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Abdominal fat content assessment by computed tomography in toy breed dogs

*Juhyang Park, Daji Noh, Kija Lee**

Department of Veterinary Medical Imaging, College of Veterinary Medicine, Kyungpook National University, Daegu, Republic of Korea

**Corresponding author: leekj@knu.ac.kr*

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Abstract: The aim of this retrospective study was to assess the abdominal fat distribution in toy breed dogs using computed tomography (CT) in relation to the breed, age, and sexual status. In 140 dogs (52 Maltese, 33 Poodles, 32 Shih-Tzus, and 23 Yorkshire Terriers), the total fat area (TA), visceral fat area (VA), subcutaneous fat area (SA) and body area (BA) were measured at the third and sixth lumbar vertebral level on non-contrast transverse CT images. The differences in the TA/BA and VA/SA according to the breed, age, and sexual status, and correlations with the age were analysed. The differences in the TA/BA and VA/SA among the breeds were revealed ($P < 0.05$). There was no difference for the TA/BA among the sexual statuses, but the VA/SA was higher in spayed females than in intact females ($P = 0.001$). Positive correlation of the age with the TA/BA in the Maltese, Poodles, and intact females, and the age with the VA/SA in the Maltese, Shih-Tzus, Yorkshire Terriers, neutered males, and spayed females were found. The results showed that the abdominal fat composition varied according to the breed, age, and sex, which may have implications on defining obesity-related disease risks in different populations. Careful monitoring of the VA/SA in the breed (Maltese, Shih-Tzu, and Yorkshire Terrier), age (senior dogs), and sexual status (neutered dogs) may be required.

Keywords: body fat; CT; small-sized dogs; visceral obesity

Obesity is a multifactorial disease defined as abnormal, excessive and ectopic fat accumulation (Muller et al. 2014). The incidence of canine obesity was reported as being 20.4–44.4% in previous studies (Courcier et al. 2010; Mao et al. 2013). Ageing, neutering, and becoming overweight or obese owners are risk factors for obesity in dogs (Bjornvad et al. 2019). Obesity increases inflammatory cytokines causing chronic low-grade inflammations (German et al. 2010). In humans, it is associated with insulin resistance, neoplasia, osteoarthrosis and earlier morbidity (Kealy et al. 2002; Gayet et al. 2004; Lund et al. 2006). Compared to subcutaneous obesity, visceral obesity may influence the adipokine, insulin and cytokine regulation which are related to the development of obesity-related diseases, such as insulin resistance and heart disease in human and veterinary medicine (Fujioka et al. 1987; Park and Lee 2005; Kamimura et al. 2013; Muller et al. 2014; Thengchaisri et al. 2014; Turner et al. 2020). Thus, the accurate diagnosis of visceral obesity is important.

The primary methods used in veterinary practice to assess the body fat are the body weight measurement and the body condition score (BCS) system. The measurement of the body weight is a rapid,

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objective, and repeatable method, but it cannot quantify the body fat (Santarossa et al. 2017). The BCS system is also a rapid, inexpensive, and non-invasive method. However, it is a subjective method based on a visual inspection and palpation, which may result in inter-observer variation and unreliability when assessing the visceral fat (Linder et al. 2013). As the visceral fat is related to the insulin resistance and heart disease in veterinary medicine (Muller et al. 2014; Thengchaisri et al. 2014), the accurate assessment of the visceral fat may be helpful in small breed dogs. Computed tomography (CT) has become more widely used for evaluating obesity because it is well-correlated with the true body fat content (Ishioka et al. 2005; Kamimura et al. 2013). Furthermore, CT enables the separate assessment of the visceral and subcutaneous fat to define the fat distribution (Ishioka et al. 2005). The use of CT for the body fat assessment in dogs has been reported in several studies (Ishioka et al. 2005; Purushothaman et al. 2013; Kobayashi et al. 2014; Kim et al. 2018; Nagao et al. 2019; Turner et al. 2020).

