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Low depression scores among pet dog owners – a comparative cross-sectional study from

Anuradhapura Sri Lanka

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ABSTRACT

Introduction and aim. The role of canine companionship in psychosocial and cardiovascular health is

increasingly explored. However, such studies are scarce in South Asia. Hence, we aim to compare

psychosocial and cardiovascular indices between pet dog owners and age and sex-matched non-pet owners

in Anuradhapura, Sri Lanka.

Material and methods. A community-based, comparative, cross-sectional study was done among 52 pet

dog owners and 52 age, sex-matched non-pet owners. An allostatic load was calculated using predefined

cutoffs. Chi-square (or Fisher's exact), Mann Whitney U, and Spearman's rho were performed (p<0.05).

Results. The pet dog owners' group had a significantly lower mean (SD) [7.8 (7.2)], and median

(interquartile range) [6 (2–12.5)] depression score when compared to the non-pet owners [11.2 (8.6)], 10

(6–14.5) respectively (p=0.03). Also, the pet dog owners showed a significant negative correlation between

the pet bond scale score and cortisol (r=-0.36, p=0.01).

Conclusion. A significantly lower depression score in the pet dog owners' group than in the non-pet

owners' group was observed. And, the pet bond score had a significant negative correlation with the stress

hormone cortisol. Thus, pet dog ownership could improve psychosocial health and will guide towards one-

health interventions and research among South Asian communities.

Introduction

Humans and dogs have had an emotional bond for thousands of years. Canine companionship is identified as an integral part of a family.² Also, pet ownership contributes to a healthy neighbourhood relationship.³ Further, dog ownership contributes towards human health and well-being. 4 Variable results such as positive, mixed, negative, and no impact of pet ownership on mental health are reported in prior literature.⁵ Thus, an objective assessment of the association between dog ownership and mental well-being is warranted. Moreover, canine companionship is associated with reduced mortality possibly due to decreased cardiovascular mortality. Dog owners are more likely to achieve optimum cardiovascular health metrics. And, ownership is protective against coronary artery disease.⁴ Animal companionship regulates the autonomic responses in patients with non-communicable diseases. 8 Dog owners had a significantly higher chance of survival within one year after acute myocardial infarction than those who did not own dogs.9 Also, pet ownership reduces the risk of high blood pressure and improves the control of hypertension among owners. 10 Further, owners who regularly walked their dogs are significantly less likely to have self-reported diabetes, hypertension, hypercholesterolemia, and depression than those who did not own dogs. 11 Moreover, dog owners had higher high-density lipoprotein, lower cholesterol, lower triglycerides and lower low-density lipoprotein.^{7,12,13} The cardiovascular benefits of pet ownership are explained via many possible mechanisms including increased physical activity, reduced sympathetic response to stress, and improved physiological parameters, mental well-being and social interaction. And, the American Heart Association finds a probable association and a causal role of dog ownership with a decreased cardiovascular disease risk but, recommends against pet adoption, rescue, or purchase for the primary purpose of reducing cardiovascular disease risk. ¹⁴ The role of canine companionship in psychosocial and cardiovascular health is increasingly explored. However, such studies are scarce in South Asia.

Stress is identified as a pervasive risk factor for cardiovascular disease. ¹⁵ Work-related stress and social isolation have an increased risk of coronary heart disease. ¹⁶ Stress is associated with cardiovascular disease independent of familial background, history of somatic or psychiatric diseases, and psychiatric comorbidity. ¹⁷ Stress-related biomarkers like cortisol have been analysed to find an association between canine companionship and cardiovascular health. ^{18–20} A significant reduction of cortisol in canine companionship and dog-assisted therapy along with a significant reduction in heart rate, systolic blood pressure, mean arterial blood pressure or total cholesterol was reported in a systematic review. ²¹ However, the precise mechanism underlining the above phenomenon is unclear. A hypothesis states that "canine companionship reduces stress and could decrease: (A) neuropeptide Y (NPY) level, (B) cortisol level, and (C) NPY-induced potentiation of cortisol levels. The net effect would be a reduction in the end processes that contribute to cardiovascular disease". ²² A high level of cortisol is associated with hypertension,

diabetes, dyslipidaemia, and immune modulation.²³⁻²⁶ Also, body physique correlates with cortisol.²⁷ Further, NPY stimulates cortisol secretion and plays a key role in cardiovascular disease.^{28,29} NPY plays an important role in regulating emotional-affective behaviour, stress coping and feeding.³⁰ NPY is implicated in hypertension, arrhythmia, immune modulation, and dyslipidaemia.^{29,31} Hence, both cortisol and NPY regulations have an essential role in cardiovascular therapeutics.^{26,29} And, NPY is a prospect for the treatment of metabolic syndrome.³² Moreover, NPY regulates neuroprotection, restores bone marrow dysfunction, and regulates the composition of the bone marrow microenvironment.³³ Hence, the modulation of NPY has a significant role in various diseases.³⁴

Natural stressors are associated with an increased level of cortisol.³⁵ And, cortisol is a potential diagnostic biomarker of stress.³⁶ Therefore, the analysis of cortisol is helpful in stress-related assessments.³⁷ Also, cortisol has a positive association with incident cardiovascular disease. 38 Cortisol has an important role in the circadian rhythm and the regulation of cardiac function.³⁹ An Indian study showed a stronger association between cortisol and cardiovascular risk factors. 40 Further, a study from Scotland revealed cortisol excretion rate to be positively correlated with anthropometry.²⁷ Hence, cortisol regulation has a vital role in cardiovascular therapeutics.²⁶ The role of NPY is extensively explored for the treatment of metabolic syndrome.³² Therefore, the assessment of NPY has a pivotal role in research. Also, assessing stress and emotional-affective behaviour involves the measurement of biomarkers like NPY. Hence, the NPY levels among healthy individuals are essential in interpreting values related to disorders. A study from the United Kingdom (UK) reported a mean NPY of 55 pmol/l in 18 healthy controls by using specific radioimmunoassay to measure NPY concentrations. 41 Also, a study from the United States of America (USA) reported a mean (SD) plasma NPY of 79.8 (34.9) pmol/l among 100 sedentary healthy controls by using a competitive radioimmunoassay. 42 Further, a Chinese study reported a mean (SD) serum NPY of 478.89 (145.53) pg/mL among 71 metabolically healthy obese participants by using a specific enzymelinked immunosorbent assay (ELISA).⁴³ The interpretation of biomarkers like NPY and cortisol requires baseline levels for the region, which is deficient in prior research, especially for the South Asian region.

Aim

Initially, we aim to determine the measures of central tendency, variation for NPY and cortisol, and its relationship with socio-demographic factors, anthropometry, blood pressure, scale scores, and biochemical tests among healthy dwellers of Anuradhapura, Sri Lanka. Then the study intends to compare socio-demographic factors, anthropometry, blood pressure, psychosocial scale scores, NPY, cortisol, and other biochemical tests related to cardiovascular health between pet dog owners and age, sex-matched individuals who do not own a pet in Anuradhapura, Sri Lanka. Moreover, it intends to sort significant differences or correlations for the variables of interest with the pet bond score among pet dog owners.

Material and methods

Study design, setting, and population

We report a community-based, comparative, cross-sectional study (pet dog owners and non-pet owners) with laboratory analysis in the Anuradhapura district, Sri Lanka. Since baseline data on NPY and cortisol were not available in Sri Lankan literature, the present study was also designed to identify the natural variation for NPY and cortisol among healthy dwellers of Anuradhapura. Data collection was initiated in May 2022. The dwellers in the district during the study period were considered the study population. Anuradhapura is a predominantly rural, agrarian district of Sri Lanka. Anuradhapura is the largest district by surface area in Sri Lanka. However, the district's population density (per 131 km²) is much lower than the country's density (per 350 km²). 46,47 Fifty-seven per cent of households in Anuradhapura owned a pet, the dog was the most common pet owned by 41% of the households. And, the dog owners of Anuradhapura experienced companionship with their pets. 49

Sample size

Pet dog owners and non-pet owners

Previous data on NPY among dog and non-dog owners was unavailable in the literature. The data for serum cortisol levels among dog and non-dog owners were available in only a Mexican study and were used for sample size calculation. The following formula was used for sample size calculation: $n_B=(1+1/k)$ [$\sigma \times (Z_{1-\alpha/2}+Z_{1-\beta})/(\mu_A-\mu_B)$]. Where n_A and n_B are the calculated sample size for the non-pet owners' and pet dog owners' groups respectively, k is n_A/n_B (matching ratio) (=01), σ is the standard deviation (=4.73), $Z_{1-\alpha/2}$ is the type I error (=1.96), $Z_{1-\beta}$ is the power (=0.80), μ_A is the non-pet owners' group mean (=14.77) and μ_B is the pet dog owners' group *mean* (=12.12). Accordingly, the sample size was 50 for each group. Fifty-two individuals were recruited for each group while maintaining a male-to-female ratio of 1:1.

