The chronic effect of physical activity on postprandial triglycerides in postmenopausal women: A randomized controlled study

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ABSTRACT

Objective: This study examined the chronic effect of increased physical activity on postprandial triglycerides in older women.

Methods: Twenty-six women, aged 72 ± 5 years (mean ± SD), participated in this study. Participants in the physical activity group (n = 11) were asked to increase their activities above their usual lifestyle levels for 12 weeks. Participants in the control group (n = 15) maintained their usual lifestyle for 12 weeks. All participants rested and consumed a standardized breakfast after a 24-h period of physical activity avoidance at baseline, 4 weeks, and 12 weeks. Blood samples were collected in the fasted state (0 h) and at 2, 4, and 6 h after breakfast.

Results: The average increased time spent in self-selected activities per day was 1.1 ± 19.3 min over the 12 weeks compared with the baseline in the physical activity group. There was no difference in the postprandial time-averaged triglyceride area under the curve at baseline (1.59 ± 0.81 vs. 1.39 ± 0.67 mmol/L, p = 0.515) or over the 12-week intervention (1.78 ± 1.00 vs. 1.31 ± 0.67 mmol/L, p = 0.212) between the physical activity and control groups.

Conclusion: Postprandial triglyceride concentrations were not reduced after performing self-selected activities under free-living conditions in older women when these responses were determined 24 h after the last physical activity bout. (Trial registration ID: UMIN000037420).

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Introduction

In 1979, Zilversmit1 proposed that atherosclerosis is a postprandial phenomenon. This notion is now supported by the findings of large prospective cohort studies which addressed that repeated daily episodes of elevated non-fasting triglycerides (TG) and prolonged residence in the circulation of TG-rich lipoproteins were risk factors for cardiovascular disease and all-cause mortality in men and women.2,3 Given that people spend up to three-quarters of each day in the postprandial state, it is important to consider lifestyle modifications which may be effective in reducing diurnal exaggerations in postprandial TG.

A large body of evidence supports the notion that an acute bout of aerobic exercise reduces postprandial TG concentrations in humans (for a review of relevant studies see Freese et al.4). It is worth noting that although the majority of these findings have been observed in controlled laboratory protocols,4 a previous study has shown that acute exercise-induced reduction in postprandial TG observed can also be seen when exercise performs under free-living conditions.5 Such research implications would be important from a public health viewpoint as the majority of adults do not perform adequate amounts of physical activity (i.e., exercise and daily activities) to meet public health guidelines established by expert panels.6,7

To our knowledge, eight studies8–15 have investigated that the chronic ≥ 4 weeks effect of exercise on postprandial TG concentrations in humans. Seven studies8–14 found that chronic exercise training failed to attenuate postprandial TG concentrations, but one exercise training study15 demonstrated a reduction in postprandial...
TG concentrations. The timing of postprandial testing from the last bout of the exercise training session is a possible reason for the discrepant findings among these studies. Indeed, seven of these studies were designed to exclude the acute exercise-induced reduction in postprandial TG\(^{10}\) – postprandial TG concentrations were determined 24–72 h after the last bout of exercise training session. Apart from such methodological issues, all previous studies\(^{8,10,11,12}\) except our previous study with a duration of 4 weeks,\(^{13}\) examining the chronic effect of exercise on postprandial TG concentrations employed “structured or supervised exercise training”. In our previous study, there was no reduction in postprandial TG concentration in postmenopausal women after performing self-selected activities under free-living conditions over a 4-weeks period, even though the participants in the active group increased their step counts by 600 steps per day and their total physical activity times by 5 min per day.\(^{13}\) The Ministry of Health, Labour and Welfare of Japan recommends that citizens incorporate “~10 min of physical activity” per day in anticipation of a 3.2% risk reduction in non-communicable diseases (including cardiovascular diseases).\(^{14,15}\) Therefore, it is valuable to explore whether the incorporation of a longer period and larger volume of increased physical activity under free-living conditions could result in a reduction in postprandial TG. Furthermore, given a higher dropout rate (roughly 50%) and a lower long-term adherence to physical activity than when a supervised intervention is conducted,\(^{16,17}\) it is necessary to examine whether the study participants can increase and maintain physical activities under free-living conditions. Thus, limited evidence is available to address the long-term effect (i.e., more than 4 weeks) of self-selected activities lasting more than 10 min per day performed under free-living conditions on postprandial TG.

The purpose of the present study was to examine the effects of self-selected activities performed under free-living conditions lasting for 12 weeks on postprandial TG concentrations in postmenopausal women. Postmenopausal women were employed in the present study because there is a substantial increase in the risk of cardiovascular disease with increasing concentrations of non-fasting TG and with increasing age.\(^{3,11}\) In addition, our previous study showed that encouraging postmenopausal women to accumulate additional self-selected weekend physical activity is sufficient to reduce postprandial TG.\(^{17}\) Thus, we wanted to determine whether these “acute” findings were also observed under free-living conditions where this group of individuals performed their self-selected activities over a 12-week period.

**Methods**

**Ethical approval**

This study was conducted with the approval of the Institutional Ethics Committee on Human Research (Approval number: 2019–105) and was conducted in accordance with the Declaration of Helsinki. All participants provided written informed consent to participate in this study. This study was registered in advance with the University Hospital Medical Information Network Center (UMIN), a system for registering clinical trials (ID: UMIN000037420).