We hypothesised that the breed, age, and sexual status may affect the abdominal visceral and subcutaneous fat distribution. There are few reports comparing the fat content evaluated by CT among different sexual statuses (Kobayashi et al. 2014; Kim et al. 2018; Turner et al. 2020), and one report analysing the effect of ageing on the body fat content using CT (Turner et al. 2020). The purpose of this study was to assess the relative fat distribution to the visceral and subcutaneous department by CT in relation to the breed, age, and sexual status in Maltese, Poodle, Shih-Tzu, and Yorkshire Terrier dogs without diseases affecting the abdominal fat status.

MATERIAL AND METHODS

Animals

This retrospective, cross-sectional, descriptive study included Maltese, Poodle, Shih-Tzu, and Yorkshire Terrier dogs. To exclude the effect of the brown fat, which has a higher water content compared to white fat (Stieger-Vanegas and Frank 2018), dogs under one year old were excluded from the study. The medical records were searched for the four breed dogs of one year old or older, that had a non-contrast CT scan of the abdomen performed for diagnostic purposes between March 2015 and September 2019. Due to the retrospective nature of this study, approval by the Animal Care and Use Committee of the institution was not required. All the clients had previously signed informed consent for the use of their dogs' data for research purposes. Patients with large abdominal masses, abdominal herniation, ascites, lipoma, subcutaneous masses, or mammary gland tumours at the L3 or L6 level were excluded. Patients with peritonitis or a recent history of abdominal surgery, which could change the abdominal fat status, were also excluded. The medical records were reviewed, and the following data were recorded: breed, age, sexual status, and body weight.

The dogs were grouped by breed, age, and sexual status. The dogs were grouped into four breeds: Maltese, Poodles, Shih-Tzus, and Yorkshire Terriers. Based on previous studies, the dogs were categorised into three age groups: young (one–three years old), mid-age (four–eight years old), and senior (≥ nine years old) (White et al. 2011; Bjornvad et al. 2019). The dogs were also classified by four sexual statuses: intact male, neutered male, intact female, and spayed female.

CT examinations

Under general anaesthesia, the dogs were positioned in dorsal recumbency on the CT table. The CT examination was performed using a 32-Multislice CT scanner (Alexion; Toshiba Medical System, Otowara, Japan). The scanning parameters were as follows: 120 kV, 200 mA, and 1.0 mm slice thickness, and 0.75 s of rotation time, with field of view adjusted to the patient size. The CT images were acquired from the diaphragm to the coxofemoral joint, with a craniocaudal direction. All the CT data were reconstructed and evaluated with non-contrast transverse images on a soft-tissue window [window width, 450 Hounsfield units (HU); window level, 40 HU] and sagittal images on a bone window (window width, 2 000 HU; window level, 600 HU).

Imaging assessment

On the transverse CT images where spinous processes of L3 and L6 were the largest, the body area

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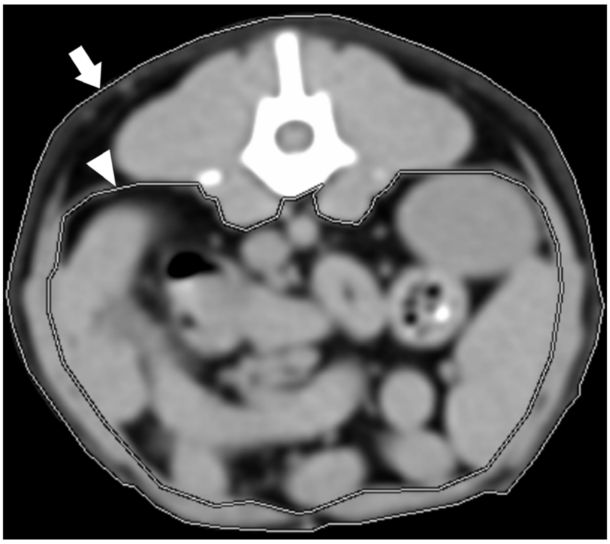

