($p < 0.001$), but was not associated with changes in either lean or FM expressed as a percentage of body weight or in absolute terms ($p > 0.1$), which was presumed to be explained by small sample size and variation. Food intake was not found to a significant factor for lean or FM loss ($p > 0.1$). These findings indicate that weight loss is common in hospitalized canine patients, which is not explained by simple under-eating. Other factors such as inflammation and inactivity should be evaluated in future studies to determine their role in influencing muscle and FM changes in hospitalized canine patients.

KEYWORDS

deuterium, dog, fat, hospital, lean, muscle

1 | INTRODUCTION

The loss of lean and fat mass (FM) during illness have different aetiologies and consequences for hospitalised patients. The loss of muscle can occur due to insufficient caloric/protein intake, metabolic derangements, systemic inflammation and disuse atrophy, whereas the loss of FM occurs primarily due to inadequate caloric intake, though losses can also be exaggerated in cachexic patients (Amitani et al., 2013; Bonaldo & Sandri, 2013; Evans et al., 2008; Lenk et al., 2010). In humans, having a low muscle mass or losing muscle while in the hospital is associated with a poorer prognosis (Anker et al., 1997; Caporossi et al., 2012; Chan, 2013; Futter et al., 2011; Slinde et al., 2005; Thomas, 2007; Vestbo et al., 2006). Much less is known in hospitalized companion animals, however, in dogs and cats with cancer or cardiac disease, having a low body condition score, a semiquantitative method of assessing body FM, during their treatment period, is associated with significantly shorter survival times (Baez et al., 2007; Finn et al., 2010; Romano et al., 2016). When FM is lost, there is an accompanying drop in the adipose-derived hormone leptin, which plays a vital role in modulating both innate and adaptive immunity (Jeusette et al., 2005; La Cava & Matarese, 2004; Mayo et al., 2003; Sánchez-Margalet et al., 2010). In addition, compared to lean individuals, being overweight is associated with reduced mortality in human patients with septicaemia and chronic disease (Bornstein et al., 1998; Fonarow et al., 2007; Kuperman et al., 2013; Oreopoulos et al., 2008; Park et al., 2014). Therefore, it appears that preservation of not only lean, but also of FM, may be of importance for hospitalised patients.

It has been documented that the longer a veterinary patient stays in hospital, the greater the risk is of reductions in body weight and body condition scoring (BCS) (Molina et al., 2018). Body weight, muscle condition scoring (MCS), and BCS are the most commonly used parameters for assessing malnutrition in veterinary medicine (Freeman et al., 2018; Laflamme, 1997b; Santarossa et al., 2018). However, the information provided by these frequently used measures are limited. Total body weight change is a crude measure that does not differentiate between lean and FM, or changes in hydration. Conversely, BCS and MCS have been developed to specifically assess FM and lean mass respectively. Nevertheless, the scoring categories are broad and may be insensitive to subtle changes that may occur during hospitalisation. Further, neither have been validated for use in hospitalized patients. Thus, we currently lack a clear understanding as to what changes occur in lean and FM during hospitalisation in veterinary patients, and whether the current scoring systems accurately measure these changes.

The aims of this study were to determine the changes in body composition during 'long-stay' hospitalisation using an isotopic dilution technique, and to compare the results with the current standard methods of measuring body composition in veterinary patients. In addition, we aimed to identify risk factors for body composition changes. We hypothesised that current methods of measuring body composition would not be accurate in measuring changes in lean and FM in long-stay hospitalised patients.

2 | MATERIALS AND METHODS

2.1 | Historical clinical records collection

In order to define 'long-stay' for the prospective study, clinical data were extracted from the records of dogs seen by the internal medicine or surgical department at Massey University Veterinary Teaching Hospital (MUVTH). Records included were for hospital admissions between November 2013 and March 2014, and data were extracted from the practice's patient management software[1] retrospectively. Exclusion criteria included patients who stayed < 24 h, patients whose records had either admission and/or discharge body weights missing, and those patients who received intravenous fluid (IVF) therapy, in order to avoid the effect of hydration on body weight. Patients who received IVF only during an anaesthesia period were still included. Each patient's age, final diagnosis, length of hospital stay and body weight changes between admission and discharge were examined.

2.2 | Assessment of body composition changes in hospitalised dogs

For the assessment of body composition, dogs were prospectively recruited from MUVTH's internal medicine and surgical departments between August 2015 and September 2017. The recruitment criteria included patients with any disease process where the expected hospital stay was equal to or greater than the defined length of time for a 'long-stay' patient as determined by the retrospective study. The suitability for enrolment of a patient was determined based on the clinical signs at admission and the primary clinician's recommendation as to the likelihood of the duration of stay for the patient. Once the primary clinician's approval was obtained, each patient's owner(s) was contacted for permission to include their dog in the study. Exclusion criteria included dogs with critical illness (American Society of Anaesthesiologists [ASA] status grade four or five), those with disturbances in water balance (e.g., kidney failure, chylothorax, haemoabdomen), or those who received IVF therapy during their hospitalisation at times other than during anaesthesia (ASo, 2014); any of which could have affected the distribution of the stable isotope used for body composition measurements. An a priori sample size calculation revealed that 34 dogs would have a power of 0.9 to detect a difference of 15% in fat-free mass (FFM), with a variance of 30%.

