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Short-term effects of polyphenol-rich black tea on blood pressure in men and women

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Abstract

There is increasing evidence that black tea polyphenols contribute to vascular health. We have recently shown that regular ingestion of polyphenol-rich black tea over 6 months results in lower systolic and diastolic blood pressure. However, the time course of these effects remains unclear. Therefore, our objective was to determine if short-term effects of tea on blood pressure could contribute to longer-term benefits of regular tea consumption on blood pressure. Men and women (n=111) were recruited to a randomised placebo-controlled double-blind parallel designed trial. During a 4-wk run-in, all participants consumed 3 cups/d of black tea. Participants then consumed 3 cups over 1 d of either powdered black tea solids containing 429 mg of polyphenols (tea), or a control product matched in flavour and caffeine content, containing no tea solids. The 24 h ambulatory blood pressure and heart rate was measured at the end of the 4-week run-in (baseline) and again during the 24 h intervention period. The 24 h, day-time and night-time blood pressures were not significantly different between tea and control ($P>0.05$). Baseline-adjusted net effects on mean 24 h ambulatory blood pressure for systolic and diastolic blood pressure were -0.2 mm Hg (95% CI, -1.5 to 1.0), $P=0.72$, and 0.0 mm Hg (95% CI, -1.0 to 0.9), $P=0.95$, respectively. Heart rate was significantly lower for tea compared to control during the night-time and early-morning periods (-2.0 (95% CI, -3.2,-0.8) bpm; and -1.9 (95% CI, -3.7,-0.2) bpm, respectively, $P<0.05$ for both), but not during the day-time. These results suggest that the longer-term benefits of black tea on blood pressure are unlikely to be due to short-term changes.

39 **Introduction**

40 Population changes to diet and lifestyle are the primary means of combating chronic lifestyle-
41 related diseases. High blood pressure is a leading risk factor contributing to the global burden of
42 disease^{1,2}, and small changes in blood pressure due to dietary change may significantly impact on
43 risk of hypertension and cardiovascular disease^{3,4}. Regular consumption of black tea (*Camellia*
44 *sinensis*) may lower blood pressure, but the acute and short-term effects of tea on blood pressure are
45 less clear.

46 Results of population studies indicate that a higher intake of black tea can protect against
47 hypertension^{5,6} and cardiovascular disease^{7,8}. There is also evidence from randomised controlled
48 trials that regular consumption of black tea can lower blood pressure^{9,10}, but the time-course of this
49 effect is not clear. We have recently demonstrated that regular consumption of 3 cups per day of
50 polyphenol-rich black tea over 6 months, compared to a control beverage matched for caffeine
51 content, results in lower systolic and diastolic blood pressures by between 2 and 3 mm Hg⁹. Grassi
52 et al¹⁰ found that regular consumption of black tea for 1 week resulted in lower office blood
53 pressure by between 2 and 3 mm Hg, but not ambulatory blood pressure. We have also found that
54 regular consumption of black tea for 1 week did not alter ambulatory blood pressure¹¹. In contrast,
55 our studies exploring the acute effects of black tea on blood pressure indicate that when tea is
56 consumed after an overnight fast it can cause a transient rise in blood pressure^{11,12}. However, the
57 relevance of these findings to usual tea consumption in the general population may be limited. The
58 acute intake of polyphenols and/or caffeine in these studies was high in comparison to intakes
59 during usual tea consumption in the general population. Therefore, our objective of this analysis
60 was to determine if short-term changes in blood pressure could contribute to the benefits of regular
61 tea consumption on blood pressure.

62 **Subjects and methods**

63 *Participants*

64 Volunteers were regular tea drinkers recruited from the general population using print media
65 advertisements. The primary inclusion criterion was for participants to have normal to mildly
66 elevated systolic blood pressures between 115 and 150 mm Hg. This was based on a 4 h day-time
67 ambulatory blood pressure measurement performed at screening. Participants could be included if
68 they were taking up to three antihypertensive medications, they were aged 35 to 75 y, had a body
69 mass index of 19 to 35 kg/m², and were non-diabetic, and were otherwise healthy. The trial was
70 approved by the University of Western Australia Ethics Committee, and registered at the Australian
71 New Zealand Clinical trials Registry as **ACTR#**:12607000543482.