**Participants**

A participant flow diagram shows in Fig. 1. Inclusion criteria included Japanese postmenopausal women 55–85 years in age who are currently non-smoking, not reporting a major illness/disease, or physical problems (acute or chronic) limiting the ability to perform activities of daily living, and had been weight stable (±3 kg) for 3 months before the study. After obtaining written informed consent for participation in the present study, 30 participants were initially enrolled, and the investigator generated the randomization sequence using computer-generated random number. Then, they were randomly allocated into a physical activity group (n = 15) or a control group (n = 15). However, three participants in the physical activity group informed us that they may not be able to comply with the requirements of the study before the main experiment. In addition, one participant in the physical activity group requested us to withdraw from the study because of venipuncture difficulties at the baseline measurement of the blood test. Therefore, 26 participants (i.e., the physical activity group, n = 11; control group, n = 15) were ultimately included in the present study. No participants had difficulties with mobility or performing activities of daily living. The baseline physical characteristics of the participants are provided in Table 1. In the physical activity group, eight women possessed the E3/3 apolipoprotein E phenotype, two women possessed the E2/3 apolipoprotein E phenotype, and one woman possessed the E3/4 apolipoprotein E phenotype. In the control group, twelve women possessed the E3/3 apolipoprotein E phenotype, one woman possessed the E2/3 apolipoprotein E phenotype, one woman possessed the E2/4 apolipoprotein E phenotype, and one woman possessed the E3/4 apolipoprotein E phenotype.

**Anthropometry**

Body mass was measured to the nearest 0.05 kg using a digital scale (Inner Scan 50, Tanita Corporation, Tokyo, Japan). Height was measured to the nearest 0.1 cm using a stadiometer (YS-OA, AS ONE Corporation, Osaka, Japan). Body mass index was calculated as weight in kilogrammes divided by the square of height in metres. Arterial blood pressure was measured from the left arm after 5 min of seated rest using a standard mercury sphygmomanometer (605P, Yagami Co Ltd, Yokohama, Japan). Two measurements were taken, and the mean of these values was recorded.

**Study design**

This was a parallel-group, controlled randomized trial designed to compare 12 weeks of a physical activity intervention (i.e., the physical activity group) versus 12 weeks of maintained individual’s habitual lifestyle (i.e., the control group). Participants in the physical activity group were asked to increase their activities above their usual lifestyle levels for 12 weeks by engaging in some examples of activities such as taking a walk, going up and down the stairs, playing with grandchildren and running. Participants freely decided the intensity of their chosen activities for at least 10 min per day. An activity amount of at least 10 min per day was chosen since this is recommended by the Ministry of Health, Labour and Welfare of Japan to promote physical activity for public health and can be incorporated into daily activity routines.\(^{18}\) Participants in the control group maintained their usual lifestyle for 12 weeks. All participants were asked to wear a uniaxial accelerometer for 12 consecutive weeks and to maintain their daily diet during the entire study period. In addition, all participants were instructed to record their daily log (physical activity, food intake, and sleeping time) in a log sheet. The primary outcome was the difference in postprandial TG between the physical activity and control groups. Postprandial TG was determined for all participants in three separate periods: at baseline and then 4 and 12 weeks after each intervention was completed. In addition to these measurements, fasting blood samples were collected at 8-week intervention in both groups. However, the postprandial blood measurement at 8-week intervention was omitted in order to improve study adherence and reduce the dropout rate for the intervention by shortening...
the participants’ time constraints and reducing their physical and mental burden of participation.

**Baseline physical activity assessment**

For the determination of physical activity levels, participants were asked to wear a uniaxial accelerometer (Lifecoder-EX, Suzuken Co. Ltd., Nagoya, Japan) on the hip to monitor their daily activity objectively for one week before the main experiments. The accelerometer defined 11 levels of activity intensity (0, 0.5, and 1–9), with 0 indicating the lowest intensity and 9 being the highest intensity. A level of 4 corresponds to an intensity of ~3 metabolic equivalents. In addition, the total step count (steps per day) was recorded and calculated from the accelerometer using computerised software (Lifelyzer 05 Coach, Suzuken Co Ltd, Nagoya, Japan).
Postprandial blood tests

Participants in the physical activity and control groups reported to the laboratory at 0845 h after a 10-h overnight fast (no food or drink except water) at three separate periods (i.e., at baseline, 4, and 12 weeks). Then, body mass was measured to the nearest 0.05 kg using a digital scale (Inner Scan 50, Tanita Corporation, Tokyo, Japan). After a 15-min rest, a fasting venous blood sample was collected in a seated position by venipuncture at 0 h (0900 h) for the measurement of circulating concentrations of total cholesterol (T-C), low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C), TG, non-esterified fatty acids (NEFA), 3-hydroxybutyrate (3-OHB), insulin, and glucose. Participants then consumed a standardized meal for breakfast (further details provided in “Test meal” section) and rested thereafter in a seated position (reading or writing) for 6 h (1500 h). Participants were not allowed to take a nap during the observation period. Researchers observed the participants throughout the day to ensure their compliance with the testing protocol. Further venous blood samples were collected by venipuncture at 2 h (1100 h), 4 h (1300 h), and 6 h (1500 h) after the initiation of breakfast for the measurement of circulating concentrations of TG, NEFA, 3-OHB, insulin, and glucose. NEFA was measured as a surrogate marker of substrate delivery to the liver for TG synthesis and 3-OHB as a marker of hepatic fatty acid oxidation affecting TG secretion. In addition, we used insulin and glucose as indicators of carbohydrate metabolism.