Figure 1. Measurement of the body area and the fat area at the L3 level

The areas were measured using non-contrast transverse CT imaging (window width, 450 HU; window level, 40 HU). The ROI is drawn around the body (arrow) to measure the BA and TA. To measure the TA, an attenuation range of −135 HU to −105 HU was used. Another ROI is drawn surrounding the peritoneal cavity (arrowhead) to measure the VA. The VA is subtracted from the TA to calculate the SA

BA = body area; CT = computed tomography; HU = Hounsfield units; L3 = third lumbar vertebra; ROI = region of interest; SA = subcutaneous fat area; TA = total fat area; VA = visceral fat area

(BA), total fat area (TA), visceral fat area (VA), and subcutaneous fat area (SA) were measured based on previous studies (Ishioka et al. 2005; Kim et al. 2018; Nagao et al. 2019) (Figure 1). Regions of interest (ROI) were drawn manually along the skin-air interface to assess the BA and TA. To gauge the VA, another ROI was drawn along the abdominal wall musculature. The SA was calculated by subtracting the VA from the TA. The fat areas were measured using an attenuation range of −135 to −105 HU (Ishioka et al. 2005).

To standardise the fat areas, the TA, VA, and SA were divided by the L6 length ($_r$TA, $_r$VA, and $_r$SA). The length of the L6 was measured on the sagittal CT images with the bone window. Based on previous studies, the ratio of the TA to BA (TA/BA) and the ratio of the VA to SA (VA/SA) (Ishioka et al. 2005; Kobayashi et al. 2014; Kim et al. 2018; Nagao et al. 2019) were calculated to compare the abdominal fat content among the breeds, age groups and sexual statuses.

Statistical analysis

The statistical analyses were performed using SPSS v25.0 (IBM SPSS Statistics, USA). The normality of the $_r$TA, $_r$VA, $_r$SA, TA/BA, and VA/SA was evaluated using the Shapiro-Wilk test. For the parametric data, a paired *t*-test, one-way analysis of variance (ANOVA) with a Bonferroni post-hoc test, and Pearson's correlation coefficient were used. For the non-parametric data, Wilcoxon's signed rank test, the Kruskal-Wallis test with a Bonferroni correction, the Mann-Whitney *U*-test, and Spearman's correlation coefficient were used. A P-value of < 0.05 was considered statistically significant in all the analyses.

Furthermore, for identification of the fat accumulation sites, the differences in the fat area ($_r$TA, $_r$VA, and $_r$SA) between L3 and L6 in each breed, age group, and sexual status were analysed using a paired *t*-test or Wilcoxon's signed rank test. To identify the differences in the fat ratios according to the breed, age, and sexual status, the differences of the TA/BA at L3 and L6 among the breeds, and age groups, the VA/SA at L6 among the breeds, and the VA/SA at L3 and L6 among the age groups were analysed using a one-way ANOVA with a Bonferroni post-hoc test.

In addition, the differences in the TA/BA and VA/SA at L3 and L6 among the sexual statuses, and the VA/SA at L3 among the breeds were analysed using the Kruskal-Wallis test with the Bonferroni correction. The correlations of the age with the TA/BA, and with the VA/SA at L3 and L6 in each breed and sexual status were analysed using Pearson's correlation coefficient or Spearman's correlation coefficient.

RESULTS

A total of 140 dogs met the inclusion criteria, aged from one year to 18.3 years (mean age 9.4 years). The dogs consisted of 52 Maltese, 33 Poodle, 32 Shih-Tzu, and 23 Yorkshire Terrier dogs. There were 11 young, 49 mid-age, and 80 senior dogs. There were 61 males (9 intact and 52 neutered) and 79 females (29 intact and 50 spayed).