Healthy dwellers

Forty healthy individuals were recruited from the district. Ten participants were recruited from each of the following age groups while maintaining a male-to-female ratio of 1:1: 18 to \leq 30, 31 to \leq 40, 41 to \leq 50, and 51 to \leq 60 years.

Participants, selection criteria and sampling method

Pet dog owners and non-pet owners

A community-based convenience sampling was done to recruit suitable participants. The experience gained by the investigators during a previous project conducted in Anuradhapura, Sri Lanka, on the prevalence of pet ownership was used to recruit the participants.⁴⁸ Individuals who owned $\leq 3 \log(s)$ as the only pet for the last 1 year or more and had a pet bond score of >50 were included in the pet dog owners' group. And,

individuals who did not own any pets for the last 1 year or more were included in the non-pet owners' group. Participants aged ≥41 to ≤65 years, Sinhala Buddhist, permanently residing in Anuradhapura district for ≥5 years, having an estimated glomerular filtration rate ≥60 ml/min/1.73m² according to Chronic Kidney Disease Epidemiology Collaboration equation, and having a Body Mass Index of 18.5 to 29.9 kg/m² were included for both the groups. Individuals with the following were excluded from both groups: acute illness, on medications, history of chronic diseases (organic or psychiatric), history of long-term drug treatment, history of immunosuppression (steroid treatment or chemotherapy), pregnancy, everyday smoking, and heavy alcohol use.^{50,51} Age-matched non-pet owners were recruited by selecting participants with the same age ±2 years. A male-to-female ratio of 1:1 was maintained.

Healthy dwellers

A community-based convenience sampling was done to recruit suitable participants. Participants aged ≥ 18 to ≤ 60 years, Sinhala Buddhist, permanently residing in Anuradhapura district for ≥ 5 years, having an estimated glomerular filtration rate (eGFR) ≥ 60 ml/min/1.73m² according to Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation, and having a body mass index (BMI) of 18.5 to 29.9 kg/m² were included. Individuals with the following were excluded: acute illness, on medications, history of chronic diseases (organic or psychiatric), history of long-term drug treatment, history of immunosuppression (steroid treatment or chemotherapy), pregnancy, everyday smoking, and heavy alcohol use. 50,51

Study instruments

The questionnaire on (i) socio-demographic factors was interviewer-administered by the first author while the (ii) Depression Anxiety Stress Scale-21 (DASS-21) translated into the Sinhala language and (iii) Multi-dimensional Scale of Perceived Social Support (MSPSS) translated into the Sinhala language were self-administered by the participants with instructions from the first author. ⁵²⁻⁵⁵ Also, the (iv) pet bond scale was self-administered among pet dog owners in the Sinhala language. ^{56,57} The following socio-demographic factors were considered: years residing in Anuradhapura, age, sex, whether the participant was the head of household, education level, employment, marital status, whether the participant was living alone, number of adults and children at home (males and females), sector, household income, alcohol consumption, smoking, betel chewing, and sleeping hours per day. The face validity of the questionnaire on socio-demographic factors was established. Subsequently, the questionnaire on socio-demographic factors and the pet bond scale were pre-tested in 05 subjects to improve language and sequence. The content validity of the pet bond scale was acceptable as it is a 25-item 5-point Likert scale ranging from 0 (strongly disagree) to 4 (strongly agree) which assesses the bonding between the owner and pet. ⁵⁶ A total score of 0 to 100 can

be achieved on the pet bond scale. The corresponding author has provided permission to translate and use the pet bond scale.⁵⁶

Anthropometric measurements of weight, height, waist circumference and hip circumference were measured using a standard digital bathroom scale (SECA robusta 813 CE), stadiometer (SECA 213 CE 0123) and measuring tape respectively. Two blood pressure measurements within a minute interval were measured using a standard aneroid desk model sphygmomanometer (ACCOSON CE 0413) after a 5-minute rest in the seated position. The average of the two measurements was used.

A blood sample of 6 mL for fasting blood sugar, lipid profile, serum creatinine, c-reactive protein, NPY and cortisol was obtained at 9 am following overnight fasting of 12 hours. Blood samples for fasting blood sugar, lipid profile, serum creatinine, and c-reactive protein were analysed at the Durdans Hospital laboratory, Anuradhapura.⁵⁸ It is a Joint Commission International-accredited hospital in Sri Lanka. Procedures for measurement of the above investigations were well established and routinely done at the above laboratory. The method used for the analysis of fasting blood sugar, total cholesterol, triglyceride, high-density lipoprotein (HDL) and serum creatinine is enzymatic colorimetric assay; for low-density lipoprotein (LDL), if triglyceride is ≥400 mg/dL, is enzymatic end-point; for c-reactive protein is turbidimetric method. Quality control for fasting blood sugar, total cholesterol, triglyceride and serum creatinine was done using Bio-Rad lyphochek assay chemistry control; for HDL and LDL using Gernorm and Gerpath Quality control; for c-reactive protein using Diagam Protein C Reactive High and Low Control. eGFR was calculated according to the CKD-EPI equation. Duplicate measurements of NPY and cortisol were analysed using ELISA at the Department of Animal and Food Sciences, Faculty of Agriculture, Rajarata University of Sri Lanka. For NPY, ELISA kits and quality controls from Elabscience ®, Elabscience Biotechnology Inc., USA were used (E-EL-H1893).⁵⁹ For cortisol, ELISA kits and quality controls from Abia Cortisol, AB Diagnostic Systems, Germany were used (DK.038.01.3). 60 ELISA reading was done using the Thermo Scientific Spectrophotometer Multiskan Sky, Singapore (51119700DP).

Data collection

Ethical clearance was obtained from the Ethics Review Committee of the Faculty of Medicine and Allied Sciences, Rajarata University of Sri Lanka (ERC/2020/76). Informed written consent was obtained from all participants. Prior permission was obtained from the Regional Director of Health Services, Anuradhapura. Further, all Medical Officers of Health in the Anuradhapura district were informed by the Regional Director of Health Services to facilitate data collection in their areas. Recruited participants, were invited to the Durdans Hospital laboratory, Anuradhapura for data and sample collection. The first author was involved in the following: describing the study, obtaining informed written consent, collecting data, and collecting anthropometric measurements. All necessary measures were taken to preserve the participant's privacy and confidentiality. The process lasted for about 30 minutes.