Body composition for each patient was measured as close as possible to the time of admission, and repeated immediately before the patient was discharged from the hospital. Body composition was measured using isotopic dilution, as has been previously described (Backus et al., 2000; Cave et al., 2007; Speakman, 2001). Enrolled dogs were fasted overnight but allowed *ad libitum* water up until the procedure began. The dogs received a physical examination in the morning and were not found to be overtly dehydrated. The dogs were then taken outside for urination and defecation, and afterwards their body weights were measured on a large floor scale located in the hospital. Based on their body weight, an injectate solution was made for each dog using a newly opened and weighed syringe and needle. To make the solution, 0.4 mg/kg body weight of deuterium solution[2] was aspirated and combined with 0.13 mg/kg body weight

[1] Cornerstone Veterinary Software 8.3 NEXT (version 8.3.6), IDEXX Laboratories.
[2] Deuterium Oxide (99.8% purity), Cambridge Isotope Laboratories, Inc.

of 3% saline.[3] The syringe and needle were reweighed at each step to determine the exact amount of each compound in the syringe. All syringe weights were measured using the same electronic scales[4] to a milligram accuracy.

A catheter was placed in a peripheral vein of each dog, and a 3 mL blood sample taken to determine the background deuterium concentration. Then, the deuterium mixture was slowly injected through the catheter, the syringe removed, and the catheter flushed with 0.9% saline[5] to ensure complete administration. The syringe and needle were then reweighed to obtain a postinjection weight to allow for residual deuterium to be accounted for. Deuterium was allowed to evenly distribute throughout the body for two hours, after which a second 3 mL blood sample was taken from a different vein to the one used for injection. Water and food were withheld during the 2 h of equilibration. Blood samples were collected into EDTA blood vacutainer tubes.[6] Plasma was obtained from the vacutainers after undergoing centrifugation for 10 minutes at 3000 rcf at 4°C, and the plasma stored in vials[7] with crimp caps[8] at −80°C until analysis.

2.3 | Deuterium analysis

Deuterium was measured in the plasma samples by gas-isotope ratio mass spectrometry.[9] For reference solutions, Vienna Standard Mean Ocean Water, International Atomic Energy Agency enriched water and Aberdeen tap water were used. Aberdeen tap water and International Atomic Energy Agency enriched water were also used as controls. Abundance of deuterium was expressed as parts per 1000 difference from the reference Vienna Standard Mean Ocean Water (δ^2H) and was converted to parts per million (ppm) using the equation:

$$\frac{1{,}000{,}000}{\left(1+\frac{1}{\left(\frac{\delta^2H}{1000}+1\right)\times 0.00015576}\right)},$$

where 0.00015576 is the accepted 2H/1H ratio of Vienna Standard Mean Ocean Water.

2.4 | Total body water

Total body water was determined using the methods from Speakman (2001). For this, the weight of the syringe postadministration was subtracted from the weight of the empty syringe to determine the weight of the residual injectate. This was then multiplied by the percentage concentration of deuterium solution in the residual injectate which was determined for each dog as:

$$\%\text{ concentration of deuterium in injectate} = \left(\frac{\text{deuterium solution (g)}}{\text{deuterium solution (g)} + \text{saline solution (g)}}\right)$$

Then, the weight of the deuterium solution administered was calculated by subtracting the residual weight from the initial weight. This was then multiplied by 0.998 to correct for the purity of the deuterium solution.

Once the exact amount of deuterium solution administered was calculated, the volume of distribution (Vd) was determined as the total deuterium solution administered in grams multiplied by 1000 and then divided by the difference between the deuterium abundance (ppm) in enriched plasma and in baseline plasma. Total body water in kilograms was calculated as Vd divided by 1.04 to account for isotopic exchange of deuterium with the hydrogen present in proteins, carbohydrates and fats (Speakman, 2001).

2.5 | FFM and FM

Lean mass in kilograms, also known as FFM, was calculated as total body water divided by 0.713, which is the hydration constant for lean mass in dogs as determined by carcass desiccation (Burkholder & Thatcher, 1998). Once FFM was calculated, body FM in kilograms was determined by subtracting FFM from body weight.

2.6 | Body and muscle condition, caloric intake, disease state and activity

Body condition score (BCS; 1–9) and muscle condition score (MCS; normal muscle mass [3], mild muscle loss [2], moderate muscle loss [1] and severe muscle loss [0]) were recorded on the days of body composition assessments (Laflamme, 1997b; World Small Animal Veterinary Association: Global Nutrition Committee, 2013). To reduce variation, all scorings were performed by the same researcher (Y. B. L.). If a dog was considered overweight (BCS ≥ 6) or underweight (BCS ≤ 3) at admission, ideal body weight was calculated using the assumption of a 5% change in body fat for every BCS (Laflamme, 1997b). Dogs who were overweight had an ideal body weight estimated to a BCS of 5 out of 9, and underweight dogs had an ideal body weight estimated to a BCS of 4 out of 9. This ideal weight was then used to calculate resting energy requirement (RER) for the patient.