The abdominal fat area and the length of the L6 for each breed, age, and sexual status are summarised in Table 1. Table 2 summarises the abdominal fat content distribution for each breed, age, and

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Table 1. Mean ± standard deviation values of the abdominal fat areas and the length of the L6 in each group

Variable	Level	BA (mm^2)	TA (mm^2)	VA (mm^2)	SA (mm^2)	L6 (mm)
Maltese	L3	10 064.5 ± 3 714.0	2 516.6 ± 1 738.1	1 557.8 ± 989.9	958.8 ± 843.0	14.4 ± 1.8
	L6	8 436.8 ± 3 315.7	2 217.6 ± 1 711.7	874.4 ± 576.5	1 343.2 ± 1 309.8	
Poodle	L3	9 312.7 ± 3 219.2	2 050.1 ± 1 351.6	1 289.6 ± 829.7	760.5 ± 589.1	14.0 ± 1.9
	L6	7 671.1 ± 2 832.1	1 747.0 ± 1 053.9	615.7 ± 430.2	1 131.3 ± 692.8	
Shih-Tzu	L3	9 780.3 ± 3 465.6	2 264.2 ± 1 520.3	1 343.1 ± 841.6	921.1 ± 797.1	14.5 ± 2.1
	L6	8 041.9 ± 3 041.0	1 847.8 ± 1 274.8	598.0 ± 431.7	1 249.8 ± 990.1	
Yorkshire Terrier	L3	10 086.4 ± 3 285.7	2 527.0 ± 1 671.4	1 595.2 ± 1 091.6	931.8 ± 702.1	15.0 ± 2.5
	L6	8 423.6 ± 2 891.9	1 998.9 ± 1 352.2	856.8 ± 656.2	1 142.1 ± 814.1	
Young	L3	7 930.0 ± 2 785.8	1 440.4 ± 1 108.3	815.5 ± 607.2	625.0 ± 603.4	13.4 ± 2.4
	L6	6 679.1 ± 2 546.0	1 185.9 ± 1 003.0	448.5 ± 369.3	737.3 ± 688.2	
Mid-age	L3	10 307.3 ± 3 668.4	2 620.8 ± 1 734.3	1 626.6 ± 977.6	994.2 ± 854.2	14.5 ± 1.8
	L6	8 618.0 ± 3 317.2	2 306.4 ± 1 722.4	914.4 ± 569.5	1 392.0 ± 1 333.1	
Senior	L3	9 791.7 ± 3 344.5	2 310.4 ± 1 515.8	1 432.0 ± 920.6	878.4 ± 700.6	14.5 ± 2.1
	L6	8 089.9 ± 2 921.2	1 900.2 ± 1 210.4	686.1 ± 514.9	1 214.1 ± 834.3	
IM	L3	9 549.9 ± 3 330.1	2 061.1 ± 1 298.6	1 428.7 ± 913.9	632.4 ± 396.8	14.0 ± 1.2
	L6	8 037.8 ± 2 859.6	1 725.6 ± 1 128.6	750.2 ± 574.1	975.4 ± 566.9	
NM	L3	9 870.8 ± 3 449.5	2 389.0 ± 1 610.8	1 448.9 ± 972.2	940.1 ± 770.1	14.7 ± 2.3
	L6	8 174.8 ± 3 028.8	1 910.6 ± 1 320.0	699.1 ± 546.6	1 211.5 ± 937.5	
IF	L3	9 182.1 ± 3 104.2	1 976.8 ± 1 324.9	1 207.8 ± 783.3	769.0 ± 602.3	13.9 ± 2.0
	L6	7 551.1 ± 2 699.5	1 713.1 ± 1 019.7	571.0 ± 394.3	1 142.1 ± 691.7	
SF	L3	10 202.4 ± 3 705.8	2 579.7 ± 1 740.9	1 600.1 ± 985.5	979.6 ± 851.7	14.5 ± 1.8
	L6	8 530.7 ± 3 340.7	2 270.3 ± 1 723.7	899.3 ± 573.5	1 371.0 ± 1 327.8	

BA = body area; NM = neutered male; IF = intact female; IM = intact male; L3 = third lumbar vertebra; L6 = sixth lumbar vertebra; SA = subcutaneous fat area; SF = spayed female; TA = total fat area; VA = visceral fat area