Test meal

Breakfast consisted of grilled salmon, scrambled eggs (egg and salts with tomato ketchup), a bowl of white rice, soup (made with soybean curd, seaweed, soybean paste, and deep-fried soybean curd), vegetables (sauteed cabbage and carrot with ham, steamed potato and broccoli, and fresh cucumber with a mayonnaise dressing), and a cream cracker. Breakfast provided 0.45 g fat, 1.39 g carbohydrate, 0.46 g protein, and 48.7 kJ (11.6 kcal) energy per kilogramme of body mass. For the physical activity group, mean macronutrient content of the breakfast was 25.5 ± 5.1 g fat, 79.0 ± 15.7 g carbohydrate, and 26.1 ± 5.2 g protein, which provided 2.8 ± 0.6 MJ (663 ± 132 kcal) energy (35% from fat, 49% from carbohydrate, and 16% from protein). For the control group, mean macronutrient content of the breakfast was 24.9 ± 2.7 g fat, 77.2 ± 8.4 g carbohydrate, and 25.5 ± 2.8 g protein, which provided 2.7 ± 0.3 MJ (648 ± 70 kcal) energy (35% from fat, 49% from carbohydrate, and 16% from protein). Participants were asked to consume breakfast within 30 min, and consumption time was recorded and replicated in subsequent postprandial blood tests (i.e., at 4 and 12 weeks). Mean time to consume breakfast of physical activity and control groups was 20.1 ± 4.7 and 20.3 ± 4.6 min, respectively. None of the participants reported nausea or any gastrointestinal discomfort during or after breakfast. Participants consumed water ad libitum during the baseline postprandial blood test, and the pattern and volume ingested were replicated in subsequent postprandial blood tests (i.e., at 4 and 12 weeks). The average water intake of the physical activity and control groups was 765 ± 187 and 690 ± 296 mL over 6 h, respectively.

Standardization of dietary intake and physical activity

Participants weighed and recorded all food and drink consumed the day before the baseline postprandial blood test and refrained from drinking alcohol during this period. Participants replicated their dietary intake from the baseline postprandial blood test in all subsequent postprandial blood tests (i.e., one day before the postprandial blood test at 4 weeks and 12 weeks after each intervention was completed) to ensure that dietary intake was standardized across three postprandial blood tests. Food diaries were analyzed by a registered dietitian to determine energy intake and macronutrient content. In addition, all participants were asked to remain inactive the day before each postprandial blood test and wore a uniaxial accelerometer (LifeCodeX; Suzuken Co Ltd, Nagoya, Japan) on the hip to monitor their daily activity objectively during this period. This time phase has been suggested to distinguish from acute physical activity-induced TG lowering effects. Thus, this practise ensures us to examine the “true” effect of chronic physical activity on postprandial TG metabolism.

Analytical methods

For serum T-C, LDL-C, HDL-C, TG, NEFA, and 3-OHB measurements, venous blood samples were collected into tubes containing clotting activators for isolation of serum (Venoject 2; Terumo Corporation, Tokyo, Japan). Thereafter, samples were allowed to clot for 30 min at room temperature and then centrifuged at 1861 g for 10 min at 4 °C. Serum was removed, divided into aliquots, and stored at −80 °C for later analysis. For plasma insulin measurements, venous blood samples were collected into tubes containing disopropylamine sodium salt-EDTA (Venoject 2; Terumo Corporation, Tokyo, Japan). For plasma glucose measurements, venous blood samples were collected into tubes containing sodium fluoride-EDTA (Venoject 2; Terumo Corporation, Tokyo, Japan). Thereafter, both tubes were immediately centrifuged and the plasma supernatants were stored at −80 °C for later analysis. Enzymatic colourimetric assays were used to measure serum T-C (T-cho Determiner L TC II; Hitachi Chemical Diagnostics Systems Co Ltd, Tokyo, Japan), serum LDL-C (Matabo Lead LDL-C; Hitachi Chemical Diagnostics Systems Co Ltd, Tokyo, Japan), serum HDL-C (Matabo Lead HDL-C; Hitachi Chemical Diagnostics Systems Co Ltd, Tokyo, Japan), serum TG (Pure Auto S TG–N; Sekisui Medical Co Ltd, Tokyo, Japan), serum NEFA (NEFA–HR; Wako Pure Chemical Industries, Ltd, Osaka, Japan), serum 3-OHB (KAINOS 3-HB; Kainos Laboratories, Inc, Tokyo, Japan), and plasma glucose (GLU–HK(M); Shino-Test Corporation, Kanagawa, Japan). Enzyme-linked immunosorbent assays (ELISAs) were used to measure plasma insulin (Mercodia Insulin ELISA; Mercodia AB, Uppsala, Sweden). Samples for each participant were analyzed within the same run for each measure. The intra-assay coefficients of variation were 0.62% for T-C, 0.75% for LDL-C, 1.16% for HDL-C, 0.60% for TG, 0.90% for NEFA, 1.40% for 3-OHB, 0.05% for insulin, and 0.92% for glucose.