No specific nutritional intervention occurred in this study, and the nutritional care of the dog was prescribed by the primary clinician assigned to the case. However, all dogs were fed diets formulated to meet the nutrient requirements for adult dogs as defined by the American Association Food Control Official (AAFCO, 2015). Body weight and food intake were recorded daily. Daily caloric intake was summed and divided by the number of days of hospitalisation to

[3]3% saline, Baxter Healthcare Pty Ltd, Toongabbie.
[4]Kern PLJ 600-3NM; Kern & Sohn GmbH.
[5]0.9% saline; Baxter Healthcare Pty Ltd.
[6]BD Vacutainer®; Becton, Dickson and Company.
[7]Polypropylene vial 250 µL, catalog #5188-0278; Agilent Technologist Inc.
[8]Aluminium cap, PFTE/butyl septa, catalog #8010-0051; Agilent Technologist Inc.
[9]Isotope Ratio Mass Spectrometry Unit, Otago University, Dunedin, New Zealand.

obtain the mean caloric intake for each patient (kcals/day). Mean caloric intake was then expressed as a percentage of the patient's estimated RER, which was calculated using the following formula (Kleiber, 1975):

$$\text{RER (kcal)} = (\text{ideal body weight})^{0.75} \times 70.$$

It was assumed that each dog's energy requirements was similar to their RER, as has been shown for hospitalised dogs on average (O'Toole et al., 2004; Walton et al., 1996). The severity of disease for each patient was determined at admission and scored using an adapted patient severity score (PSS) (Brunetto et al., 2010; Lumb & Jones, 1984; Molina et al., 2018; Remillard et al., 2001). For this scoring system, we used a score of (0) to indicate an animal with no disease, (1) mild systemic disease, (2) severe systemic disease limiting activity but not incapacitation, (3) incapacitating systemic disease that is a constant threat to life and (4) a moribund animal not expected to live 24 h with or without any type of intervention. In addition, daily activity was measured using a tri-axial accelerometer[10] fitted to the collar of each dog. The daily activity was summed and divided by the number of days of hospitalisation to obtain the mean activity (ΔG) per day for each patient.

2.7 | Statistics

All statistical analyses were performed using the R software.[11] From the hospital records, the median and interquartile range (IQR) for hospital duration of the dogs were established. The first quartile (Q1) value was then used to define the minimum amount of days for a dog to be considered a 'long-stay' hospitalised patient in the second study. Linear regression was used to determine the relationship between body weight change and the duration of stay. The normal distribution of the residuals of the final regression was confirmed by visual appearance and with a Shapiro–Wilks normality test.

Changes in BCS, MCS, FM (expressed as total kg and % body weight), FFM (expressed as total kg and % body weight) for each patient were obtained by subtracting the discharge value from the admission value. Changes in body weight were expressed as the percentage change [(body weight at admission − body weight at discharge) / body weight at admission]. Given that BCS and MCS are ordinal categorical data, Kendall rank correlation was used to compare associations between BCS and FM (expressed as % body weight) and MCS and FFM (expressed as % body weight) at the time of admission and at discharge. Prior to running the correlation test, data exploration was performed, and no outliers were detected (Tukey, 1977).

Models for changes in body weight, FFM and FM during hospitalisation were built using multiple regression analysis in a manual, stepwise technique. Age, sex, disease severity, average daily caloric intake, duration of stay and activity (ΔG) were treated as independent variables. Model fit was assessed using Akaike information criterion (AIC) and adjusted R^2. The collinearity of the variables in the final models was checked by variance inflation factor (VIF). The normal distribution of the residuals of the final regression was confirmed by visual appearance and with a Shapiro–Wilks normality test. Results are presented as mean with standard deviation (SD) unless stated otherwise. P values < 0.001 were considered indicative of very strong evidence, $p < 0.01$ of strong evidence, $p < 0.05$ of moderate evidence, $p < 0.1$ of weak evidence and $p \geq 0.1$ of insufficient evidence (Ganesh & Cave, 2018).

3 | RESULTS

3.1 | Historical clinical records

Sixty clinical records of dogs seen at the MUVTH referral internal medicine and surgical departments were included. The mean age of the dogs was 7 (SD 3.5) years. The majority (57%) of these dogs were hospitalised because of spinal disease, including intervertebral disc disease (IVDD) and fibrocartilaginous embolism (FCE) (Table 1).

The median duration of stay was 5 days, with an IQR of 3 – 7.2 days (Figure 1). On that basis, a hospital duration of 3 days or longer was used to define 'long-stay' patients for the subsequent prospective study. This was chosen as it was hypothesized that 3 days would be sufficient in duration to see changes in body composition.