Table 2. Mean ± standard deviation values and the differences in the fat areas between the L3 and L6 in each group

Variable	*n*	$_{r}$TA		$_{r}$VA		$_{r}$SA	
		L3	L6	L3	L6	L3	L6
Maltese	52	121.3 ± 71.3	101.0 ± 61.4*	75.9 ± 46.2	37.3 ± 27.2*	45.4 ± 34.9	63.7 ± 43.6*
Poodle	33	164.1 ± 94.0	140.6 ± 78.4*	98.9 ± 52.8	53.8 ± 33.3*	65.2 ± 47.6	86.8 ± 62.8*
Shih-Tzu	32	204.3 ± 105.4	174.1 ± 100.9*	126.1 ± 66.4	64.7 ± 36.9*	78.2 ± 46.3	109.4 ± 70.4*
Yorkshire Terrier	23	176.6 ± 125.0	148.5 ± 121.9*	111.9 ± 67.7	56.8 ± 36.4*	64.7 ± 63.6	91.7 ± 95.7*
Young	11	48.0 ± 34.7	32.11 ± 31.9	26.5 ± 19.4	12.8 ± 11.7*	21.5 ± 17.0	30.2 ± 21.8*
Mid-age	49	147.6 ± 94.4	122.6 ± 73.6*	85.0 ± 53.2	38.8 ± 26.7*	62.6 ± 47.9	83.8 ± 55.7*
Senior	80	182.0 ± 97.6	154.9 ± 97.0*	117.0 ± 57.1	63.1 ± 33.9*	65.0 ± 47.9	91.8 ± 73.9*
IM	9	144.6 ± 89.3	122.2 ± 80.6*	100.2 ± 63.4	53.3 ± 41.9*	44.4 ± 26.8	68.9 ± 39.4*
NM	52	158.8 ± 97.7	126.9 ± 81.0*	96.0 ± 58.2	45.9 ± 32.1*	62.8 ± 48.5	81.0 ± 59.7*
IF	29	137.6 ± 82.5	119.3 ± 62.8*	84.8 ± 51.7	40.0 ± 26.8*	52.8 ± 35.7	79.2 ± 40.7*
SF	50	175.5 ± 111.3	154.4 ± 112.3*	109.4 ± 63.8	61.3 ± 36.3*	66.1 ± 54.5	93.1 ± 87.4*

IF = intact female; IM = intact male; L3 = third lumbar vertebra; L6 = sixth lumbar vertebra; *n* = number of dogs; NM = neutered male; $_{r}$SA = ratio of the subcutaneous fat area to the length of L6 body; $_{r}$TA = ratio of the total fat area to the length of L6 body; $_{r}$VA = ratio of the visceral fat area to the length of L6 body; SF = spayed female

*$P < 0.05$ when compared with the value at L3

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Table 3. TA/BA and VA/SA among the groups and correlation coefficients with the age