Data description and analysis

Collected data were entered into a Microsoft Excel spreadsheet. Mean with SD was reported for continuous variables and frequency with percentages for categorical variables. Also, the World Health Organization/ International Society of Hypertension (WHO/ISH) risk prediction chart for Southeastern Asian Region B was used to predict the 10-year risk of a fatal or non-fatal cardiovascular event.⁶¹ Further, the following were used to calculate the allostatic load: (A) For the cardiovascular system, systolic blood pressure, diastolic blood pressure, mean arterial blood pressure, total cholesterol, HDL, triglyceride, and LDL; (B) For the metabolic system, body mass index, waist circumference, waist-to-hip ratio, fasting blood sugar, and estimated glomerular filtration rate; (C) For the immune system, c-reactive protein; (D) For the neuroendocrine system, NPY and cortisol. Cut-off values for the parameters used to calculate the allostatic overload are shown in Table S1. Scores for depression, anxiety and stress in DASS-21 were calculated by summing the scores for the relevant items. Scores on the DASS-21 were multiplied by 2 to calculate the final score.⁵² A total score for Perceived Social Support was calculated by adding all values across the twelve items and subsequently, a mean score was obtained. Moreover, separate scores for social support from a significant other, family and friends were analysed. The analysis was performed using Microsoft Excel and add-ins. Continuous variables with a p<0.2 were included in a multivariable model and a backward stepwise multiple linear regression was used to identify continuous variables with a p<0.05. The stopping rule was satisfied when all remaining variables in the model had a p<0.05. The Cronbach's alpha of questions representing pet bond scale, depression, anxiety, and stress of DASS-21 and social support from a significant other, family and friends of MSPSS was derived for pet dog owners' group, non-pet owners' group and healthy dwellers group.

Pet dog owners and non-pet owners

The chi-square test (or Fisher's exact) and Mann-Whitney U test were performed to determine a significant difference in the proportions and distributions of variables (p<0.05). Spearman's rho was sorted for the pet bond score against the continuous variables of interest (p<0.05).

Healthy dwellers

Measures of the central tendency of NPY and cortisol were described. Mann Whitney U test was performed to determine a significant difference in the distribution of NPY and cortisol between the two groups of the categorical variables of interest (p<0.05). Spearman's rho was sorted for NPY and cortisol against the continuous variables of interest (p<0.05). Kruskal-Wallis test (and when significant post hoc Dunn's multiple comparisons test) was performed to determine significant differences between the age groups in relation to the NPY and cortisol levels (p<0.05).

Ethics approval

Ethical clearance was obtained from the Ethics Review Committee of the Faculty of Medicine and Allied Sciences, Rajarata University of Sri Lanka (ERC/2020/76). Informed written consent was obtained from all participants. The study was performed in accordance with the Declaration of Helsinki. All methods were carried out in accordance with relevant guidelines and regulations. All necessary measures were taken to preserve the participant's privacy and confidentiality.

Results

Demographic data

Pet dog owners and non-pet owners

Fifty-two participants (male: female=1:1) were recruited each for the pet dog owners' and non-pet owners' groups. Most of the participants have completed the general certificate of education (advanced level; the highest level of examination sat within the Sri Lankan high-school education system) or above (pet dog owners=71%, non-pet owners=69%), were employed (pet dog owners=79%, non-pet owners=92%), currently married (pet dog owners=98%, non-pet owners=90%), not living alone (pet dog owners=100%, non-pet owners=98%), had >Rs 100,000 monthly household income (pet dog owners=69%, non-pet owners=56%), never consumed alcohol (pet dog owners=56%, non-pet owners=67%), never smoked (pet dog owners=92%, non-pet owners=88%), never chewed betel (pet dog owners=92%, non-pet owners=94%), and had <10% 10-year risk of a fatal or non-fatal cardiovascular event (pet dog owners=98%, non-pet owners=100%). The Cronbach's alpha of questions representing pet bond scale, depression, anxiety, and stress of DASS-21 and social support from a significant other, family and friends of MSPSS in the pet dog owners' group were 0.91, 0.78, 0.76, 0.83, 0.82, 0.88, 0.92 respectively, indicating good internal consistency in the responses. The Cronbach's alpha of questions representing depression, anxiety, and stress of DASS-21 and social support from a significant other, family and friends of MSPSS in the non-pet owners' group were 0.81, 0.79, 0.80, 0.65, 0.86, 0.79 respectively, indicating good internal consistency in the responses.

Healthy dwellers

Most of the study participants were non-head of household (70% - 28/40), completed the general certificate of education (advanced level; the highest level of examination sat within the Sri Lankan high-school education system) or above (75% - 30/40), employed (90% - 36/40), currently married (73% - 29/40), not living alone at the household (100% - 40/40), rural residents (53% - 21/40), and having a monthly household income of more than 100,000 Sri Lankan rupees (53% - 21/40). Also, most had ≥ 2 adult males in the household (53% - 21/40), ≥ 2 adult females in the household (58% - 23/40), no male children in the

household (65% - 26/40), no female children in the household (60% - 24/40), never consumed alcohol (68% - 27/40), never smoked (83% - 33/40), and never chewed betel (95% - 38/40). All participants above the age of 40 years had <10% 10–year risk of a fatal or non–fatal cardiovascular event according to the World Health Organization/ International Society of Hypertension (WHO/ISH) risk prediction chart for Southeastern Asian Region B. The Cronbach's alpha of questions representing depression, anxiety, and stress of DASS–21 and social support from a significant other, family and friends of MSPSS in the healthy dwellers were 0.90, 0.84, 0.85, 0.78, 0.82, and 0.86 respectively, indicating good internal consistency in the responses.

Comparison of the variables of interest among pet dog owners and non-pet owners

The categorical and continuous variables of the pet dog owners and non-pet owners' groups are compared in Table S2 and Table 1 respectively. Proportions related to the sector, being the head of household, education level, employment, marital status, living alone, number of adults and children at home (males and females), monthly household income, alcohol consumption, smoking, betel chewing, and 10-year risk of a fatal or non-fatal cardiovascular event were not significantly different between the two groups.

Table 1. Comparison of continuous variables between the pet dog owners' group and the non-pet owners' group of the biochemical tests

No	Item	Pet dog owners	Non-pet owners'	p of Mann Whitney U	
		(n=52)	(n=52)		
	G	Mean (SD) and	Mean (SD) and	test	
		median	median		
		(Interquartile	(Interquartile		
		range)	range)		
1	Age (years)	46 (5)	46 (6.1)	0.50	
		46 (42.8–48)	44 (42–48)		
2	Duration of stay at Anuradhapura	35.9 (13.3)	36.7 (15.3)	0.77	
	(years)	40.0 (22.8–46)	41.5 (26–45.3)		
3	Hours of sleep per day	6.9 (1)	6.9 (1.1)	0.85	
		7 (6–8)	7 (6–8)		
4	Weight (kg)	66.9 (9)	64.1 (10.8)	0.20	
		64.6 (60.8–71.8)	62.6 (56.7–72.8)		
5	Height (cm)	163 (9.3)	161 (8.8)	0.90	
		163.8 (155.8–168.9)	160.5 (155–166.6)		
6	BMI (kg/m ²)	25.2 (2.8)	24.7 (3.3)	0.57	

		24.9 (22.9–27.4)	25.1 (22.4–27)	
7	Waist circumference (cm)	82.6 (6.9)	82.1 (7.8)	0.97
		82.0 (77.5–87.3)	82.5 (75–89)	
8	Hip circumference (cm)	97.5 (6.4)	96.4 (7.5)	0.62
		96.5 (93–101.3)	97.0 (92–101)	
9	Waist/Hip ratio	0.9 (0)	0.9 (0.1)	0.40
		0.9 (0.8–0.9)	0.9 (0.8–0.9)	2
10	Heart rate (per minute)	67 (4.6)	67.4 (6.9)	0.56
		66.0 (64.0–69.0)	65 (62–70)	17
11	Systolic blood pressure (mmHg)	118.7 (5.7)	117.8 (8)	0.78
		119.0 (115.5–123)	119.5 (111–122.3)	
12	Diastolic blood pressure (mmHg)	75.5 (4.8)	75.2 (5.8)	0.71
		75.0 (71–79)	73.0 (71–79)	
13	Mean arterial blood pressure	89.9 (4.5)	89.4 (6)	0.57
	(mmHg)	89.7 (85.9–94.1)	89.2 (86.5–92.3)	
14	Depression score (Depression	7.8 (7.2)	11.2 (8.6)	0.03
	Anxiety Stress Scale-21)	6 (2–12.5)	10.0 (6–14.5)	
15	Anxiety score (Depression Anxiety	7 (6.7)	8.5 (7.5)	0.23
	Stress Scale-21)	4 (2–12)	6 (10–14.5)	
16	Stress score (Depression Anxiety	13.8 (8.9)	15.5 (8.6)	0.26
	Stress Scale-21)	12 (8–20)	14 (10–20)	
17	Total score for perceived social	5.2 (0.8)	5 (0.5)	0.14
	support (Multi-dimensional Scale	5.1 (4.7–5.6)	4.9 (4.7–5.2)	
	of Perceived Social Support)			
18	Perceived social support from the	5.3 (0.9)	5.1 (0.8)	0.20
	significant other (Multi-	5.1 (4.8–6)	5.0 (4.8–5.5)	
	dimensional Scale of Perceived			
	Social Support)			
19	Perceived social support from the	5.4 (1)	5.2 (0.9)	0.10
	family (Multi-dimensional Scale of	5.3 (4.9–6)	5.0 (4.5–5.8)	
	Perceived Social Support)			
20	Perceived social support from	4.9 (0.9)	4.8 (0.7)	0.42
	friends (Multi-dimensional Scale	4.8 (4.4–5.3)	4.8 (4.5–5)	
	of Perceived Social Support)			