The mean percentage (%) body weight change was −3.97% (SD 4.45). Percentage body weight change was moderately correlated to the duration of stay for these patients ($r^2 = 0.4238$, $p < 0.0001$, Figure 2).

3.2 | Assessment of body composition changes in hospitalised dogs

A total of 23 dogs were recruited, which consisted of seven females and sixteen males and encompassed 16 different breeds including American bulldog (1), Bichon frisé (2), border collie (1), Chihuahua (1), crossbreed (2), dachshund (3), fox terrier (1), greyhound (2), Harrier hound (2), husky (1), Labrador retriever (1), Maltese (1), Pekingese (1), poodle (2), Staffordshire bull terrier (1) and Weimaraner (1). The mean age of the dogs was 7.2 (SD 3.2) years, and the median duration of stay was 5 [(IQR) 4–9] days. Dogs with spinal disease made up a large majority (18/23) of recruited patients (Table 2). One dog seen by the medicine department had multiple comorbidities including hyperadrenocorticism, diabetes and urolithiasis.

Activity, food intake, body weight, BCS, MCS, FM and FFM at admission and discharge for each dog are presented in Table 3. Median activity was 22,904 (IQR 19,445–47,993) ΔG. In one dog, activity could not be recorded as the surgical incision site was located where the collar and activity monitor were required to be placed. The dogs ate on average 77.5% (SD 28) of their estimated RER. Most

[10] Heyrex®, Say Systems.

[11] R version 3.1.0; R Development Core Team (2012); R Foundation for Statistical Computing.

TABLE 1 Final diagnoses of dogs ($n = 60$) in a retrospective study to investigate the range in hospitalisation duration at Massey University's Veterinary Teaching Hospital's referral service between November 2013 and March 2014.

Diagnosis	Count
Congenital vertebral malformation	1
Discospondylitis	2
Fibrocartilaginous embolism	5
Granulomatous meningoencephalitis	2
Head trauma	1
Inflammatory bowel diseases	1
Immune-mediate polyarthritis	1
Intervertebral disc disease	29
Leg fracture	1
Lumbosacral stenosis	3
Meningitis	1
Myasthenia gravis	1
Neoplasm (all types)	6
Spinal fracture	1
Trauma	3
Not definitive	2

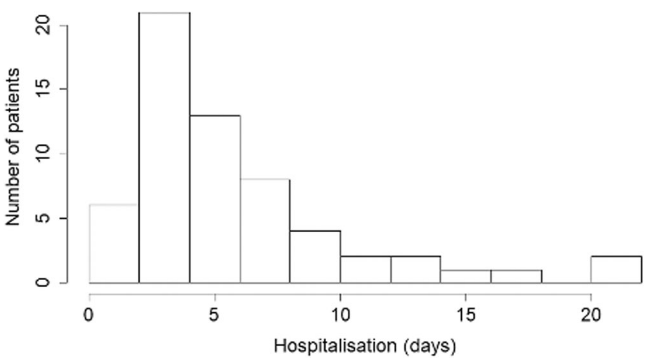

FIGURE 1 Bar graph of the length of stay for 60 dogs in a retrospective study to investigate the range of hospitalisation duration in dogs seen at Massey University's Veterinary Teaching Hospital's referral services between November 2013 and March 2014.

(78.3%) of the dogs lost weight, while 13.0% maintained and 8.7% gained weight. Six dogs (26.1%) lost one BCS, and three dogs (13%) lost one MCS during their hospitalisation. Of the dogs that lost condition, three lost both BCS and MCS during their stay.

The dogs lost on average −0.79 kg (SD 1.3) which represented 3.9% (SD 3.9) of their total body weight during their hospitalisation. The mean FFM difference was −0.49 kg (SD 1.6), which represented 61.8% of the total body weight lost. For FM, the mean difference was −0.3 kg (SD 1.7), which was 38.2% of the total body weight lost.

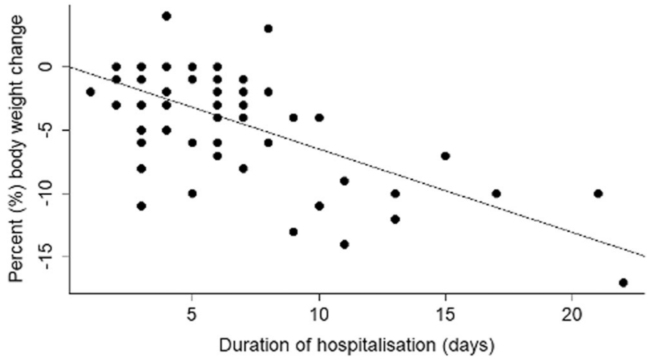

FIGURE 2 Correlation between body weight change (%) and duration of hospitalisation (days) in sixty dogs seen at Massey University's Veterinary Teaching Hospital's referral services between November 2013 and March 2014 ($r^2 = 0.4238$, $p < 0.0001$).