72 *Study design*

73 A randomised placebo controlled double-blind parallel designed intervention study was performed.
74 During a 4-week run-in period the participants were asked to consume 3 cups/d of regular black leaf
75 tea prepared in the usual manner. This is because all participants were regular tea drinkers, but tea
76 intake varied between individuals. Assessing the effects of tea against a background of regular tea
77 drinking therefore allowed our results to be generalizable to tea-drinking populations. In order to
78 achieve a degree of uniformity of background flavonoid intake, the intake of a few specific high
79 flavonoid foods, including dark chocolate, red wine and fruit juices, was limited throughout the
80 study. During the fourth week of the run-in period 24 h ambulatory blood pressure measurement
81 was performed (baseline). Participants were then assigned to consume 3 cups over 24 h of either
82 powdered black tea solids containing a total of 429 mg of polyphenols and 96 mg of caffeine (tea),
83 or a placebo matched in flavour and caffeine content, containing no tea solids. Intake of regular leaf

tea ceased during this 24 h period. Ambulatory blood pressure was measured again during this 24 h period (intervention). The powdered black tea was derived from a blended black tea (*Camellia sinensis*). Participants were advised to consume the tea or placebo products at the time that they would normally consume tea. The contents of a sachet was added to approximately 200 ml of boiled hot water and stirred until it was completely dissolved. Addition of sugar, milk, cream or other additives was not allowed, and the product was consumed while still hot.

Volunteers were randomized (1:1) using computer generated random numbers. Randomization codes were sealed in envelopes, which were opened in consecutive order as each participant deemed eligible was entered into the study.

Blood pressure

Screening blood pressure was assessed as 4-hour day-time ambulatory blood pressure, with blood pressure and heart rate measured every 20 min. During the study, blood pressure was assessed as 24 h ambulatory blood pressure with blood pressure and heart rate measured every 20 min during the day (6:00 am to 9:59 pm) and every 30 min overnight (10:00 pm to 5:59 am)¹³. These measurements were performed at the end of the 4-week run-in and during the 24 h intervention period. A trained researcher fitted a Spacelabs ambulatory blood pressure monitor (Spacelabs Medical Inc. Redmond, WA, USA) and explained its use to the participants. The monitor was fitted to the non-dominant arm approximately 2.5 cm above the antecubital fossa. Adult or large adult size cuffs were used depending on upper arm circumference assessed at baseline. Participants were instructed to continue their usual daily activities and to avoid any vigorous exercise. Measurements showing an error code or those with a pulse pressure of less than 20 mm Hg were excluded from the analysis. Blood pressure traces were regarded as being complete if more than 80% of the recordings were valid.

107 *Statistical analysis*

108 Analysis was performed according to a pre-specified statistical analysis plan, which was finalized
109 prior to breaking the randomization code. Descriptive statistics are presented as mean and standard
110 deviation. Categorical variables are summarized by number and percentage in each category.

111 At baseline, characteristics of participants in the two groups were compared using the independent-
112 samples t-test and the chi-squared test for categorical variables. Ambulatory blood pressure data
113 were analysed using linear mixed models in STATA. The STATA “xtmixed” and “margins”
114 commands were used to determine baseline-adjusted between-group differences. Subject was
115 included as a random factor in each model. Fixed effects included baseline value, group (placebo or
116 tea), hour (categories 1 to 24) and group X hour. The baseline-adjusted ambulatory blood pressure
117 and heart rate values and between-group differences are presented as least squares means and 95%
118 CI. $P < 0.05$ was used to indicate statistical significance. Separate models were used to assess 24-hr,
119 early-morning, daytime and night-time effects.