21	Fasting blood sugar (mg/dL)	95.0 (16.1)	94.9 (12.9)	0.52
		91.5 (85.0–97.5)	93.5 (86.0–99.3)	
22	C-reactive protein (mg/l)	0.7 (1.2)	1.1 (2.5)	0.82
		0.1 (0.1–0.6)	0.1 (0.1–0.3)	
23	Total cholesterol (mg/dL)	200.2 (32.9)	203.8 (3.7)	0.80
		205.5 (176–220.3)	204.5 (177–223)	
24	HDL (mg/dL)	38.4 (7.8)	40.2 (7.6)	0.13
		36.0 (32–42)	38.0 (34.8–45.3)	K C
25	Triglyceride (mg/dL)	170.0 (79.7)	164.1 (87.1)	0.42
		147.0 (115–192)	138.0 (97–214.3)	
26	LDL (mg/dL)	126.7 (31.5) 125.1	132.7 (27.9) 131.7	0.35
		(107.9–148)	(112.2–152.9)	
27	Very-low-density lipoprotein	33.3 (13.8)	30.9 (13.5) 27.6	0.32
	(mg/dL)	29.4 (23–38.4)	(19.4–40)	
28	Total cholesterol/HDL	5.3 (1)	5.2 (1.1)	0.49
		5.5 (4.6–6.1)	5.1 (4.5–6)	
29	Triglyceride/HDL	4.6 (2.4)	4.3 (2.5)	0.24
		4.4 (3–5.4)	3.4 (2.4–5.5)	
30	Serum creatinine (mg/dL)	0.8 (0.2)	0.8 (0.2)	1.00
		0.8 (0.6–0.9)	0.8 (0.6–0.9)	
31	Estimated glomerular filtration rate	103.3 (13.8)	100.1 (13.8)	0.20
	(ml/min/1.73m ²)	105.0 (95–112.3)	100.5 (91–110)	
32	NPY (pg/mL)	388.2 (225.4)	408 (215)	0.61
		341.1 (223–512.8)	347.6 (211.2–592.1)	
33	Cortisol (nmol/L)	234.4 (73.1)	234.8 (122.2)	0.31
		235.3 (188.9–292.2)	193.0 (149.1–286.9)	
34	Allostatic load	2.4 (1.4)	2.4 (1.5)	0.96
		2 (1–3)	2 (1–3)	

The pet dog owners' group had a significantly lower *mean* (SD) [7.8 (7.2)], and median (interquartile range) [6 (2–12.5)] depression score in DASS-21 when compared to the non-pet owners' group [11.2 (8.6)], 10 (6–14.5) respectively (p=0.03). However, the distributions were not significantly different in the two groups for age, duration of stay at Anuradhapura, hours of sleep per day, weight, height, body mass index, waist

circumference, hip circumference, waist/hip ratio, heart rate, systolic blood pressure, diastolic blood pressure, mean arterial blood pressure, DASS-21 scores for anxiety and stress, MSPSS scores (total, significant other, family, friend), fasting blood sugar, c-reactive protein, total cholesterol, HDL, triglyceride, LDL, very-low-density lipoprotein, total cholesterol/HDL ratio, triglyceride/HDL ratio, serum creatinine, estimated glomerular filtration rate, and allostatic load.

The pet dog owners' group had a mean (SD) of 388.2 (225.4) pg/mL, and a median (interquartile range) of 341.1 (223–512.8) pg/mL for NPY when compared to 408 (215) pg/mL and 347.6 (211.2–592.1) pg/mL for the non-pet owners' group. However, the difference was not significant (p=0.61). Further, the pet dog owners' group had a mean (SD) of 234.4 (73.1) nmol/L, and a median (interquartile range) of 235.3 (188.9–292.2) nmol/L of cortisol when compared to 234.8 (122.2) nmol/L and 193 (149.1–286.9) nmol/L for the non-pet owners' group. However, the difference was not significant (p=0.31).

Pet bond score and the variables of interest among pet dog owners

The mean pet bond score of the pet dog owners' group was 68.4 (11.2) with a median (interquartile range) of 66 (60.8–79.3). The distribution of the pet bond score was not significantly different in relation to sex, sector, whether the participant was the head of household, education level, employment, number of children at home (males and females), monthly household income, alcohol consumption, and number of pet dogs at home. Table S3 summarises the p-values for the categorical variables of interest against the pet bond score. The categorical variables of the high (scores 70 to 100) and low (scores 50 to 69) pet bond groups are compared in Table 2. The high pet bond group had a significantly higher percentage of females (67%) when compared to the low pet bond group (39%) (p=0.047). The high pet bond group had a significantly higher percentage of participants having a monthly household income of more than Rs 100,000 (86%) when compared to the low pet bond group (58%) (p=0.03). However, proportions related to the sector, being the head of household, education level, marital status, number of children at home (males and females), alcohol consumption, smoking, betel chewing, the 10-year risk of a fatal or non-fatal cardiovascular event, and number of pet dogs at home were not significantly different between the high and low pet bond groups. Cortisol (r=-0.36, p=0.01), perceived social support from a significant other (r=0.34, p=0.01), and height (r=-0.31, p=0.02) were significantly correlated with the pet bond score. A backward stepwise regression found only the cortisol to have a p<0.05 (Figure 1). The R² of the single variable regression was 0.13 with the predictive model, pet bond score; + 81.43 - 0.06 (cortisol). However, age, duration of stay at Anuradhapura, hours of sleep per day, weight, body mass index, waist circumference, hip circumference, waist/hip ratio, heart rate, systolic blood pressure, diastolic blood pressure, mean arterial blood pressure, DASS-21 scores for depression, anxiety and stress, MSPSS score (total, family, friend), fasting blood sugar, c-reactive protein, total cholesterol, HDL, triglyceride, LDL, very-low-density lipoprotein, total cholesterol / HDL ratio, triglyceride / HDL ratio, serum creatinine, estimated glomerular filtration rate, NPY, and

allostatic load did not have a significant correlation with the pet bond score. Table 3 summarises the p-values for the continuous variables of interest against the pet bond score.