Statistical analysis

Data were analyzed using the Predictive Analytics Software version 26.0 for Mac (IBM SPSS Statistics 26.0, SPSS Japan Inc., Japan). The Shapiro–Wilk test was used to check for normality of distribution. Normally distributed values were compared between groups using Welch’s t-test. The other values, which were found not to be normally distributed, were compared between groups using the Mann–Whitney U test. Time-averaged total area under the serum or plasma concentration versus time curves (AUC) were calculated using the trapezium rule. The linear mixed model was used to examine between-group differences over the 12-week intervention for body mass, physical activity levels, fasting serum or plasma concentration, and time-averaged AUC values. Time-averaged insulin AUC values were also analyzed by the fasting plasma insulin concentration and time–averaged AUC value at the baseline modelled as a covariate. Where significant group by week interactions and group and weekly effects were found (i.e., 6-h postprandial blood test at baseline, 4 weeks, and 12 weeks in two
groups), the data were subsequently analyzed using post-hoc analysis using the Bonferroni method. Single regression analysis was conducted to examine the association between changes in postprandial time-averaged TG AUC and changes in total physical activity (i.e., minutes/day) at 4 weeks and 12 weeks. Data are expressed as mean ± SD. Statistical significance was set at p < 0.05.

Results

Physical characteristics

There were no differences in terms of participant age, height, body mass, BMI, systolic diastolic blood pressure, or diastolic blood pressure between the physical activity and control groups at baseline (Table 1).

Physical activity data

The body mass and physical activity data over the 12-week intervention in the physical activity and control groups are shown in Table 2. There were no significant main effects of group (p = 0.714), week (p = 0.054), or an interaction (group × week, p = 0.248) on body mass. According to the daily logs, the participants in the physical activity group commonly engaged in walking, going up and down the stairs, and performing household chores over 12-week intervention. There were no differences in terms of step counts (p = 0.236), time spent in total physical activity (p = 0.247), time spent in moderate to vigorous (≥3 METs) physical activity (p = 0.204), or time spent in light (<3 METs) physical activity (p = 0.231) at baseline between the groups. On average over the 12-week intervention, step counts increased by 139 ± 1748 steps/day in the physical activity group and by 525 ± 1668 steps/day in the control group. There were no significant main effects of group (p = 0.287), week (p = 0.570), or an interaction (group × week, p = 0.401). On average over the 12-week intervention, time spent in the physical activity increased by 11 ± 19.3 min/day in the physical activity group and by 3.3 ± 15.3 min/day in the control group. There were no significant main effects of group (p = 0.216), week (p = 0.572), or an interaction (group × week, p = 0.312). On average, over the 12-week intervention, time spent in moderate (3–6 metabolic equivalents (METs)) to vigorous (>6 METs) physical activity increased by 2.8 ± 8.8 min/day in the physical activity group and by 4.9 ± 11.2 min/day in the control group. There were no significant main effects of group (p = 0.636), week (p = 0.055), or an interaction (group × week, p = 0.686). On average, over the 12-week intervention, time spent in light (<3 METs) physical activity increased by 1.7 ± 15.1 min/day in the physical activity group and by 1.6 ± 11.3 min/day in the control group. There was no significant main effect of group (p = 0.088), week (p = 0.088), or an interaction (group × week, p = 0.312).

Table 2

| Physical activity group (n = 11) | Control group (n = 15) | Interaction (group × week) p value |
|---------------------------------|-----------------------|---------------------------------|
| Body mass (kg)                  |                       |                                 |
| Baseline                        | 57.4 (11.8)           | 57.7 (11.8)                     | 0.248                           |
| 4 weeks                         | 57.9 (11.7)           | 56.1 (7.0)                      |                                 |
| 8 weeks                         | 56.2 (6.9)            | 56.4 (6.7)                      |                                 |
| 12 weeks                        | 56.6 (6.6)            |                                 |                                 |
| Steps (steps/day)               | 7646 (2840)           | 8120 (2354)                     | 0.401                           |
| 4 weeks                         | 7676 (1793)           | 7559 (2004)                     |                                 |
| 8 weeks                         | 6250 (2595)           | 6566 (2743)                     |                                 |
| 12 weeks                        | 6929 (2955)           | 6830 (3327)                     |                                 |
| Total PA (min/day)              | 80 (29)               | 85 (24)                         |                                 |
| 4 weeks                         | 79 (17)               | 77 (18)                         |                                 |
| 8 weeks                         | 66 (28)               | 68 (24)                         |                                 |
| 12 weeks                        | 71 (26)               | 69 (30)                         | 0.312                           |
| ≥3 METs PA (min/day)            | 20 (17)               | 23 (12)                         |                                 |
| 4 weeks                         | 23 (10)               | 16 (16)                         |                                 |
| 8 weeks                         | 18 (18)               | 21 (19)                         |                                 |
| 12 weeks                        | 22 (21)               |                                 |                                 |
| <3 METs PA* (min/day)           | 59 (15)               | 63 (14)                         | 0.686                           |
| 4 weeks                         | 56 (11)               | 54 (9)                          |                                 |
| 8 weeks                         | 50 (17)               | 49 (13)                         |                                 |
| 12 weeks                        | 50 (13)               | 47 (16)                         | 0.312                           |