120

Results

Descriptive and baseline data

A total of 111 participants were randomised (55 to placebo and 56 to tea). Complete ambulatory blood pressure data were available for 84 participants: 43 in the placebo group and 41 in the tea group. Baseline characteristics of the participants are presented in **Table 1**. There were no significant differences between the groups in these variables at baseline. The two groups were also well matched for use of antihypertensive medications. In the placebo group 11 participants were taking antihypertensive medication, and in the tea group 7 participants were taking antihypertensive medication ($P=0.59$).

Blood pressure

The mean 24 h, day-time, night-time and early-morning systolic and diastolic blood pressures and heart rates during baseline and intervention are presented in **Table 2**. Tea compared with placebo did not alter mean 24 h ambulatory blood pressure. Baseline-adjusted net effects for systolic and diastolic blood pressure were -0.2 mm Hg (95% CI, -1.5 to 1.0), $P=0.72$, and 0.0 mm Hg (95% CI, -1.0 to 0.9), $P=0.95$, respectively. In addition, black tea did not significantly alter blood pressure during pre-specified periods during the 24 h. Tea compared to placebo resulted in lower heart rate at night-time ($P<0.001$) and early-morning ($P=0.03$), but not during the day-time period ($P=0.20$).

Discussion

Black tea is the most widely consumed type of tea. There is growing evidence that black tea and its polyphenols (primarily flavonoids) can make an important contribution to vascular health^{7,8,14}. Our study explored the short-term effects of polyphenol-rich black tea, over 1 d, on ambulatory blood pressure. Background regular tea intake was standardised and maintained at 3 cups per day. Participants then consumed either tea or control, 3 cups during 1 d. The 24 h ambulatory blood pressures were not different between beverages, and blood pressures were not influenced during specific time periods within the 24 h intervention period. That is, tea consumption did not significantly influence ambulatory blood pressure in the short-term. However, heart rate was significantly lower with tea compared with control during the night-time and early-morning periods. Results of population studies indicate that a higher intake of tea may protect against hypertension^{5,6}. There is also evidence from randomised controlled trials that regular consumption of black tea^{9,10} and green tea^{15,16} can result in lower blood pressure. We have recently demonstrated that regular consumption of 3 cups/d of black tea over 6 months resulted in lower systolic and diastolic blood pressures of between 2 and 3 mm Hg⁹. We have addressed here any short-term contribution to longer-term effects on blood pressure in this study, and demonstrated that longer-term benefits of tea on blood pressure are unlikely to be due to immediate effects on ambulatory blood pressure. Our data are consistent with a recent short-term controlled trial that assessed the effects of green tea on blood pressure¹⁷. Rudelle et al¹⁷ demonstrated that green tea consumption over 3 days did not alter mean 24 h blood pressure measured while participants resided in an indirect calorimeter chamber. Several previous studies have investigated acute effects of tea and its components on blood pressure^{11,12,18-20}. These studies have primarily been interested in the acute effects of a single dose

of tea and/or its components, and often in the morning after an overnight fast. In this setting, tea and caffeine have been shown to cause a transient increase in blood pressure^{11,12,18}. These changes in blood pressure may have a bearing on acute improvements in cognitive performance^{19,20}. However, their relevance to the impact of regular tea consumption on ambulatory blood pressure and ultimately to the risk of hypertension and cardiovascular disease is uncertain.

We have shown here that a powdered black tea rich in polyphenols, in comparison to a control beverage matched for caffeine, as it would usually be consumed in the general population did not influence ambulatory blood pressure over a single day. We also found that blood pressure was not significantly altered in the early-morning period, a time comparable to previous acute studies, or during day-time or night-time periods. Our study assessed the effects of black tea against a background of regular tea drinking. It remains possible that effects of black tea could be different if participants were not consuming tea, coffee or caffeine during the run-in period. These results indicate that the previously observed rapid onset and short-lived pressor response to tea^{11,12} has minimal impact on ambulatory blood pressures in the short-term. Therefore, although non-caffeine components in black tea when consumed at high levels in the fasting state have the potential to raise blood pressure acutely, their effects on ambulatory blood pressure during usual tea consumption are likely to be minimal.