Table 2. Comparison of categorical variables between the high pet bond score group and the low pet bond score group among the pet dog owners^a

No	Variable	Description	Percentage in	Percentage in	р
			the high pet	the low pet	2
			bond group	bond group	
			(Score 70 to	(Score 50 to	
			100)	69)	
			(n=21)	(n=31)	
1	Gender	Male	33	61	<0.05*
		Female	67	39	
2	Sector	Rural	33	52	0.19*
		Urban	67	48	
3	Head of the household	Yes	33	58	0.08*
		No	67	42	
4	Education	Completed or	81	65	0.20*
		above the			
		general			
		certificate of			
		education			
	\sim	(advanced level;			
		the highest level			
		of examination			
		sat within the			
		Sri Lankan			
		high-school			
		education			
		system)			
		Below or up to	19	35	
		general			
		certificate of			
		education			
		(advanced level)			

5	Employment	Employed	86	74	0.52#
		Unemployed	14	26	
6	Marital status	Currently	95	100	0.80#
		married			
		Never married	5	0	
		or separated			
7	Living alone	Yes	0	0	Not
					performed
		No	100	100	17
8	No of adult males (≥12	≥1	100	100	Not
	years) at home			4	performed
		None	0	0	
9	No of adult females (≥12	≥1	100	100	Not
	years) at home		~^^		performed
		None	0	0	
10	No of male children (<12	≥1	19	32	0.29*
	years) at home				
		None	81	68	
11	No of female children (<12	≥1	57	32	0.08*
	years) at home				
		None	43	68	
12	Monthly household	>100,000	86	58	0.03*
	income (Rs)				
		≤100,000	14	42	
13	Alcohol consumption	Never	67	48	0.19*
		Moderation	33	52	
14	Smoking	Never	100	87	0.23#
		Somedays or	0	13	
)	former			
15	Betel chewing	Never	91	94	0.99#
		Somedays or	9	6	
		former			
16	10-year risk of a fatal or	<10%	100	97	0.99#
	non-fatal cardiovascular				

event according to the
WHO/ISH risk prediction
chart for the Southeastern
Asian region B

	10% to <20%	0	3	
17 No of pet dogs	>1	38	23	0.23*
	=1	62	77	

^a* – Chi-square test was performed, [#] – Fisher's exact was performed

Table 3. Continuous variables of interest against the pet bond score among pet dog owners

No	Variables	Spearman's rho	p
1	Age (years)	0.18	0.20
2	Duration of stay at Anuradhapura (years)	-0.04	0.77
3	Hours of sleep per day	-0.10	0.50
4	Weight (kg)	-0.20	0.16
5	Height (cm)	-0.31	0.02
6	BMI (kg/m²)	0.09	0.54
7	Waist circumference (cm)	0	0.98
8	Hip circumference (cm)	0.12	0.42
9	Waist/Hip ratio	-0.06	0.67
10	Heart rate (per minute)	-0.06	0.66
11	Systolic blood pressure (mmHg)	0.02	0.91
12	Diastolic blood pressure (mmHg)	-0.08	0.56
13	Mean arterial blood pressure (mmHg)	-0.07	0.61
14	Depression Anxiety Stress Scale-21 – depression score	0.05	0.75
15	Depression Anxiety Stress Scale-21 – anxiety score	0.07	0.63
16	Depression Anxiety Stress Scale-21 – stress score	-0.06	0.67
17	Multi-dimensional Scale of Perceived Social	0.18	0.20
	Support – total score		
18	Multi-dimensional Scale of Perceived Social	0.34	0.01
	Support – score for significant other		
19	Multi-dimensional Scale of Perceived Social	0.16	0.26
	Support – score for family		

20	Multi-dimensional Scale of Perceived Social	0.16	0.26
	Support – score friend		
21	Fasting blood sugar (mg/dL)	-0.08	0.58
22	C-reactive protein (mg/l)	-0.19	0.19
23	Total cholesterol (mg/dL)	-0.10	0.50
24	HDL (mg/dL)	0.10	0.50
25	Triglyceride (mg/dL)	-0.13	0.38
26	LDL (mg/dL)	-0.13	0.38
27	Very-low-density lipoprotein (mg/dL)	-0.12	0.39
28	Total cholesterol/High-density lipoprotein	-0.13	0.34
29	Triglyceride/High-density lipoprotein	-0.12	0.39
30	Serum creatinine (mg/dL)	-0.10	0.50
31	Estimated glomerular filtration rate (ml/min/1.73m ²)	-0.22	0.12
32	Cortisol (nmol/L)	-0.36	0.01
33	Neuropeptide Y (pg/mL)	0.10	0.50
34	Allostatic load	0.10	0.48

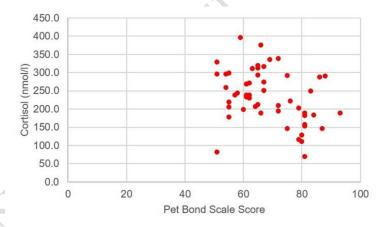


Fig. 1. Cortisol against pet bond scale score among pet dog owners (r=-0.36, p=0.01)

Cortisol and its association with the variables of interest among healthy dwellers

The mean (SD) of cortisol was 317.3 (132.5) nmol/L with a lower and upper 95% CI of 274.9 and 359.7 respectively. The cortisol ranged from 153.9 to 713.7 nmol/L. The median was 275.4 nmol/L with an interquartile range of 217.5 to 362.7. Skewness and kurtosis for the distribution were + 1.14 and + 0.95 respectively showing asymmetrical and potentially mesokurtic shape. The post hoc power was 0.99 for the distribution of cortisol. The following parameters were used to calculate the power: study mean (317.3 nmol/L), mean (SD) from previous literature among healthy controls [381.9 (97.7) nmol/L]⁶², sample size

(40) and type I error (0.05). Participants who were never married had a significantly higher mean (SD) [426.8 (167.9) nmol/L], and median (390.4 nmol/L) of cortisol when compared to those who were married [275.8 (88.7) nmol/L], (272.8 nmol/L) respectively (p=0.01). Participants who consumed alcohol in moderation had a significantly higher mean (SD) [381.8 (133.7) nmol/L], and median (361.5 nmol/L) of cortisol when compared to those who never consumed alcohol [286.2 (122.4) nmol/L], (272.8 nmol/L) respectively (p=0.04). However, the distribution of cortisol was not significantly different between the two groups related to sex, sector, whether the participant was the head of a household, education level, number of adults and children at home (males and females), and household income. Table 4 summarises the p-values for the categorical variables of interest against cortisol.

Table 4. Categorical variables of interest against NPY and cortisol among healthy dwellers

N	Variable	Groups	n (%)	NPY	_	Cortis	ol
0			(Total=40)	Mean (SD)	p of	Mean (SD)	p of
				and median	Mann	and median	Mann
				(Interquartile	Whitney	(Interquartile	Whitney
				range)	U test	range)	U test
				pg/mL		nmol/L	
1	Sex	Male	20 (50)	434.9 (243.3)	0.96	334.1 (132.9)	0.41
				361.3 (226.4–		299.3 (247.7–	
				610.7)		395.8)	
			5				
		Female	20 (50)	428 (221.6)		300.5 (133.3)	
				428.1 (280.8–		275.4 (210.8–	
				577.5)		328.8)	
2	Sector	Rural	21 (53)	417.8 (219.5)	0.72	331.7 (153.9)	0.87
				385.5 (227–		275.9 (218.5–	
				580.6)		370.4)	
		Urban	19 (47)	446.5 (245.7)		301.3 (105.8)	
				431.4 (252.6–		274.9 (225.9–	
				661.9)		353.7)	
3	Head of	No	28 (70)	436.1 (222.5)	0.79	337.1 (142.5)	0.14
	household			428.1 (255.1–		320.7 (237.1–	
				628.1)		375.4)	
		Yes	12 (30)	420.6 (255.7)		271.1 (95)	