TABLE 2 Final diagnoses of dogs ($n = 23$) recruited for body composition analysis from Massey University's Veterinary Teaching Hospital.

Diagnosis	Count
Medical reason (diabetes, hyperadrenocorticism, urolithiasis, insulinoma)	2
Injury from a dog fight	1
Atlantoaxial instability	1
Fibrocartilaginous embolism	1
Orthopaedic fracture	2
Paralumbar abscess requiring a hemilaminectomy	1
Intervertebral disc disease (IVDD)	15

Conversely, 39.1% of dogs gained FFM and 34.8% gained FM during their stay (Table 3).

There was only a moderate correlation between BCS and percentage FM measured at admission (Kendall's $\tau = 0.51$; $p = 0.002$, Figure 3), and at discharge (Kendall's $\tau = 0.55$; $p = 0.001$, Figure 4). However, there was no correlation between MCS and FFM at either admission or discharge ($p > 0.1$). Finally, duration of stay was associated with change in overall body weight in kilograms, but was not associated with changes in either FFM or FM. No other factors (age, sex, duration, PSS, caloric intake or activity) were found to be associated with either changes in body weight, FFM or FM. The final models are presented in Table 4.

4 | DISCUSSION

The loss of total body weight, lean mass and fat mass is associated with poorer outcomes in both human and veterinary patients (Anker et al., 1997; Baez et al., 2007; Caporossi et al., 2012; Finn et al., 2010; Oreopoulos et al., 2008; Romano et al., 2016; Slinde et al., 2005; Vestbo et al., 2006). The majority of the dogs in this study lost body

TABLE 3 Average activity (ΔG) and caloric intake per day (% resting energy requirement, % RER), physical status score (PSS), body weight (BW), body condition score (BCS), muscle condition score (MCS) and body composition (fat-free mass, FFM, fat mass, FM) at admission (a) and discharge (d) of 23 long-stay hospitalised dogs.

Dog	Activity (ΔG)	Intake (% RER)	PSS	BWa (kg)	BWd (kg)	BCSa	BCSd	MCSa	MCSd	FFMa (kg)	FFMa (% BW)	FFMd (kg)	FFMd (% BW)	FMa (kg)	FMa (% BW)	FMd (kg)	FMd (% BW)
1	22,493	61.3	1	7.80	7.70	6	6	3	3	5.41	69.37	5.13	66.60	2.39	30.63	2.57	33.40
2	187,587	106.7	1	23.50	23.80	6	6	3	3	17.28	73.52	15.38	64.63	6.22	26.48	8.42	35.37
3	98,680	117.7	1	5.80	5.80	8	8	3	3	3.73	64.28	2.52	43.39	2.07	35.72	3.28	56.61
4	19,344	44.6	1	9.57	9.47	6	6	3	3	7.16	74.78	7.18	75.86	2.41	25.22	2.29	24.14
5	9874	69.3	1	6.50	6.14	4	4	3	3	4.25	65.35	4.14	67.49	2.25	34.65	2.00	32.51
6	20,727	34.1	1	12.5	12.30	5	5	3	3	8.87	70.99	9.37	76.21	3.63	29.01	2.93	23.79
7	37,101	59.2	1	7.10	6.50	5	4	3	2	5.61	79.00	4.48	68.92	1.49	21.00	2.02	31.08
8	19,750	91.0	1	6.27	6.32	5	5	3	3	4.26	67.94	4.26	67.36	2.01	32.06	2.06	32.64
9	30,543	100.6	1	5.14	5.00	4	4	3	3	4.45	86.64	4.13	82.64	0.69	13.36	0.87	17.36
10	52,661	58.6	1	7.50	6.50	5	4	3	3	5.33	71.01	5.51	84.71	2.17	28.99	0.99	15.29
11	48,229	120.4	2	8.60	7.70	4	3	2	0	7.52	87.39	6.86	89.04	1.08	12.61	0.84	10.96
12	9887	34.9	1	24.00	24.00	5	5	3	3	17.63	73.44	13.39	55.78	6.37	26.56	10.61	44.22
13	14,015	90.1	1	21.10	19.90	4	4	3	3	17.60	83.41	17.49	87.89	3.50	16.59	2.41	12.11
14	22,544	104.0	2	19.40	19.20	3	3	3	3	18.42	94.96	19.07	99.34	0.98	5.04	0.01	0.05
15	NA	88.2	3	3.87	3.78	6	6	3	3	2.40	61.95	2.52	66.61	1.47	38.05	1.26	33.39
16	23,265	98.2	2	27.40	26.80	3	3	3	3	27.38	99.92	24.52	91.48	0.02	0.08	2.28	8.52
17	17,265	60.0	3	30.80	30.80	6	6	3	3	19.75	64.12	22.51	73.08	11.05	35.88	8.29	26.92
18	54,260	78.6	1	32.70	30.00	7	6	3	3	21.69	66.33	22.93	76.42	11.01	33.67	7.07	23.58
19	10,917	39.0	1	28.00	27.00	4	4	2	2	19.12	68.3	18.87	69.88	8.88	31.70	8.13	30.12
20	20,326	92.6	1	60.00	54.40	8	7	3	3	42.94	71.57	38.78	71.29	17.06	28.43	15.62	28.71
21	47,286	56.1	1	37.50	35.00	5	4	3	2	29.23	77.94	28.50	81.41	8.27	22.06	6.51	18.59
22	31,865	54.8	1	28.40	27.60	4	4	3	3	25.84	90.99	26.99	97.80	2.56	9.01	0.61	2.20
23	61,937	122.4	1	6.57	6.18	4	4	3	3	4.97	75.60	5.10	82.55	1.60	24.40	1.08	17.45