Variable	TA/BA				VA/SA			
	L3		L6		L3		L6	
	mean ± SD	r	mean ± SD	r	mean ± SD	r	mean ± SD	r
Maltese	18.6 ± 5.8^{ab}	0.33*	18.8 ± 6.3^{ab}	0.30*	1.8 ± 0.9	0.47*	0.6 ± 0.3	0.39*
Poodle	23.8 ± 7.7^{a}	0.46*	24.8 ± 7.0^{a}	0.28	1.5 ± 0.6^{a}	0.21	0.6 ± 0.4	0.15
Shih-Tzu	25.7 ± 7.9^{b}	0.27	26.3 ± 8.0^{bc}	0.21	1.6 ± 0.5	0.08	0.6 ± 0.3	0.44*
Yorkshire Terrier	21.0 ± 8.4	–0.32	20.3 ± 7.1^{c}	–0.19	2.2 ± 1.1^{a}	0.63*	0.7 ± 0.3	0.69*
Young	11.1 ± 6.9^{ab}	NA	11.7 ± 7.2^{ab}	NA	1.3 ± 0.5^{a}	NA	0.4 ± 0.2^{a}	NA
Mid-age	20.9 ± 7.5^{a}	NA	21.1 ± 7.1^{a}	NA	1.4 ± 0.6^{b}	NA	0.5 ± 0.3^{b}	NA
Senior	23.5 ± 6.4^{b}	NA	23.3 ± 6.7^{b}	NA	2.2 ± 1.1^{ab}	NA	0.8 ± 0.4^{ab}	NA
IM	19.6 ± 8.0	0.27	19.4 ± 7.6	0.57	2.1 ± 0.5	0.52	0.7 ± 0.2	0.56
NM	21.8 ± 8.5	0.13	20.9 ± 8.3	0.08	1.9 ± 1.1	0.49*	0.6 ± 0.3^{a}	0.55*
IF	19.1 ± 8.4	0.58*	20.6 ± 7.3	0.62*	1.6 ± 5.3	0.29	0.5 ± 0.2^{b}	0.34
SF	22.8 ± 6.0	0.26	23.1 ± 5.4	0.28	2.0 ± 1.0	0.33*	0.8 ± 0.4^{ab}	0.05

IF = intact female; IM = intact male; L3 = third lumbar vertebra; L6 = sixth lumbar vertebra; NA = not applicable; NM = neutered male; r = correlation coefficient of the fat ratio with age for each measuring level; SD = standard deviation; SF = spayed female; TA/BA = the ratio of the total fat area to body area; VA/SA = the ratio of the visceral fat area to subcutaneous fat area

[a–c]Significantly different between the groups with the same superscripted letter at the same measuring level; *P < 0.05

sexual status compared by the respective measuring levels. The $_r$TA was significantly higher at L3 than L6 in all the breeds, age, and sexual status groups (P < 0.05), except in the young dogs. In the young dogs, no significant difference in the $_r$TA between L3 and L6 was found. The $_r$VA was significantly higher at L3 than at L6. In contrast, the $_r$SA was significantly higher at L6 than L3 in all the breed, age, and sexual status groups (P < 0.05). Table 3 summarises the differences in the TA/BA and VA/SA among the breeds, age groups, and sexual statuses. In addition, correlations of the age with the TA/BA, and with the VA/SA in each breed and sexual status were analysed and the results are summarised in Table 3.

DISCUSSION

CT is useful for quantifying the fat content and distribution. This enables the separate assessment of the visceral and subcutaneous fat (Ishioka et al. 2005). This is the first study to analyse the effect of the breed on the body fat content distribution in toy breed dogs using CT. In the present study, the L3 and L6 levels were chosen for the quantification of the abdominal fat content, which are suitable locations for the evaluation of the total and visceral fat, and the evaluation of the subcutaneous fat, respectively (Ishioka et al. 2005; Kim et al. 2018). In addition, the fat area measured at L3 using the attenuation range of –135 HU to –105 HU had the best correlation with the true body fat as calculated by the deuterium oxide dilution method (Ishioka et al. 2005). Therefore, the same attenuation range was chosen for the quantification of the abdominal fat content in the present study.

Our results showed that the total and visceral fat tend to accumulate at L3, whereas the subcutaneous fat tends to accumulate at L6 in all four breeds, findings that are consistent with the results of previous studies (Kim et al. 2018; Nagao et al. 2019). Therefore, it is thought that the fat distribution tendency remains the same regardless of the respective breed. On the other hand, the fat accumulation ratio may have a breed effect as the Shih-Tzu and Poodle dogs had a higher TA/BA than the Maltese dogs at both the L3 and L6 levels. In addition, at the L6 level, the Yorkshire Terriers showed a lower TA/BA than the Shih-Tzus, and a lower VA/SA than the Poodle dogs. These differences in the fat accumulation ratio may imply differences in the body shapes according to the breed, providing the first objective evidence