				313.6 (223.1–		269.4 (212.8–	
				601.8)		273.7)	
4	Education	completed	30 (75)	395 (222.9)	0.08	325 (122.5)	0.33
	level	the general		348.5 (225.1–		296.8 (247.8–	
		certificate		567.9)		365.0)	
		of					
		education					2
		(advanced				(
		level) or					
		above					
		up to or	10 (25)	540.8 (225.5)		294.2 (164)	
		below the		578.3 (360.6–	_	262.4 (184.4–	
		general		674.4)		309.7)	
		certificate			1	Y	
		of					
		education					
		(advanced		2 3			
		level)					
5	Employed	Yes	36 (90)	422.1 (238.6)	Test not	322.5 (137.6)	Test not
				348.5 (226.4–	done	274.0 (217.5–	done
			5	610.7)		367.3)	
		No	4 (10)				
		110	4 (10)	515.2 (107.4)		270.3 (62.5)	
		110	4 (10)	515.2 (107.4) 493.9 (429.7–		270.3 (62.5) 289.8 (252.1–	
		10	4 (10)				
6	Marital	Currently	29 (73)	493.9 (429.7–	0.32	289.8 (252.1–	0.01
6	Marital status			493.9 (429.7– 579.3)	0.32	289.8 (252.1– 307.9)	0.01
6		Currently		493.9 (429.7– 579.3) 412.1 (237.0)	0.32	289.8 (252.1– 307.9) 275.8 (88.7)	0.01
6		Currently		493.9 (429.7– 579.3) 412.1 (237.0) 340.9 (224.4–	0.32	289.8 (252.1– 307.9) 275.8 (88.7) 272.8 (200.0–	0.01
6		Currently married	29 (73)	493.9 (429.7– 579.3) 412.1 (237.0) 340.9 (224.4– 600.2)	0.32	289.8 (252.1– 307.9) 275.8 (88.7) 272.8 (200.0– 320.9)	0.01
6		Currently married Never	29 (73)	493.9 (429.7– 579.3) 412.1 (237.0) 340.9 (224.4– 600.2) 482.5 (211)	0.32	289.8 (252.1– 307.9) 275.8 (88.7) 272.8 (200.0– 320.9) 426.8 (167.9)	0.01
		Currently married Never	29 (73)	493.9 (429.7– 579.3) 412.1 (237.0) 340.9 (224.4– 600.2) 482.5 (211) 518.2 (342.2–	0.32	289.8 (252.1– 307.9) 275.8 (88.7) 272.8 (200.0– 320.9) 426.8 (167.9) 390.4 (297.2–	0.01
	status	Currently married Never married	29 (73)	493.9 (429.7– 579.3) 412.1 (237.0) 340.9 (224.4– 600.2) 482.5 (211) 518.2 (342.2– 653.5)		289.8 (252.1– 307.9) 275.8 (88.7) 272.8 (200.0– 320.9) 426.8 (167.9) 390.4 (297.2– 551.3)	
7	Status No of adult	Currently married Never married	29 (73)	493.9 (429.7– 579.3) 412.1 (237.0) 340.9 (224.4– 600.2) 482.5 (211) 518.2 (342.2– 653.5) 413.7 (207.7)		289.8 (252.1– 307.9) 275.8 (88.7) 272.8 (200.0– 320.9) 426.8 (167.9) 390.4 (297.2– 551.3) 329.5 (135.7)	

				356.0 (270.2–		271.5 (248.2–	
				644.8)		336.5)	
8	No of adult	≥2	23 (58)	412 (207.9)	0.58	332.8 (142.3)	0.46
	females			385.5 (256.5-		275.9 (231.6–	
	(≥12 years)			543.1)		378.3)	
		<2	17 (42)	457.7 (260.6)		296.3 (118.8)	
				424.8 (247.5–		273.0 (195.5–	
				687.2)		352.5)	
9	No of male	None	26 (65)	441.4 (229.2)	0.63	318.1 (126.7)	0.88
	children			408.4 (250–		296.8 (225–	
	(<12 years)			619.5)		363.4)	
		≥1	14 (35)	413.0 (238.1)		315.8 (147.6)	
				376.6 (240.9–		275.4 (228–	
				542.7)		347)	
10	No of	None	24 (60)	472.5 (219.1)	0.14	314.5 (137.1)	0.86
	female			497.7 (324.9–		275.4 (198.9–	
	children			630)		375.4)	
	(<12 years)						
		≥1	16 (40)	369.9 (238.4)		321.5 (129.5)	
				205.5 (286.5–		288.2 (264.8–	
			5	549)		356.5)	
11	Monthly	>Rs	21 (53)	428.9 (212.8)	0.98	313.7 (134)	1.00
	household	100,000	<i>y</i>	424.8 (247.5–		303.6 (195.5–	
	income			623.9)		361.5)	
		≤Rs	19 (47)	434.3 (253)		321.2 (134.3)	
		100,000		356 (257.7–		272.8 (254.6–	
				603.3)		343.6)	
12	Alcohol	Never	27 (68)	389.8 (227)	0.10	286.2 (122.4)	0.04
	consumption			340.9 (186.5–		272.8 (197.8–	
				537.3)		323.2)	
		Moderation	13 (32)	517.9 (218.6)		381.8 (133.7)	
				580.6 (290.2-		361.5 (271.5–	
				623.9)		519.7)	

13	Smoking	Never	33 (83)	433.5 (237.8)	Test not	313 (133.5)	Test not
				424.8 (247.5–	done	275.9 (214.4–	done
				623.9)		361.5)	
		Former and	7 (17)	559.8 (205.6)		337.7 (135.9)	
		somedays		337 (258.6–		273 (269.4–	
				565.0)		403.4)	A
14	Betel	Never	38 (95)	418.5 (227.8)	Test not	321.7 (134.2)	Test not
	chewing			370.7 (232.1–	done	282.9 (225–	done
				599.9)		365)	
		Somedays	2 (5)	677.1 (108.8)		234.3 (54.8)	,
				677.1 (638.7–		234.3 (214.9–	
				715.5)		253.6)	

Age (r=-0.38, p=0.02), hours of sleep per day (r=0.34, p=0.03) and DASS-21 scores for stress (r=0.33, p=0.04) were significantly correlated with cortisol (Fig. 2). A backward stepwise regression found age, height, and stress to have a p<0.05. The R² of the three variable regression was 0.38 with the predictive model, cortisol level; - 452.84 – 3.39 (age) + 5.08 (height) + 4.98 (stress). Spearman's rho for the following combinations was performed: age vs. hours of sleep, age vs. stress score, and hours of sleep vs. stress score. Out of the above combinations, age had a significant negative correlation with stress score (r=-0.37, p=0.02). Also, cortisol levels showed a significant difference between the age groups of 18 to \leq 30 (median 380.4 nmol/L), 31 to \leq 40 (median 285.5 nmol/L), 41 to \leq 50 (median 273.2 nmol/L), and 51 to \leq 60 (median 245.7 nmol/L) years (p=0.04). The significant pair was 18 to \leq 30 vs 51 to \leq 60 years. However, duration of stay at Anuradhapura, weight, height, BMI, waist circumference, hip circumference, waist/hip ratio, systolic blood pressure, diastolic blood pressure, mean arterial blood pressure, DASS-21 scores for depression and anxiety, MSPSS score (total, significant other, family, friend), fasting blood sugar, c-reactive protein, total cholesterol, HDL, triglyceride, LDL, very-low-density lipoprotein, total cholesterol/HDL ratio, serum creatinine, estimated glomerular filtration rate, NPY and allostatic load did not have a significant correlation with cortisol. Table 5 summarises the p-values for the continuous variables of interest against cortisol.

Table 5. Continuous variables of interest against NPY and cortisol among healthy dwellers

No	Variable	Mean	NPY		Cortisol	
		(SD)	Spearman's	p-value	Spearman's	p-value
			rho		rho	
1	Age (years)	39.5	-0.12	0.46	-0.38	0.02
		(11.8)				

2	Duration of stay at	33.0	-0.07	0.69	-0.09	0.59
	Anuradhapura (years)	(13.6)				
3	Hours of sleep per day	7.2 (1.2)	0.28	0.08	0.34	0.03
4	Weight (kg)	62.6	0	0.99	0.04	0.79
		(11.0)				
5	Height (cm)	163.7	0.11	0.49	0.27	0.09
		(10.2)				2
6	BMI (kg/m ²)	23.3 (3.0)	-0.12	0.47	-0.19	0.25
7	Waist circumference	79.8 (9.0)	-0.10	0.54	-0.24	0.14
	(cm)					·
8	Hip circumference (cm)	95.9 (6.5)	-0.07	0.67	-0.06	0.72
9	Waist/Hip ratio	0.8 (0.1)	0.01	0.96	-0.16	0.34
10	Systolic blood pressure	113.6	-0.10	0.56	-0.11	0.51
	(mmHg)	(10.1)			<i>y</i>	
11	Diastolic blood pressure	74.4 (7.9)	-0.23	0.15	0.08	0.65
	(mmHg)					
12	Mean arterial blood	87.5 (8.2)	-0.18	0.26	-0.05	0.78
	pressure (mmHg)					
13	Depression Anxiety	9.9 (10.4)	-0.02	0.89	0.29	0.07
	Stress Scale-21 –	~^	· /			
	Depression score	25				
14	Depression Anxiety	8.2 (8.6)	-0.13	0.41	0.19	0.25
	Stress Scale-21 – anxiety					
	score					
15	Depression Anxiety	14.5 (9.9)	-0.16	0.31	0.33	0.04
	Stress Scale-21 – stress					
	score					
16	Multi-dimensional Scale	5 (0.8)	0.02	0.88	0.08	0.61
	of Perceived Social					
	Support – total score					
17	Multi-dimensional Scale	5 (1)	-0.06	0.71	-0.00	0.99
	of Perceived Social					
	Support – score for					
	significant other					