Abbreviations: BCS, body condition score; BW, body weight; FFM, fat-free mass; FM, fat mass; MCS, muscle condition score; PSS, patient severity score; RER, resting energy requirement.

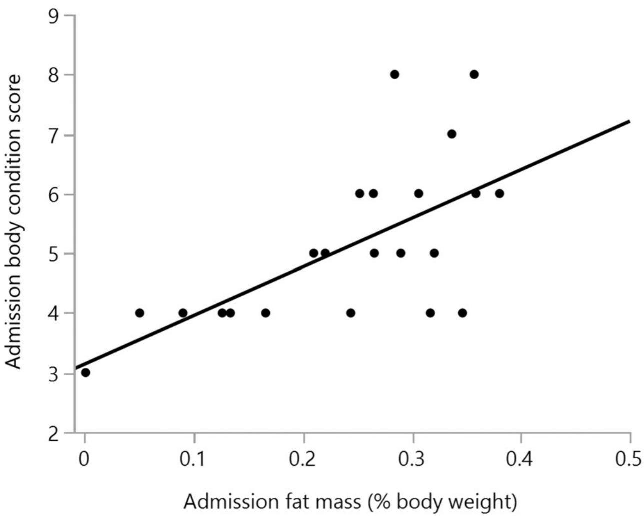

FIGURE 3 Correlation between body fat mass (expressed as % of body weight) and body condition scores at admission to a Veterinary Teaching Hospital in 23 long-stay patients (Kendall's $\tau = 0.51$; $p = 0.002$).

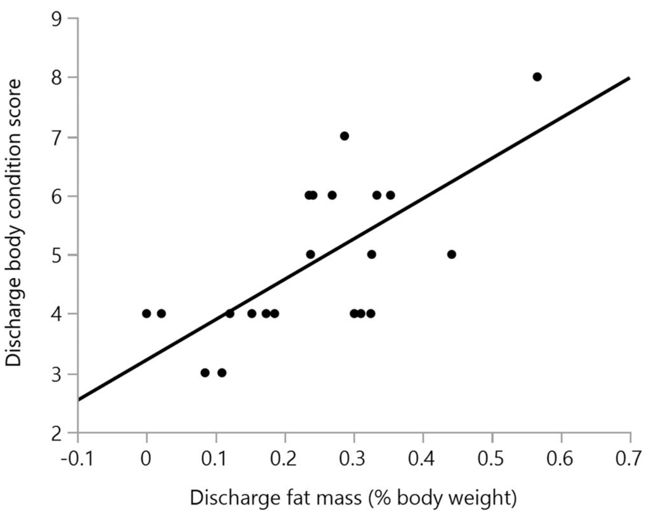

FIGURE 4 Correlation between body fat mass (expressed as % of body weight) and body condition scores at discharge from a Veterinary Teaching Hospital in 23 long-stay patients (Kendall's $\tau = 0.55$; $p = 0.001$).

weight, which consisted of a much greater proportion of lean mass (61.8%) than fat mass (38.2%). By comparison, in healthy dogs that were fed 50% of their estimated requirements for 3 months, only 30% of the total weight lost was lean mass (Blanchard et al., 2004). For humans, the percentage lean mass lost when undergoing a purposeful weight loss programme is approximately 20% of their total weight loss (Das et al., 2003; Leibel et al., 1995). In this study, dogs lost a much higher percentage of lean mass, despite them consuming on average 77.5% of their estimated energy requirements. In addition, intake was not found to be a significant factor in the multiple regression model for lean loss. This is in

TABLE 4 Multiple regression models for factors associated with changes in body weight, fat-free mass (FFM), and fat mass (FM) in hospitalised dogs.