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of the slender body shape of Maltese dogs as compared to Shih-Tzu and Poodle dogs, and a slenderer loin of Yorkshire Terrier dogs as compared to Shih-Tzu dogs. Regarding positive correlations of the TA/BA with the age in Maltese and Poodle dogs, the risk of obesity is expected to increase with ageing. The positive correlations of the VA/SA with the age in Maltese, Shih-Tzu, and Yorkshire Terrier dogs imply the re-allocation of the adipose tissue from the subcutaneous compartment to the visceral compartment. The result is consistent with a recent study which showed increased fat distribution to the peritoneum relative to the subcutaneous space with an increasing age (Turner et al. 2020). These re-allocations may be a concern because of the protective effects of the subcutaneous fat against metabolic syndromes in humans, particularly among those carrying a high visceral fat load (Demerath et al. 2008; Demerath et al. 2011). Although no study has reported such effects of the subcutaneous fat in veterinary medicine, Muller et al. (2014) and Thengchaisri et al. (2014) found a positive correlation between the visceral to subcutaneous fat volume ratio and insulin resistance, and cardiovascular disease. Further studies to define the protective effect of the subcutaneous fat against metabolic syndromes in dogs are needed. In addition, careful monitoring of the VA/SA in Maltese, Shih-Tzu, and Yorkshire Terrier dogs may be required with ageing to protect against the development of visceral obesity-related diseases. The Maltese especially, in which not only the VA/SA, but also the TA/BA showed positive correlations with the age, may require more circumspective monitoring as the dogs get older.

This is also the second study to analyse the effect of ageing on the visceral and subcutaneous fat distribution by CT. Consistent with a previous study of fat distribution in Beagle dogs (Kim et al. 2018), the total and visceral fat tended to accumulate at L3 and the subcutaneous fat tended to accumulate at L6 in all three age groups, except that the total fat content at L3 and L6 were not significantly different in young dogs. These results support the trends of total fat distribution may differ by age groups. However, as there were only a small number of young dogs included in the present study, a further study with a larger group of young dogs is needed. In addition, the TA/BA and VA/SA were different among age groups, implying an age effect on the fat accumulation ratio. In previous studies, the peak prevalence of obesity has appeared at about 10 years of age and then declined, suggesting young and mid-age dogs as primary targets for obesity prevention (Lund et al. 2006). In the studies, obesity was diagnosed based on the BCS of the dog, which is correlated more with the subcutaneous fat rather than with the visceral fat (Linder et al. 2013). In addition, it has a strong correlation with the TA/BA, and a poor correlation with the VA/SA (Kim et al. 2018). In the present study, the TA/BA was not different between mid-age and senior dogs, and the VA/SA was higher in senior than mid-age dogs. The result implies the silent increase in the VA/SA without a change in the BCS, consistent with a recent study which showed an increased fat distribution to the peritoneum relative to the subcutaneous space with an increasing age (Lund et al. 2006). Therefore, the careful monitoring of the VA/SA in senior dogs against the risk of visceral obesity-related diseases would be helpful even when an increase in the BCS does not take place. In addition, senior dogs should also be included in obesity prevention programmes, especially for those which target visceral obesity.

There are only few studies comparing the fat distribution evaluated by CT among different sexual statuses (Kobayashi et al. 2014; Kim et al. 2018; Turner et al. 2020). Our study is the first study to analyse the correlation of the fat ratio with age in detailed sexual status, including whether or not neutered, without distinguishing only as male or female. A previous study comparing the fat accumulation between male and female dogs revealed higher visceral fat than subcutaneous fat accumulation in male dogs, and higher subcutaneous than visceral fat accumulation in female dogs (Kim et al. 2018). However, the study had an imbalanced number of males and females and did not consider the effect of neutering. On the other hand, a more recent study found no significant difference in the visceral to subcutaneous fat ratio between male and female dogs (Turner et al. 2020). The difference may have resulted from the different methodology in the fat measurement using CT between the studies. Another study comparing the visceral and subcutaneous fat distribution before and after neutering reported an increase in the total fat and a more pronounced accumulation of subcutaneous fat than visceral fat after neutering, but the study was performed in only three male dogs (Kobayashi et al. 2014).