18	Multi-dimensional Scale of Perceived Social	5.1 (0.9)	0	1.00	0.08	0.62
	Support – score for					
	family					
19	Multi-dimensional Scale	4.8 (0.9)	0.03	0.84	0.11	0.50
	of Perceived Social					
	Support – Score friend					2
20	Fasting blood sugar	91.6 (7.9)	0.09	0.58	0.06	0.71
	(mg/dL)					
21	C-reactive protein (mg/l)	2.5 (2.6)	-0.01	0.94	0.15	0.37
22	Total cholesterol (mg/dL)	210.2	-0.06	0.71	-0.20	0.21
		(30.5)				
23	HDL (mg/dL)	42.2 (9.8)	0.19	0.25	0.09	0.58
24	Triglyceride (mg/dL)	130.8	0.01	0.96	-0.09	0.59
		(66.4)		, , , , , , , , , , , , , , , , , , ,		
25	LDL (mg/dL)	143.2	-0.15	0.34	-0.23	0.15
		(28)		2		
26	Very-low-density	24.8 (9.9)	0.07	0.69	-0.08	0.61
	lipoprotein (mg/dL)					
27	Total cholesterol/HDL	5.2 (1.2)	-0.16	0.33	-0.13	0.43
28	Serum creatinine	0.9 (0.2)	0.12	0.48	0.07	0.67
	(mg/dL)					
29	Estimated glomerular	96.1	-0.05	0.78	0.30	0.06
	filtration rate	(14.5)				
	$(ml/min/1.73m^2)$					
30	Cortisol (nmol/L)	317.3	0.14	0.38	Not	Not
		(132.5)			applicable	applicable
31	NPY (pg/mL)	431.4	Not	Not	0.14	0.38
) >	(229.7)	applicable	applicable		
32	Allostatic load against	1.7 (1.4)	-0.15	0.37	Not	Not
	neuropeptide Y				applicable	applicable
33	Allostatic load against	1.7 (1.5)	Not	Not	-0.18	0.27

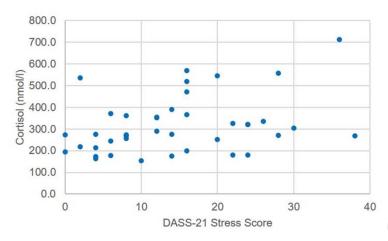


Fig. 2. Cortisol against DASS-21 stress score among healthy dwellers (r=0.33; p=0.04)

NPY and its association with the variables of interest among healthy dwellers

The mean (SD) of NPY was 431.4 (229.7) pg/mL with a lower and upper 95% CI of 360.2 and 502.6 respectively. The NPY ranged from 106.7 to 907.2 pg/mL. The median was 405.1 pg/mL with an interquartile range of 242.4 to 610.7. Skewness and kurtosis for the distribution were +0.31 and -0.92 respectively showing a potentially symmetrical and mesokurtic shape. The post hoc power was 0.98 for the distribution of NPY. The following parameters were used to calculate the power: study mean (101.4 pmol/l), mean (SD) from previous literature among healthy controls from the USA [79.8 (34.9) pmol/l], sample size (40) and type I error (0.05).⁴² The study mean NPY in pg/mL was multiplied by 0.235 to convert to pmol/l.63 The distribution of NPY was not significantly different in relation to sex, sector, whether the participant was the head of household, education level, marital status, number of adults and children at home (males and females), household income, and alcohol consumption. Table 4 summarises the p-values for the categorical variables of interest against NPY. Also, age, duration of stay at Anuradhapura, hours of sleep per day, weight, height, BMI, waist circumference, hip circumference, waist/hip ratio, systolic blood pressure, diastolic blood pressure, mean arterial blood pressure, DASS-21 scores for depression, anxiety and stress, MSPSS score (total, significant other, family, friend), fasting blood sugar, c-reactive protein, total cholesterol, HDL, triglyceride, LDL, very-low-density lipoprotein, total cholesterol/HDL ratio, serum creatinine, estimated glomerular filtration rate, cortisol, and allostatic load did not have a significant correlation with NPY. Table 5 summarises the p-values for the continuous variables of interest against NPY. However, a backward stepwise regression found only the hours of sleep per day to have a p<0.05. The R^2 of the single variable regression was 0.11 with the predictive model, NPY level; -27.71 + 63.99 (hours of sleep/day). NPY levels did not show a significant difference between the age groups of 18 to \leq 30 (median 549.4 pmol/l), 31 to \leq 40 (median 463.1 pmol/l), 41 to \leq 50 (median 257.7 pmol/l), and 51 to \leq 60 (median 386.1 pmol/l) years (p=0.41).

Comparison of NPY and cortisol of healthy dwellers against the pet dog owners and non-pet owners

The healthy dwellers between the age of 41 to \leq 60 years had a non-significant mean (SD) [268.4 (83.4) nmol/L], and median (interquartile range) [272.2 (209.7–297.6) nmol/L] of cortisol when compared to the pet dog owners' group [234.4 (73.1) nmol/L, 235.3 (188.9–292.2) nmol/L] (p=0.22) and non-pet owners' group [234.8 (122.2) nmol/L, 193 (149.1–286.9) nmol/L] (p=0.08) respectively. Also, the healthy dwellers between the ages of 41 to \leq 60 years had a non-significant mean (SD) [378 (237) pg/mL], and median (interquartile range) [339 (158.3–567.4) pg/mL] of NPY when compared to the pet dog owners' group [388.2 (225.4) pg/mL, 341.1 (223–512.8) pg/mL] (p=0.71) and non-pet owners' group [408 (215) pg/mL, 347.6 (211.2–592.1) pg/mL] (p=0.49) respectively.

Discussion

To the best of our knowledge, it is the first study from the South Asian region to compare NPY, cortisol, and other biochemical tests related to psychosocial and cardiovascular health between pet dog owners and age, sex-matched non-pet owners. The study outcomes are helpful for one-health research in low-middle income and South Asian regions. Our study did find a significantly lower depression score in the pet dog owners' group when compared to the non-pet owners' group. Depression scores of pet owners have been significantly lower than non-pet owners in prior literature as well.⁶⁴ Also, pet owners were less depressed than non-pet owners.⁶⁵ Similar findings have been reported in other settings during the COVID-19 pandemic.⁶⁶ Thus, the above evidence suggests a lower depression level among pet owners when compared to non-pet owners. Further, a community-based three-arm controlled study concludes that pet dog acquisition may reduce loneliness among owners.⁶⁷ Moreover, pet ownership has been found to provide companionship, give a sense of purpose, reduce loneliness and increase socialisation among community-dwelling older adults which may increase resilience against mental illnesses.⁶⁸ However, few studies did not find an association between pet ownership and depression in adolescents and older adults.^{69,70} Owners perceived stress reduction when interacting with their pet dogs.⁴⁹ Dog ownership is associated with