Values are means (standard deviation). Means were compared using linear mixed model. METs; metabolic equivalents; PA; physical activity. * a minimum threshold is 1.8 METs.
Table 3
Fasting biochemical and hormonal variables at baseline, 4 weeks, 8 weeks, and 12 weeks in the physical activity and control groups.

| Variable                          | Physical activity group (n = 11) | Control group (n = 15) | Interaction (group × week) p value |
|-----------------------------------|---------------------------------|------------------------|-----------------------------------|
|                                  | Baseline 4 weeks 8 weeks 12 weeks | Baseline 4 weeks 8 weeks 12 weeks |                                  |
| Total cholesterol (mmol/L)       | 6.09 (1.19) 6.24 (1.28) 6.28 (1.16) 6.30 (1.26) | 5.92 (0.68) 5.96 (0.77) 5.94 (0.66) 6.02 (0.89) | 0.544 |
| LDL-cholesterol (mmol/L)         | 3.58 (1.00) 3.62 (0.97) 3.62 (1.02) 3.67 (1.02) | 3.47 (0.58) 3.43 (0.70) 3.27 (0.56) 3.45 (0.72) | 0.430 |
| HDL-cholesterol (mmol/L)         | 1.89 (0.62) 1.87 (0.65) 1.98 (0.64) 2.03 (0.66) | 1.83 (0.42) 1.94 (0.49) 2.01 (0.44) 2.03 (0.50) | 0.182 |
| Triglyceride (mmol/L)            | 1.11 (0.51) 1.55 (0.97) 1.34 (0.68) 1.18 (0.54) | 1.02 (0.51) 0.99 (0.51) 0.93 (0.47) 0.97 (0.58) | 0.004* |
| NEFA (mmol/L)                    | 0.80 (0.26) 0.68 (0.23) 0.73 (0.20) 0.74 (0.19) | 0.81 (0.22) 0.86 (0.42) 0.69 (0.21) 0.75 (0.18) | 0.108 |
| Glucose (mmol/L)                 | 5.62 (0.58) 5.57 (0.57) 5.52 (0.44) 5.50 (0.45) | 5.56 (0.44) 5.46 (0.47) 5.50 (0.39) 5.43 (0.56) | 0.938 |
| Insulin (µIU/mL)                 | 5.12 (2.50) 5.83 (2.77) 5.56 (3.22) 5.34 (1.97) | 5.41 (8.93) 3.54 (1.06) 0.10 (0.11) 0.13 (0.12) | 0.507 |

Values are means (standard deviation). Means were compared using linear mixed model and post-hoc analysis was adjusted for multiple comparisons using the Bonferroni method. * main effect of week for NEFA (p < 0.001) and TG (p = 0.015). Post-hoc analysis revealed that the fasting serum TG concentration at 4 weeks was significantly higher in the physical activity group than in the control group (p = 0.024). HDL; high-density lipoprotein, LDL; low-density lipoprotein, NEFA; non-esterified fatty acids, 3-OHB; 3-hydroxybutyrate.

The relationship between increments of physical activity and serum TG concentrations in the postprandial state

Physical activity was not increased over the 12-week intervention in either group and there was a large variation in physical activity between individuals. The changes in total physical activities ranged from −31 to 40 min per day at 4 weeks and from −34 to 28 min per day at 12 weeks. Therefore, all data was pooled together and the association between changes in postprandial TG AUC and changes in total physical activity time was examined. No associations between changes in postprandial TG AUC and changes in total physical activity time were observed at 4 weeks (r² = 0.017, p = 0.531) or 12 weeks (r² = 0.037, p = 0.445).

Dietary data – a day before the postprandial tests

Reported energy intake for the 24 h preceding postprandial testing was 7.7 ± 1.4 MJ/day (1832 ± 327 kcal/day) (14 ± 4% from fat, 70 ± 5% from carbohydrate and 16 ± 3% from protein) in the physical activity group and 7.3 ± 1.2 MJ/day (1735 ± 280 kcal/day) (15 ± 5% from fat, 68 ± 7% from carbohydrate and 17 ± 4% from protein) in the control group.

Discussion

The present study examined whether an increase in self-selected physical activity for at least 10 min per day over 12 weeks under free-living conditions altered postprandial TG concentration in postmenopausal women. The present study demonstrated that there was no reduction in postprandial TG concentration, with no meaningful averaged increment in physical activities over the 12-weeks investigation. In addition, the present study found that there was no association between changes in postprandial TG concentration and changes in physical activity time (range: −34 to 40 min per day) when these responses were determined 24 h after the last physical

intervention (p = 0.002) and among the weeks (p = 0.008). Post-hoc analysis revealed that the time-averaged plasma insulin AUC was significantly higher in the physical activity group than in the control group. The time-averaged plasma insulin AUC was significantly higher at 4 weeks than at baseline (p = 0.027) and 12 weeks (p = 0.021). There was no significant interaction (group × week, p = 0.776). On average, the difference between the groups over the 12-weeks intervention remained significant after adjusting for fasting insulin concentration at baseline as a covariate (p = 0.002). However, after adjusting for time-averaged plasma insulin AUC at the baseline as a covariate, the difference between the groups was no longer significant (p = 0.183).