We have also shown that heart rate was significantly lower with black tea compared to control during the night-time and early-morning periods. These effects can be interpreted as an effect of black tea polyphenols to reduce heart rate, but could equally be interpreted as an effect of withdrawal of non-caffeine black tea solids to raise heart rate. The mechanisms responsible are not clear. Caffeine can acutely influence heart rate²¹. However, the observed effects were independent of caffeine, because the two beverages were matched for caffeine content. Results of previous

human trials suggest that regular ingestion of a high polyphenol diet leads to small and mostly non-significant effects on heart rate^{9,22, 23}. A additional possibility is that other components of black tea such as L-theanine could influence heart rate^{24, 25}. Further, because heart rate differences were found only during the night-time and early-morning periods, it is possible that the withdrawal of polyphenols, L-theanine, or other component of black tea could influence sleep quality, perhaps via attenuation of sympathetic nervous activation²⁵⁻²⁷.

Elevated heart rate may be an independent risk factor for cardiovascular disease²⁸. However, the magnitude of the effects on night-time and early-morning heart rate, -2 bpm, was small. The clinical importance of small differences in heart rate is not clear. Therefore, an important question relating to the clinical relevance of these findings is whether effects are maintained longer-term. Analysis of the longer-term data⁹ showed that an effect of tea to reduce heart rate at night was still present at 3 months but was not present at 6 months. Thus the clinical relevance of these short-term effects is therefore uncertain.

Conclusions

In conclusion, we have demonstrated that black tea rich in polyphenols, taken as black tea would normally be consumed over a single day, did not alter ambulatory blood pressure. The results indicate that any benefits on blood pressure with long-term regular black tea intake are unlikely to be due to short-term blood pressure changes. These results also suggest that any acute effects of high levels of intake of tea to alter blood pressure in the fasting state have minimal impact on ambulatory blood pressures over 24 h.

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215 data analysis. The primary data analysis was performed independent of Unilever Research and
216 Development by R. J. W., employed by Flinders University, Adelaide, Australia, who followed the
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218 analysis. J. M. H. had full access to all the data in the study and takes responsibility for the integrity
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References