Outcome	Variable	Estimate	Standard error	R^2	*p* Value
ΔBody weight (kg)	Intercept	1.198	0.757		0.130
	Age	−0.086	0.071		0.240
	Sex (male)	−0.621	0.454		0.188
	Days	−0.104	0.026		<0.001*
	Overall model			0.41	0.005*
ΔFat-free mass (kg)	Intercept	2.111	1.830		0.264
	Age	−0.221	0.124		0.101
	Sex (male)	−0.754	0.708		0.301
	% RER	−0.024	0.014		0.107
	Overall model			0.10	0.218
ΔFat mass (kg)	Intercept	−1.354	1.893		0.484
	Age	0.140	0.139		0.328
	Days	−0.063	0.044		0.168
	PSS	−0.737	0.622		0.252
	% RER	0.020	0.016		0.213
	Overall model			0.01	0.400

Note: Difference in body weight, FFM and FM between discharge and admission are expressed as delta (Δ) change in kilograms (kg). Age, sex, disease severity, caloric intake, duration of stay, and activity were treated as independent variables. The best fit models shown here were assessed using Akaike information criterion and adjusted R^2. The symbol (*) denotes a *p* value of < 0.10.

Abbreviations: PSS, physical status score; RER, resting energy requirement.

agreement with a meta-analysis of human hospital intervention studies where there was no clear association between energy and protein intake, and changes in skeletal muscle mass in the patients (Lambell et al., 2018). Taken together, these findings suggest that the loss of muscle mass in hospitalised patients is not simply due to undereating, and instead, is likely to be caused by several factors.

Protein metabolism can change in human patients during hospitalisation, and may do so soon after admission. Protein synthesis decreases in ICU patients, while protein catabolism increases, and significant muscle mass loss can occur even within the first week of hospitalisation (Puthucheary et al., 2013). Nonnutritional factors that can contribute to muscle loss include increased production of cortisol and inflammatory cytokines, development of acidosis, disuse atrophy and denervation (Gordon & Mao, 1994; May et al., 1986; Paddon-Jones, Sheffield-Moore, et al., 2006; Williams et al., 1991). Injury to the muscle itself increases IL-6, TNF-α and cortisol production, leading to local and more general muscle catabolism (Castro et al., 2000; Gorgey & Dudley, 2007; Paddon-Jones, Sheffield-Moore, et al., 2006).

The majority of the patient population in this study were spinal patients, and so partial denervation and disuse atrophy were likely significant contributors to the muscle loss (Castro et al., 2000; Gordon & Mao, 1994; Gorgey & Dudley, 2007). The patients were also recovering from surgery, which is a process that also induces muscle injury. An elevation in plasma creatine kinase lasting for three days postsurgery has been described in dogs undergoing hemilaminectomy surgery (Wilson et al., 2018). Likewise, in humans following elective surgery, an increase in cortisol has been documented for three days postsurgery, which reached concentrations that have been shown experimentally to exacerbate muscle loss during bedrest (Ferrando et al., 1999; Paddon-Jones, 2006; Padova et al., 2008). Thus, it is likely that a combination of these factors played a role in the muscle loss of the patients in this study.

When examining for factors associated with changes in body weight and composition, the only significant association was between hospital duration and the loss of body weight. This is a similar finding to another study of hospitalised dogs where longer hospitalisation was associated with changes in body weight (Molina et al., 2018). Interestingly though, no association was found between the duration of hospitalisation and the amount of lean mass lost. The reason for this could be due to our small sample size, but may also be due to differences in post-op management, discharge criteria, availability of home care, and the severity of the patients' spinal disease, all of which can affect how long a patient stays in the hospital. While systemic disease severity was measured using PSS, this measurement is not specific to grading the severity of spinal disease. Additionally, there may be a nonlinear relationship between the loss of muscle and patient recovery, which would not be captured in the multiple regression analysis. Larger changes in composition in the early days or later days of hospitalisation may influence a patient's recovery and subsequent discharge from the hospital differently.

Activity was not found to be associated with changes in lean mass, which was unexpected. Activity was monitored using a tri-axial accelerometer fitted to the collar of each dog, which measures both the magnitude and direction of acceleration. Spinal patients recovering during hospitalisation are mostly inactive during cage rest, and experience only limited opportunities for activity outside the cage. In addition, spinal patients received daily physical therapy, which may reduce muscle loss, but the movements elicited during the therapy would not be fully captured using a collar-mounted accelerometer. As such, other methods of monitoring activity and movements related to physical therapy may be needed to better understand the relationship between disuse atrophy, activity and lean mass changes during hospitalisation.

While MCS and BCS are commonly used in veterinary medicine to determine nutritional status, it is unknown how accurately these measures detect changes in lean and fat mass in hospitalised patients. A correlation was found between BCS and FM in this population of hospitalised dogs, whereas there was no correlation between MCS and FFM. However, the strength of the correlation was only modest, and so the utility of BCS for monitoring hospitalised patients is low. BCS is measured at fixed points on the body and may not detect all changes in fat mass that might occur during hospitalisation. In a study of human patients with acute pancreatitis, the patients lost 12% of their visceral fat mass, but had no change in their subcutaneous fat mass after nine days of hospitalisation (Brewster et al., 2014). Thus BCS appears to be insufficiently sensitive to allow for accurate monitoring of changes in the overall fat mass of individual hospitalised patients.