Fig. 2. The time-averaged serum triglyceride (TG) area under the curve (AUC) value over 6 h after the consumption of the test meal at baseline, 4 weeks, and 12 weeks in the physical activity and control groups.

Table 4
The time-averaged serum triglyceride, serum non-esterified fatty acids (NEFA), serum 3-hydroxybutyrate (3-OHB), plasma insulin, and plasma glucose area under the curve (AUC) values over 6 h after the consumption of the test meal at baseline, 4 weeks, and 12 weeks in the physical activity and control groups.

| Variable                          | Physical activity group | Control group | Interaction (group × week) p value |
|-----------------------------------|-------------------------|---------------|-----------------------------------|
|                                  | Baseline 4 weeks 12 weeks | Baseline 4 weeks 12 weeks |                                  |
| NEFA AUC (mmol/L)                 | 0.45 (0.11) 0.39 (0.08) | 0.45 (0.08) 0.44 (0.12) 0.39 (0.11) 0.41 (0.08) | 0.960 |
| Glucose AUC (mmol/L)              | 5.90 (0.61) 5.90 (0.61) | 5.88 (0.64) 6.19 (0.60) 6.00 (0.60) 5.98 (0.68) | 0.881 |
| Insulin AUC* (µIU/mL)             | 17.3 (6.5) 21.7 (10.5) | 17.7 (9.2) 9.6 (5.5) 12.7 (6.3) 8.1 (3.4) | 0.776 |
| 3-OHB AUC (mmol/L)                | 0.06 (0.04) 0.04 (0.03) | 0.05 (0.05) 0.03 (0.03) 0.06 (0.03) | 0.581 |

Values are means (standard deviation). Means were compared using linear mixed model and post-hoc analysis was adjusted for multiple comparisons using the Bonferroni method. * main effect of week for NEFA (p = 0.002), insulin (p = 0.008) and 3-OHB (p = 0.010).
activity bout. This finding supports the findings of previous studies examining the chronic (≥4 weeks) effect of structured exercise training on postprandial TG concentrations.\textsuperscript{8–11,13,14} Furthermore, our findings support the importance of increased “frequent” physical activity in daily life for gaining the postprandial TG lowering effects. Indeed, our previous study highlighted the effectiveness of acute physical activity in reducing postprandial TG concentration.\textsuperscript{2}

No improvement in postprandial TG is consistent with results of previous studies in young,\textsuperscript{10,14} middle-aged,\textsuperscript{9,12} and older\textsuperscript{11} adults when the postprandial measurements were conducted 24–72 h after the last exercise bout. The present study extends these findings by demonstrating that no persisting effect of reduction in postprandial TG concentrations was observed through activities of daily living in postmenopausal women. Furthermore, our findings support findings of our previous study which showed that postprandial TG concentration was not reduced in postmenopausal women after participants performed self-selected activities under free-living conditions over a 4-weeks period, despite the participants in the active group increasing their step counts by 600 steps per day and their total physical activity by 5 min per day.\textsuperscript{12} In contrast, another study demonstrated reductions in postprandial TG concentrations after 11 weeks of high-volume resistance exercise in postmenopausal women.\textsuperscript{15} However, the postprandial measurements were conducted 15 h after the last exercise bout in the study by Correa et al.\textsuperscript{13} Therefore, caution is required when interpreting the findings of this study as reductions in postprandial TG concentrations by exercise are transient (i.e., the exercise-induced reduction in postprandial TG was diminished when exercise was performed for 24 h, but not 12 h prior to the postprandial measurements). Taken together, the present study supports the notion that physical activity-induced lowering of postprandial lipemia is short-lived and is reduced by the short period of detraining.\textsuperscript{9,24,25}

The majority of previous studies examining the chronic effect of physical activity on postprandial lipemia are conducted under “structured or supervised” conditions.\textsuperscript{8–11,13–15} These previous interventions administered under the supervision of experimenters have shown effectiveness in increasing physical activity levels or physical fitness.\textsuperscript{3–11,13–15} However, a higher dropout rate (roughly 50%) from supervised exercise programmes has been reported, and a lower long-term adherence to physical activity has been reported when the intervention was terminated.\textsuperscript{10,16} Given the importance of increasing or maintaining physical activities in daily life after the intervention, it is necessary for the participants to choose their activities and be easily performed in their free-living condition. The health commission report of Japan has shown that an increment of 1 MET-h/week results in a 0.8% reduction in the average relative risk for non-communicable diseases (including cardiovascular diseases), and this reduction could be interpreted as a 3.2% risk reduction with a 10-min increase in physical activity per day.\textsuperscript{17} For this reason, the Ministry of Health, Labour and Welfare of Japan recommends that people incorporate “≥10 min of physical activity” per day into their daily lives to promote physical activity for public health.\textsuperscript{18} The present study was performed in accordance with this guidance and the participants in the physical activity group were instructed to increase their activities for at least 10 min per day above their usual lifestyle levels for 12 weeks. However, the average increment of total physical activity time spent in physical activity was only 1.1 min per day in the physical activity group. Therefore, the small increment in the amount of physical activity observed in the present study may be the main reason for the lack of improvement in fasting blood lipid parameters.\textsuperscript{26–28} In the present study, the investigators instructed the participants in the physical activity group to reduce their inactivity time and to increase their physical activity in accordance with their own lifestyle by engaging in activities such as taking a walk, going up and down the stairs, playing with grandchildren, and running. Thus, we did not set a target for physical activity intensity or a targeted number of steps. Alternatively, the number of steps is an objective measurement of physical activity that can be easily measured and evaluated on a daily basis for many individuals using pedometer and smartphone applications as recommended for promoting physical activity.\textsuperscript{29,30}

To obtain better adherence to daily physical activity in older adults, an alternative intervention plan where individuals can visually check the amount of physical activity may be needed.

