1   Full title: Structured, aerobic exercise reduces fat mass and is partially compensated through

2   energy intake but not energy expenditure in women

3   Short title: Compensatory EI and EE after structured exercise in women

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23 **ABSTRACT**

24 **Background**

25 Exercise-induced weight loss is often less than expected and highly variable in men and

26 women. Behavioural compensation for the exercise-induced energy deficit could be through

27 energy intake (EI), non-exercise physical activity (NEPA) or sedentary behaviour (SB). We

28 investigated this issue in women.

29 **Methods**

30 Twenty-four overweight [body mass index (BMI) M=27.9 kg/m^2, SD=2.7] women [age

31 M=33.1 years, SD=11.7] completed 12-weeks of supervised exercise (5x500kcal per week) in

32 a non-randomised pre-post intervention study. Body mass (BM), waist circumference (WC),

33 body composition, resting metabolic rate (RMR), total daily EI, individual meals, appetite

34 sensations and appetite-related peptides were measured at baseline (week 0) and post-

35 intervention (week 12). Free-living physical activity (PA) and SB were measured (SenseWear)

36 at baseline, week 1 and 10 of the exercise intervention, and at post-intervention (week 13).

37 **Results**

38 Following the 12-week exercise intervention BM [p=.04], BMI [p=.035], WC [p<.001] and fat

39 mass (FM) [p=.003] were significantly reduced, and fat-free mass (FFM) significantly

40 increased [p=.003]. Total [p=.028], *ad libitum* [p=.03] and snack box EI [p=.048] were

41 significantly increased and this was accompanied by an increase in hunger [p=.01] and a

42 decrease in fullness [p=.03] before meals. The peptides did not explain changes in appetite

43   [p>.05]. There was no compensatory reduction in NEPA [p>.05] and no increase in SB, rather

44   there was a decrease in SB during the exercise intervention [p=.03].

45   **Conclusions**

46   Twelve-weeks of supervised aerobic exercise resulted in a significant reduction in FM and an

47   increase in FFM. Exercise increased hunger and EI which only partially compensated for the

48   increase in energy expenditure. There was no evidence for a compensatory reduction in NEPA

49   or an increase in SB. Dietary intervention, as an adjunct to exercise, may offset the

50   compensatory increase in EI and result in a greater reduction in BM.

51   **Trial registration**

52   Our trial was retrospectively registered on the International Standard Randomised Controlled

53   Trials Registry (ISRCTN78021668, 27th September 2016) and can be found here:

54   https://doi.org/10.1186/ISRCTN78021668

55   **KEY WORDS**

56   Exercise, appetite control, weight loss, compensation, non-exercise physical activity, sedentary

57   behaviour

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BACKGROUND

There is much discussion about the role of physical activity (PA) and/or exercise for reducing obesity and promoting weight maintenance. The scepticism surrounding the efficacy of PA for weight management arises from the observation that weight loss as a result of exercise interventions is often less than expected (1) and the belief that increased exercise-induced energy expenditure (EE) is automatically countered by an increase in energy intake (EI) (2). Despite this, observational studies demonstrate that habitual PA is associated with lower body mass (BM) and fat mass (FM) (3, 4). Furthermore, experimental studies have shown that structured exercise results in reduced BM and FM, often with an increase or preservation of fat-free mass (FFM) (5-7). Exercise and/or PA is also a strong predictor of weight loss maintenance (8). The evidence demonstrates that exercise is an integral component of weight management interventions (5).

Despite significant reductions in average BM and FM with exercise, weight loss is often less than the theoretically predicted reduction based on the exercise-induced EE, even when adherence to the exercise intervention is strictly supervised and monitored and compliance is high (1, 7). This less than theoretically predicted weight loss could, in part, be due to the use of overly simplistic and static predictive equations that do not account for dynamic physiological adaptations to weight loss and therefore overestimate the weight loss resulting from a particular exercise-induced energy deficit (9). Additionally, compensation in response to the energy deficit generated by the exercise regime would attenuate weight loss. This compensation could arise through an increase in EI (7, 10), or compensation that acts to reduce total daily EE such as a decrease in non-exercise physical activity (NEPA) or an increase in sedentary behaviour (SB) (or subtle combinations of all these components of energy balance) (11, 12). The literature regarding changes in EI, NEPA and SB in response to structured

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84   exercise is conflicting and many studies lack accurate and reliable measures of EI, EE, NEPA

85   and SB (13, 14).

86   This study applied objective methodology to assess the influence of an exercise regime on EI

87   (food intake, appetite sensations and appetite-related peptides) and EE (PA and SB outside of

88   the structured exercise) in women. The specific objective was to examine whether a 12-week

89   supervised, structured aerobic exercise regime generated compensation through appetite,

90   NEPA or SB.

91   **METHODS**

92   **Participants**

93   Thirty-two overweight or obese inactive women were recruited to take part in the study. Only

94   women were recruited to reduce unwanted variability in the design. Of those 32 participants,

95   24 women aged 33.1 years (SD = 11.7) with a body mass index (BMI) of 27.9 kg/m^2 (SD =

96   2.7) completed the study. The following reasons were given for participant dropouts: did not

97   like exercise (week 1; n=1); exercise related injury (week 4; n=1); did not comply with