Another concern is the accuracy of BCS to adequately assess fat mass specifically in lean animals. The BCS system was originally validated based on ideally condition or overweight animals, and its accuracy in lean animals has not been properly assessed (Laflamme 1997b, 1997a; Mawby et al., 2004). Given that the BCS system is largely based upon subcutaneous fat stores, it is likely to be inaccurate in animals with little subcutaneous fat. By comparison, the isotopic dilution technique has been validated across dogs of different sizes and body conditions, including lean dogs (Burkholder & Thatcher, 1998). Indeed in this study population, three dogs with minimal body fat as measured by isotopic dilution (0.08% –12.61%), were still scored a BCS of three. Other studies in greyhounds have also described very low body fat mass as measured by both deuterium isotopic dilution and dual-energy X-ray absorptiometry (DEXA), despite the dogs being considered ideally conditioned (Hill et al., 2001, 2005). In agreement, two of the three leanest dogs in this study were greyhounds. Therefore, in very lean dogs, BCS also appears to be an inaccurate measure of body fat mass.

The main limitation to this study is the small total study population; we only recruited 23 out of the 34 patients we were aiming for and most were patients with spinal cord disease. Patients from the medicine department were ultimately challenging to recruit due to difficulties in estimating duration of stay, or because they had water imbalances and/or needed IVF therapy. As such, the study population was heavily biased towards spinal patients. These patients may have exacerbated muscle loss from inflammatory diseases and anorexia compared to surgical patients that are otherwise systemically healthy. A study of patients undergoing hemilaminectomies found a rise in cell-free DNA and creatine kinase in plasma for several days post-operatively, which was not seen in patients undergoing surgery for cranial cruciate ruptures and ovariohysterectomies, suggesting greater tissue trauma and inflammation (Wilson et al., 2018). As such, due to the limited disease types and degrees of illness, care should be taken with extrapolating the results of this study to all hospitalized patients. Future studies of hospitalized patients should utilise a method that is not so affected by hydration status, such as DEXA or bioimpedance (although neither of those methods are completely unaffected), and include a larger, more varied patient population with different diseases and illness severity.

Another consideration is that the true energy expenditures of the patients were not measured, and so food provision was based on an estimated energy requirement. However, other studies using indirect calorimetry have shown that the energy requirements of hospitalised dogs is similar to healthy dogs (O'Toole et al., 2004; Walton et al., 1996). Thus the RER calculation used in this study is believed to be close to true requirements. In addition, ideal body weight was used to calculate RER, which was based on the body condition score and current weight of the patient at admission. This was performed to avoid overfeeding overweight patients, and underfeeding underweight patients, as the energy requirement of an animal is mostly related to the quantity of lean mass, rather than fat mass (Browning & Evans, 2015; Cameron et al., 2016; Johnstone et al., 2005; Larsson et al., 2014; Martin et al., 2022; Pouteau et al., 2000). There is currently no consensus in the veterinary community as to whether ideal body weight or current body weight should be used to calculate RER, as both methods are commonly described in the literature (Hall et al., 2018; Kathrani & Parkes, 2022; Michel & Higgins, 2006; Norton et al., 2016; Tolbert et al., 2022; Tsuruta et al., 2016). Nonetheless, in this study, there were eight patients that were considered overweight (BCS > 5) and two patients that were underweight (BCS < 4) and had their body weights adjusted when performing the energy calculation. The concern would be that using ideal weight could lead to underfeeding in overweight patients. However, only two of the overweight dogs in the study lost notable body weight and BCS. In addition, the dogs that were underweight did not gain weight as the result of excess feeding, and instead maintained their BCS throughout the duration of their hospitalization. These results indicate that the patients were not likely being under or overfed. Furthermore, the majority of the patients consumed less than the amount of food that was offered, and thus intake was limited by appetite, and not an estimation of RER.

5 | CONCLUSION

This is the first study to measure body composition changes in hospitalised dogs using isotopic dilution. It was found that long-stay hospitalised dogs usually lose weight during hospitalisation, and that the majority of tissue lost was lean tissue. In addition, the proportion of the total weight lost as lean mass was greater that obese dogs losing weight on a planned weight loss programme. Intake was not significantly associated with either changes in body weight or body condition, which highlights that the weight lost by hospitalised dogs is not simply due to undereating. However, larger studies containing patients with a wider range of disease are needed to better evaluate other factors, such as systemic inflammatory states, that may affect lean and fat loss during hospitalisation. In addition, it has been shown that the most common methods of measuring body composition (BCS and MCS) are not suitable for monitoring changes during hospitalisation, especially in lean dogs.

AUTHOR CONTRIBUTIONS

Y. Becca Leung, Nick Cave, and Timothy J. Wester contributed to the conception and design of the study. Y. Becca Leung collected the data and performed the statistical analysis with assistance from Nick Cave. Y. Becca Leung wrote the first draft of the manuscript and Nick Cave and Timothy J. Wester contributed to the manuscript revision, read, and approved the submitted version.

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CONFLICTS OF INTEREST STATEMENT

The authors declare no conflict of interest.

DATA AVAILABILITY STATEMENT

Data available on request from the authors.

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