1. M. Ezzati, A. D. Lopez, A. Rodgers, H. S. Vander and C.J. Murray. *Lancet*, 2002, **360**,: 1347-1360.
2. P. M. Kearney, M. Whelton, K. Reynolds, P. Muntner, P. K. Whelton and J. He. *Lancet*, 2005, **365**, 217-223.
3. R. Collins and MacMahon S. *Br. Med. Bull.*, 1994;**50**, 272-298.
4. S. Lewington, R. Clarke, N. Qizilbash, R. Peto and R. Collins. *Lancet*. 2002, **360**, 1903-1913.
5. J. M. Hodgson, A. Devine, I. B. Puddey, S. Y. Chan, L. J. Beilin and R. L. Prince. *J. Nutr.*, 2003, **133**, 2883-2886.
6. Y. C. Yang, F. H. Lu, J. S. Wu, C. H. Wu and C. J. Chang. *Arch. Intern. Med.*, 2004, **164**, 1534-1540.
7. L. Arab, W. Liu and D. Elashoff. *Stroke*, 2009, **40**, 1786-1792.
8. Z. M. Wang, B. Zhou, Y. S. Wang, et al. *Am. J. Clin. Nutr.*, 2011, **93**, 506-515.
9. J. M. Hodgson, I. B. Puddey, R. J. Woodman, T. Mulder, D. Fuchs, K. Scott and K. D. Croft. *Arch. Intern. Med.*, 2012, **172**, 186-188.
10. D. Grassi, T. P. Mulder, R. Draijer, G. Desideri, H. O. Molhuizen and C. Ferri. *J. Hypertens.*, 2009, **27**, 774-781.
11. J. M. Hodgson, I. B. Puddey, V. Burke and L. J. Beilin. *J. Hypertens.*, 1999, **17**, 457-463.
12. J. M. Hodgson, V. Burke and I. B. Puddey. *J. Hypertens.*, 2005, **23**, 47-54.
13. Y. P. Lee, T. A. Mori, I. B. Puddey, et al. *Am. J. Clin. Nutr.*, 2009, **89**, 766-782.
14. J. M. Hodgson and K. D. Croft. *Molec. Aspects. Med.*, 2010, **31**, 495-502.
15. T. Matsuyama, Y. Tanaka, I. Kamimaki, T. Nagao and I. Tokimitsu. *Obesity*, 2008, **16**, 1338-1348.
16. M. P. Nantz, C. A. Rowe, J. F. Bukowski and S. S. Percival. *Nutrition*, 2009, **25**, 147-154.
17. S. Rudelle, M. G. Ferruzzi, I. Cristiani, J. Moulin, K. Mace, K. J. Acheson and L. Yappy. *Obesity*, 2007, **15**, 349-355.
18. P. Quinlan, J. Lane and L. Aspinall. *Psychopharmacology*, 1997, **134**, 164-173.
19. A. Belza, S. Toubro and A. Astrup. *Eur. J. Clin. Nutr.*, 2009, **63**, 57-64.
20. T. Giesbrecht, J. A. Rycroft, M. J. Rowson, E. A. De Bruin. *Nutr. Neurosci.*, 2010, **13**, 283-290.
21. P. J. Green, R. Kirby and J. Suls. *Ann. Behav. Med.*, 1996, **18**, 201-216.
22. R. R. Zilkens, V. Burke, J. M. Hodgson, A. Barden, L. J. Beilin and I. B. Puddey. *Hypertension*, 2005, **45**, 874-879.
23. N. C. Ward, J. M. Hodgson, K. D. Croft, V. Burke, L. J. Beilin and I. B. Puddey. *J. Hypertens.*, 2005, **23**, 427-434.
24. A. Higashiyama, H. H. Htay, M. Ozeki, L. R. Juneja and M. P. Kapoor. *J. Functional Foods*, 2011, **3**, 171-178.
25. K. Kimura, M. Ozeki, L.R. Juneja and H. Ohira. *Biol. Psychol.*, 2007, **74**, 39-45.
26. M. Tanida, N. Tsuruoka, J. Shen, Y. Kiso and K. Nagai. *Metab. Clin. Exp.*, 2008, **57**, 526-534.
27. A. Steptoe, E. L. Gibson, R. Vuononvirta, E. D. Williams, M. Hamer, J. A. Rycroft, J. D. Erusalimsky and J. Wardle. *Psychopharmacology*, 2007, **190**, 81-89.
28. A. Hjalmarson. *Eur. Heart J.*, 2007, **9**, F3-F7.

Table 1 Baseline characteristics of participants in the placebo and tea groups

	Placebo (n 43)	Tea (n 41)	P ^a
Men/Women (n)	16/27	15/26	0.95
Age (y)	56.2 ± 10.9 ^b	56.1 ± 10.7	0.94
Body mass index (kg/m ²)	25.3 ± 3.5	24.8 ± 3.7	0.55
Total Cholesterol (mmol/L)	5.1 ± 0.9	5.1 ± 0.9	92
LDL Cholesterol (mmol/L)	3.2 ± 0.8	3.2 ± 0.8	0.95
HDL Cholesterol (mmol/L)	1.40 ± 0.34	1.41 ± 0.31	0.95
Triglycerides (mmol/L)	1.1 ± 0.6	1.1 ± 0.4	0.94
Glucose (mmol/L)	5.1 ± 0.5	5.2 ± 0.9	0.28
Insulin (mU/L)	7.0 ± 4.9	8.3 ± 8.9	0.41