Owners perceived stress reduction when interacting with their pet dogs. Dog ownership is associated with significant decreases in stress and post-traumatic stress symptoms among veterans. A systematic review revealed a significant reduction of the stress hormone cortisol in dog companionship and dog-assisted therapy showing both long-term (5.5 years) and short-term (15 to 30 mins) influence respectively. A session with a therapy dog significantly reduced systolic blood pressure, pulse rate, and salivary cortisol levels among nursing students. Also, a session with a facility dog and a palliative care psychologist significantly reduced the pulse rate and salivary cortisol among veterans of the armed forces receiving palliative care. Further, the canine companionship group had significantly lower total cholesterol and serum cortisol when compared to the control group. Cortisol is implicated in hypertension, type 2 diabetes mellitus, dyslipidaemia, and immune modulation. And, high cortisol level is associated with metabolic

syndrome.⁷² Although the cortisol levels were not significantly different between the pet dog owners and the non-pet owners of the present study, the pet bond score among pet dog owners had a significant negative correlation with cortisol. More the bond with the pet dog lower was level of the stress hormone cortisol. Also, the present study among healthy dwellers showed a significant positive correlation between stress scores and cortisol. Further, the hormone cortisol is known to have a positive association with incident cardiovascular disease.³⁸ Nevertheless, the loss of a pet dog is also associated with stress in the bereaved owner.⁷³ The present study used the pet bond score to measure the bond between owners and pet dogs. Such studies are scarce in the literature. The high pet bond group had a significantly higher percentage of females and participants having a monthly household income of more than Rs 100,000. In prior literature, pet ownership was significantly associated with the number of adult females at home.⁴⁸ Also, households with an adult female are more likely to own a dog.⁷⁴ Further, households with high incomes are more likely to own a dog.⁷⁵

To the best of our knowledge, it is the first, community-based study among healthy volunteers from the region to report the measures of central tendency, variation for NPY and cortisol, and its relationship with socio-demographic factors, anthropometry, blood pressure, scale scores, and biochemical tests. The study mean (SD) of cortisol [317.3 (132.5) nmol/L] among the healthy dwellers of the present study was comparable to an Asian study among healthy participants from Thailand [381.93 (97.74) nmol/L].⁶² Also, the study among healthy volunteers showed a significant positive correlation between stress scores and cortisol. A meta-analysis provides evidence for an association between natural stressors and elevated cortisol levels.³⁵ An elevation of salivary cortisol during stressful periods compared to relaxed periods was reported.⁷⁶ However, prior literature has also shown a significant association for cortisol with depression and anxiety. A significant association between current anxiety disorder and higher awakening salivary cortisol was reported.⁷⁷ And, another study found higher stress and serum cortisol levels among individuals with major depressive disorder.⁷⁸

The study among the healthy dwellers showed that the distribution of cortisol in those who were never married was significantly higher compared to those who were currently married. A study has shown that married individuals had lower salivary cortisol levels than never married or previously married. It further revealed that married had a rapid decline in cortisol during the afternoon hours compared to never married (but not with previously married). And, higher stress was associated with higher cortisol levels in previously married compared to the married and never married groups. Thus, cortisol is a candidate mechanism for the association of marital status and health.⁷⁹ Also, the study among healthy dwellers showed that the distribution of cortisol in those who consumed alcohol in moderation was significantly higher compared to those who never consumed alcohol. Among men and women alcohol consumption was positively associated with serum cortisol.⁸⁰ Also, acute binge intoxication is associated with increased blood cortisol.⁸¹ Further, a positive relationship between different indices of alcohol intake and daily release of salivary

cortisol was found in an ageing cohort. ⁸² Our study showed a significant negative correlation between age and cortisol. A study also found that the cortisol peak is advanced by 24 minutes per decade concerning the age of healthy adults. ⁸³ However, another study showed that age above 60 years was associated with an increased morning serum cortisol. ⁸⁴ Our study included only participants aged 18 to 60 years. The present study showed a significant positive correlation between hours of sleep per day and cortisol. A prior clinical trial has shown elevated plasma cortisol the next evening due to loss of sleep. ⁸⁵ Greater sleep-wake behaviour problems were associated with decreased cortisol responses among children and adolescents. ⁸⁶ Also, the present study's backward stepwise regression model found a positive association between height and cortisol levels among healthy dwellers. However, prior literature showed a negative association between fasting plasma cortisol and adult height which shows that adult height may be affected by physiological variations in adrenocortical glucocorticoid secretion. ⁸⁷

The study mean (SD) of NPY [431.4 (229.7) pg/mL] was comparable to an Asian study among metabolically healthy obese participants from China [478.89±145.53 pg/mL].⁴³ However, NPY from the study was higher when compared to the findings from the UK and the USA which used radioimmunoassay.^{41,42} Hence, the present study may hint towards a difference in NPY between the Global North and South. However, future research is necessary for confirmation. The NPY did not show a significant association with any of the possible determinants among healthy dwellers selected based on predetermined selection criteria. However, a backward stepwise regression found hours of sleep per day to be significantly associated with NPY. NPY has hypnotic properties and regulates sleep in humans.⁸⁸ And, NPY inhibits noradrenergic signalling to promote sleep.⁸⁹

The findings of the comparative study are unique, as it has compared demographic and laboratory data between pet dog owners and age, sex-matched non-pet owners of a low-middle-income South Asian region. The study produced exclusive data about the effect of canine companionship on psychosocial and cardiovascular health from a South Asian country where similar studies were scarce. Future studies among participants with non-communicable diseases and on different treatment options are methodologically challenging yet, would yield further details on the role of canine companionship in psychosocial and cardiovascular health. Also, it is crucial to assess the cost-effectiveness of canine companionship and its health benefits among socioeconomically deprived families. A positive finding on the effect of pet dog ownership on human psychosocial health will guide towards one-health interventions among the dwellers of this community. Also, the findings will help plan future research on canine companionship and human psychosocial health. Further, the study found the baseline values of NPY and cortisol among healthy dwellers of a low-middle-income South Asian country. It will help interpret such biomarkers in future research and clinical practice in the region.

Study limitations

The study among healthy dwellers was conducted in one of 25 districts in Sri Lanka which cannot be generalized. Also, the study lacks adequate comparison due to the unavailability of previous similar local or regional data. Further, the cost of laboratory investigations limited the sample size to 40 healthy dwellers. However, it produced valuable baseline data on NPY and cortisol levels among healthy dwellers of a district in a low-middle-income South Asian country where similar studies were scarce. Thus, the findings will be helpful for future research in South Asia. Geographic, sociocultural, and other demographic differences could have led to the negative findings in the present study. Publishing negative results could lead to a new understanding and interpretation of prior positive results. Further, it would help adjust future research plans and increase the chance of positive results.⁹⁰

Conclusion

A significantly lower depression score in the pet dog owners' group than in the non-pet owners' group was observed. And, the pet bond score had a significant negative correlation with the stress hormone cortisol. Taken together, our results suggest that pet dog ownership may have improved psychosocial health by reducing depression and stress. Thus, canine companionship could be an option to reduce depression and stress and will guide towards one-health interventions among South Asian communities. The findings will help plan future research on canine companionship and human psychosocial health. Also, the study found the baseline values of NPY and cortisol among healthy dwellers of a low-middle-income South Asian country which will help interpret such biomarkers in future research and clinical practice in the region.

Supplementary materials

Table S1 file format - .docx. Cut-off and points for allostatic load.

Table S2 file format - .docx. Comparison of categorical variables between the pet dog owners' group and the non-pet owners' group.

Table S3 file format - .docx. Categorical variables of interest against the pet bond score among pet dog owners.

Declarations

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Author contributions

Conceptualization, D.R.; Methodology, D.R., J.R. and K.W.; Software, D.R.; Validation, D.R., J.R. and K.W.; Formal Analysis, D.R.; Investigation, D.R.; Resources, D.R.; Data Curation, D.R.; Writing – Original Draft Preparation, D.R.; Writing – Review & Editing, J.R. and K.W.; Visualization, D.R.; Supervision, J.R. and K.W.; Project Administration, D.R.; Funding Acquisition, D.R.

Conflicts of interest

The author(s) declare no competing interests.

Data availability

All data generated or analyzed during this study are included in this published article (and its Supplementary Information files).

Ethics approval

Ethical clearance was obtained from the Ethics Review Committee of the Faculty of Medicine and Allied Sciences, Rajarata University of Sri Lanka (ERC/2020/76).

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