The present study has two limitations. Although each participant was asked to standardize their dietary intake one day before each postprandial blood test and was asked not to alter their diet over the 12 weeks, we had no data on their dietary intake during the entire experimental period. Thus, such a background diet may explain why fasting TG concentration and postprandial TG AUC at 4 weeks was significantly higher in the physical activity group than in the control group ($p = 0.024$ and $p = 0.056$, respectively) as exercise with a short-term (i.e., 4 days) dietary manipulation affected postprandial TG concentrations.\textsuperscript{11} In addition, although there was no statistically significant difference between the groups, there was a difference in physical activity levels at baseline between the groups, with women\textsuperscript{15} physical activity group having -1400 steps/day and 14 min more total physical activity compared to the control group. This difference may partly explain why the intervention did not work well in the present study, as the physical activity group could not increase their physical activity across the intervention. However, this initial variation in physical activity levels did not appear to affect postprandial TG concentrations, since there was no association between the changes in postprandial time-averaged TG AUC and changes in the total physical activity levels.

Conclusion

The present study demonstrated that self-selected activities performed under free-living conditions lasting for 12 weeks did not improve postprandial TG metabolism in postmenopausal women with no meaningful averaged increase in physical activities. The present study also observed that there was no association between changes in postprandial time-averaged TG AUC and changes in duration of physical activity when these responses were determined 24 h after the last bout of physical activity. Given that only recent physical activity appears to facilitate the physical activity-induced postprandial TG lowering effects,\textsuperscript{19} the findings of the present study imply that “regular” physical activity through an active lifestyle is important to keep circulating concentrations of postprandial TG low in postmenopausal women.

Author contribution

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Declaration of competing interest

None.