^a P-value for between-group difference analysed using the independent samples t-test and the chi-squared test for categorical variables

^b Values are presented as mean ± standard deviation

Table 2 Mean systolic blood pressure, diastolic blood pressure and heart rate according to beverage (placebo or tea) during baseline and intervention, and between group differences ^a

	Placebo		Tea		Baseline-adjusted between group difference
	Baseline	Intervention	Baseline	Intervention	
<i>24 hour</i>					
Systolic blood pressure (mm Hg)	121.1 (120.2,122.0)	122.0 (121.1,122.9)	121.3 (120.4,122.2)	121.8 (120.9,122.7)	-0.2 (-1.5,1.0) <i>P</i> =0.72
Diastolic blood pressure (mm Hg)	72.3 (71.6,73.0)	72.9 (72.2,73.6)	72.0 (71.3,72.7)	72.9 (72.1,73.6)	0.0 (-1.0,0.9) <i>P</i> =0.95
Heart rate (bpm)	69.5 (68.7,70.0)	69.0 (68.2,69.7)	68.9 (68.2,69.7)	68.4 (67.6,69.2)	-0.6 (-1.6,0.5) <i>P</i> =0.30
<i>Day-time (6:00 am – 9:59 pm)</i>					
Systolic blood pressure (mm Hg)	126.0 (125.0,127.0)	127.4 (126.4,128.3)	126.0 (125.0,127.0)	126.5 (125.5,127.5)	-0.9 (-2.2,0.5) <i>P</i> =0.22
Diastolic blood pressure (mm Hg)	76.5 (75.8,75.2)	77.4 (76.6,78.1)	76.0 (75.2,76.7)	77.0 (76.3,77.8)	-0.4 (-1.4,0.7) <i>P</i> =0.50
Heart rate (bpm)	73.9 (72.8,75.0)	72.4 (71.3,73.5)	73.6 (72.5,74.7)	73.4 (72.3,74.6)	1.0 (-0.6,2.7) <i>P</i> =0.20
<i>Night-time (10:00 pm – 5:59 am)</i>					
Systolic blood pressure (mm Hg)	111.3 (110.4,112.3)	111.3 (110.4,112.3)	111.6 (110.6,112.6)	111.9 (110.9,112.9)	0.6 (-0.8,2.0) <i>P</i> =0.42
Diastolic blood pressure (mm Hg)	63.9 (63.1,64.6)	63.9 (63.1,64.7)	63.7 (62.9,64.5)	64.2 (63.4,65.1)	0.3 (-0.8,1.5) <i>P</i> =0.57
Heart rate (bpm)	60.8 (60.0,61.6)	61.9 (61.0,62.7)	60.4 (59.6,61.3)	59.8 (59.0,60.8)	-2.0 (-3.2,-0.8) <i>P</i> <0.001
<i>Early-morning (6:00 am – 8:59 am)</i>					
Systolic blood pressure (mm Hg)	122.7 (121.5,123.8)	123.4 (122.2,124.5)	122.5 (121.4,123.7)	125.0 (123.7,126.2)	1.6 (-0.1,3.3) <i>P</i> =0.06
Diastolic blood pressure (mm Hg)	63.8 (63.1,64.6)	63.9 (63.1,64.7)	63.7 (62.9,64.5)	64.2 (63.4,65.1)	0.3 (-0.8,1.5) <i>P</i> =0.57
Heart rate (bpm)	71.9 (70.8,73.0)	71.6 (70.4,72.8)	71.8 (70.7,73.0)	69.7 (68.4,71.0)	-1.9 (-3.7,-0.2) <i>P</i> =0.03

^a Analysed using mixed models in STATA using “xtmixed” and “margins” commands to determine baseline-adjusted between-group differences during intervention. Values are estimated marginal means (95% CI) for intervention versus control.