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The total and visceral fat showed tendencies to accumulate at L3 and the subcutaneous fat at L6 in all four sexual status categories, implying that the fat distribution tendency was the same regardless of the sexual status. However, the fat accumulation ratio was found to be sexually affected. A higher VA/SA was noted in spayed female dogs compared to intact female dogs indicating that neutering may cause an increased visceral fat and a decreased subcutaneous fat content in female dogs. This result is different from recent findings, which proposed no effect of neutering on the BCS and the risk of obesity in female dogs (Bjornvad et al. 2019). The difference may have resulted from the fact that the separation of the visceral and subcutaneous fat was not conducted in the study. In the present study, considering that the TA/BA did not differ between intact female and spayed female dogs, the BCS may have not changed even if the VA/SA had increased. Therefore, spayed female dogs may require careful monitoring of the VA/SA against the risk of visceral obesity-related diseases. A similar pattern of accumulation was found in humans, as post-menopausal women had significantly more visceral fat than pre-menopausal women (Ley et al. 1992; Janssen et al. 2010; Demerath et al. 2011). In intact female dogs, only the TA/BA showed a positive correlation with the age. Thus, the risk of obesity may increase with ageing in female dogs, consistent with a previous study in which the BCS increased steadily from young to senior female dogs (Bjornvad et al. 2019). In contrast, in spayed female dogs, the TA/BA did not show a correlation with the age, but the VA/SA showed a positive correlation with the age. This difference in the correlation between intact and spayed female dogs may be the result of the aforementioned neutering effect of increasing the VA/SA (Janssen et al. 2010; Demerath et al. 2011).

Between intact and neutered male dogs, the TA/BA and VA/SA were not different, in contrast to a recent study which revealed neutering in male dogs dramatically increased the BCS (Bjornvad et al. 2019).

As a previous study proved the age at neutering influences the post-neutering obesity (Spain et al. 2004), the age at neutering in the dogs in the present study might have affected the abdominal fat composition. Apart from that, this may be due to the small number of intact male dogs in the present study, thus further study with a more balanced number of male dogs is needed. Bjornvad et al. (2019) also found that the BCS in male dogs remained at the same level with ageing, and ageing in male dogs decreased the risk of obesity. In the present study, as the intact male dogs showed no significant correlation in the TA/BA or VA/SA with the age, the risk of obesity may not change with ageing in intact male dogs. However, in neutered male dogs, a positive correlation of the VA/SA and no significant correlation of the TA/BA with the age, may imply a tacit increase in the VA/SA. The re-allocation of abdominal fat from the subcutaneous to visceral compartments in spayed female and neutered male dogs may be a concern because of the decreased protective effects of the subcutaneous fat (Demerath et al. 2011). Therefore, careful monitoring of the VA/SA in these dogs against visceral obesity-related diseases may be required, even without an increase noted in the BCS.

There were some limitations of the present study. An imbalanced number among the four sexual statuses may have an effect on the results. The age at neutering, nutrition status, owner's age or owner's health status may have influenced the body composition of the dogs; however, these data were not obtained. The use of the deuterium oxide dilution method to compare and validate the fat mass assessed by CT could have strengthened the study with its accuracy for assessment of the body fat mass.

In conclusion, the breed, age, and sex all have a distinct effect upon the visceral and subcutaneous fat accumulation patterns; the ratio of the visceral to subcutaneous fat was relatively higher in Yorkshire Terriers than in Poodles, in older dogs than in younger dogs, and in spayed females than in intact females. The careful monitoring of the VA/SA in the breeds (Maltese, Shih-Tzus, and Yorkshire Terriers), age (senior dogs), and sexual statuses (neutered male and spayed female dogs) may be required.

This study presented descriptive data of the TA/BA and VA/SA according to the breed, age, and sexual status. The results may help future studies in identifying the breeds and sex predisposed to obesity-related diseases.

Conflict of interest

The authors declare no conflict of interest.

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