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References

1. Zilversmit DB. Atherogenesis: a postprandial phenomenon. Circulation. 1979;60(3):473–485. https://doi.org/10.1161/01.cir.60.3.473.
2. Iso H, Imano H, Yamagishi K, et al. Fasting and non-fasting triglycerides and risk of ischemic cardiovascular disease in Japanese men and women: the Circulatory Risk in Communities Study (CIRCS). Atherosclerosis. 2014;237(1):361–368. https://doi.org/10.1016/j.atherosclerosis.2014.08.028.
3. Nordestgaard BC, Vatbo A. Triglycerides and cardiovascular disease. Lancet. 2014;384(9943):626–635. https://doi.org/10.1016/S0140-6736(14)61777-6.
4. Freese EC, Gist NH, Cureton KJ. Effect of prior exercise on postprandial lipemia: an updated quantitative review. J Appl Physiol. 2014;116(1):67–75. https://doi.org/10.1152/japplphysiol.00623.2013.
5. Miyashita M, Takahashi M, Burns S. Increased participation in weekend physical activity reduces postprandial lipemia in postmenopausal women. Int J Sports Med. 2014;35(13):1059–1064. https://doi.org/10.1055/s-0034-1372641.
6. Ministry of Health, Labour and Welfare of Japan. The national health and nutrition survey ([in Japanese]). https://www.mhlw.go.jp/content/000681200.pdf. Accessed December 3, 2020.
7. Department of Health and Human Services. Physical Activity Guidelines for Americans. second ed.; 2018. https://health.gov/sites/default/files/2019-09/Physical_Activity_Guidelines_2nd_edition.pdf. Accessed December 3, 2020.
8. Wirth A, Diehm C, Hanel W, Welte J, Vogel I. Training-induced changes in serum lipids, fat tolerance, and adipose tissue metabolism in patients with hypertriglyceridemia. Atherosclerosis. 1985;54(3):263–271. https://doi.org/10.1016/0021-9150(85)90120-0.
9. Aldred HE, Hardman AE, Taylor S. Influence of 12 weeks of training by brisk walking on postprandial lipemia and insulinemia in sedentary middle-aged women. Metabolism. 1995;44(3):390–397. https://doi.org/10.1016/0026-0495(95)90172-8.
10. Herd SL, Hardman AE, Boobis LH, Cairns CJ. The effect of 13 weeks of running training followed by 9 d of detraining on postprandial lipemia. Br J Nutr. 1998;80(1):57–66. https://doi.org/10.1079/000711498661779.
11. Paton CM, Brandauer J, Weiss EP, et al. Hemostatic response to postprandial lipemia before and after exercise training. J Appl Physiol. 2006;101(1):316–321. https://doi.org/10.1152/japplphysiol.01361.2005.
12. Edamoto K, Jung H-H, Kidozako T, et al. Effect of increased physical activities of daily living on postprandial triacylglycerol concentrations in postmenopausal women. Jpn J Phys Fit Sports Med. 2015;64(5):485–492. https://doi.org/10.7800/jgfpm.64.485 ([in Japanese, English abstract]).
13. Freese EC, Gist NH, Acitelli RM, et al. Acute and chronic effects of sprint interval exercise on postprandial lipemia in women at-risk for the metabolic syndrome. J Appl Physiol. 1985;118(7):872–879. https://doi.org/10.1152/japplphysiol.00380.2014.
14. Wilhelmsen L, Mallinson J, Jones R, Cooper S, Taylor T, Totszan T. Chronic effects of high-intensity interval training on postprandial lipemia in healthy men. J Appl Physiol. 1985;127(6):1763–1771. https://doi.org/10.1152/japplphysiol.00131.2019.
15. Correa CS, Teixeira RC, Cobos RCR, et al. High-volume resistance training reduces postprandial lipemia in postmenopausal women. J Sports Sci. 2015;33(18):1890–1901. https://doi.org/10.1080/02640424.2015.1017732.
16. Zhang JQ, Ji LL, Nunez G, Feathers S, Hart CL, Wan XY. Effect of exercise timing on postprandial lipemia in hypertriglyceridemic men. Can J Appl Physiol. 2004;29(5):590–603. https://doi.org/10.1139/h04-038.
17. Ministry of Health, Labour and Welfare of Japan. Physical activity reference for health promotion. Accessed December 3, 2020. [in Japanese] https://www.mhlw.go.jp/stf/houdou/20a52500000e2xett%28%2525000002%2xpat.pdf. 2013.
18. Ministry of Health, Labour and Welfare of Japan. Active Guide - Japanese official physical activity guidelines for health promotion. http://www.nibohon.go.jp/esen/ki/pdf/active2013-e.pdf. 2013. Accessed December 3, 2020.
19. Dishman RK. Increasing and maintaining exercise and physical activity. Behav Ther. 1991;22(3):345–378. https://doi.org/10.1016/0005-7944(95)0371-5.
20. Dishman RK. Motivating older adults to exercise. South Med J. 1994;87(5):579–582.
21. Emerson SR, Kurihara SD, EMerson EM, et al. Postprandial metabolic responses differ by age group and physical activity level. J Nutr Health Aging. 2018;22(1):145–153. https://doi.org/10.1016/j.jnha.2017.07.006.
22. Murakami H, Irituspe J, Kawakami R, Muzhi M. “Add 10 min for your health”: the new japanese recommendation for physical activity based on dose-response analysis. J Am Coll Cardiol. 2015;65(15):1135–1154. https://doi.org/10.1016/j.jacc.2014.10.080.
23. Kumahara H, Schutz Y, Ayabe M, et al. The use of uniaxial accelerometry for the assessment of physical-activity-related energy expenditure: a validation study against whole-body indirect calorimetry. Br J Nutr. 2004;91(2):235–243. https://doi.org/10.1079/bjn20031032.
24. Gill JMR, Caslake MJ, McAllister C, et al. Effects of short-term detraining on postprandial metabolism, endothelial function, and inflammation in endurance-trained men: dissociation between changes in triglyceride metabolism and endothelial function. J Clin Endocrinol Metab. 2003;88(9):4328–4335. https://doi.org/10.1210/jc.2003-030226.
25. Hardman AE, Lawrence JEM, Herd SL. Postprandial lipemia in endurance-trained people during a short interruption to training. J Appl Physiol. 1998;84(6):1895–1901. https://doi.org/10.1152/japplphysiol.1998.84.6.1895.
26. Durstine JL, Grandjean PW, Davis PG, Ferguson MA, Alderson NL, DuBose KD. Blood lipid and lipoprotein adaptations to exercise: a quantitative analysis. Sports Med. 2001;31(15):1033–1062. https://doi.org/10.2165/00007256-200131150-00002.
27. Igarashi Y, Akazawa M, Maeda S. Effects of aerobic exercise alone on lipids in healthy east asians: a systematic review and meta-analysis. J Atherosclerosis Thromb. 2019;26(5):488–503. https://doi.org/10.5531/jat.45864.
28. Kodama S, Tanaka S, Saito K, et al. Effect of aerobic exercise training on serum levels of high-density lipoprotein cholesterol: a meta-analysis. Arch Intern Med. 2007;167(10):1099–1108. https://doi.org/10.1001/archinte.167.10.1099.
29. Ministry of Health, Labour and Welfare of Japan. The second term of national health promotion movement in the twenty first century (health Japan 21 (the second term). http://www.mhlw.go.jp/file/06-Seisakouhoujo-1990000-kenkouyouko/1990000047330.pdf. 2012. Accessed December 3, 2020.
30. Department of Health and Social Care. UK chief medical officers’ physical activity guidelines. https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/832868/uk-chief-medical-officers-physical-activity-guidelines.pdf. 2019. Accessed December 3, 2020.
31. Burns SF, Hardman AE, Stensel DJ. Brisk walking offsets the increase in postprandial TAG concentrations found when changing to a diet with increased carbohydrate. Br J Nutr. 2009;101(12):1787–1796. https://doi.org/10.1017/S0007114508133591.