98   procedures (week 4; n=1); personal reasons (week 6; n=1); no reason provided (week 7; n=1);

99   time commitment of exercise too much (week 10; n=2); illness (week 12; n=1). Participants

100   were recruited from the University of Leeds, UK, and surrounding area using posters and email

101   mailing lists. An online screening survey was completed to assess the eligibility of potential

102   participants based on the following criteria: women aged 18-55 years, BMI between 25.0 and

103   34.9 kg/m^2, not currently dieting to lose weight, inactive (less than 150 min/week of moderate-

104   to-vigorous PA (MVPA) assessed by questionnaire), no increase in PA in previous four weeks,

105   weight stable (no significant weight loss (≥5%) in the previous 6 months), non-smokers, not

106   taking any medication or have any medical condition known to affect metabolism or appetite,

107   and acceptance of the study foods (≥3 liking of study foods on 7-point Likert scale). All

108   participants provided written informed consent before taking part in the study. The study

109   procedures and all study materials were reviewed and approved by the National Research

110   Ethics Service Committee Yorkshire & the Humber (ref: 09/H1307/7).

111   **Design**

112   This study was a non-randomised pre-post study with a 12-week supervised aerobic exercise

113   intervention. Anthropometrics, body composition and resting metabolic rate (RMR) were

114   taken before (week 0) and at the end of the exercise intervention (week 12). Participants also

115   completed two probe days prior to the exercise intervention (week 0) commencing and two in

116   the final week (week 12) of the exercise intervention to assess eating behaviour and

117   subjective appetite sensations. On both measures and probe days, the participants arrived at

118   the research unit between 07:00 and 09:00 following a 10 hour fast (no food or drink except

119   water). Free-living PA and SB were measured before (week -1), during (week 1 and week 10)

120   and after (week 13) the intervention.

121   **Measures days**

122   A range of measurements were performed at week 0 (baseline) and week 12. Participants

123   arrived at the laboratory following an overnight fast. RMR was measured (GEM, NutrEn

124   Technology Ltd, Cheshire, UK) with participants laying supine for 40 min during which

125   expired air was collected using a ventilated hood system. VO_2 and VCO_2 values were

126   sampled every 30 seconds. The average of the final 30 min values was deemed to be the

127   RMR expressed as kcal/d. BM and body composition (fat mass (FM) and fat-free mass

128 (FFM)) were measured using the BODPOD (Body Composition Tracking System, Life

129 Measurement, Inc., Concord, USA) which uses air displacement plethysmography.

130 Participants wore tight clothing and a swim cap to allow for an accurate measure of body

131 volume. Height was measured using a stadiometer (Seca Ltd., Birmingham, UK) and waist

132 circumference (WC) was measured horizontally in line with the umbilicus.

133 **Probe days**

134 Twenty-four hour EI and subjective appetite sensations were measured during the probe day

135 visits. Participants were provided with an individually fixed energy breakfast (25% of

136 measured RMR) of muesli and milk and a choice of tea, coffee or water and were instructed

137 to consume all food and drink within 10 min. The macronutrient composition of the breakfast

138 was fixed at 55%, 30% and 15% for carbohydrate, fat and protein, respectively. Participants

139 remained in the laboratory between breakfast and lunch and were able to use a desktop

140 computer/laptop, listen to music or read.

141 Four hours after breakfast, an *ad libitum* lunch consisting of chilli with rice, and strawberry

142 yoghurt with double cream was provided with water. Participant were then free to leave the

143 laboratory between lunch and dinner but were not allowed to consume any food or drink

144 except the bottle of water provided.

145 Participants returned to the laboratory four hours later for the *ad libitum* dinner of tomato and

146 herb risotto, garlic bread, salad items, chocolate brownies and water. An *ad libitum* snack box

147 containing an apple, two mandarins, roast ham, cheese, bread, margarine, crisps, chocolate

148 buttons and a vanilla yoghurt was given to participants to take home in the evening.

149 Participants could eat any food items from the snack box but were instructed not to share the

150   foods. Participants returned the snack box containing any uneaten foods and food packaging

151   the following day. All of the *ad libitum* meals were presented in excess of expected

152   consumption and participants were instructed to eat until they reached a comfortable level of

153   fullness. EI was calculated by weighing foods to the nearest 0.1 g before and after

154   consumption and using energy equivalents for protein, fat and carbohydrate of 4, 9 and 3.75

155   kcal/g, respectively, and nutritional information from the manufacturers' food labels.

156   During probe days visual analogue scales (VAS) were completed immediately before and

157   after meals and periodically between meals to assess subjective appetite sensations using a

158   validated electronic appetite rating system (15). Area under the curve (AUC) was calculated

159   using the trapezoid method for subjective feelings of hunger, fullness, desire to eat and

160   prospective foods consumption throughout the whole day (post-breakfast (0 min), +15 min,

161   +30 min, +60 min, +90 min, +120 min, +180 min, +230 min, pre-lunch (+235 min), post-

162   lunch (+260 min), +300 min, +360 min, +420 min, pre-dinner (+480 min), post-dinner (+500

163   min), +540 min, +600 min).

164   EI and subjective appetite sensations were averaged across the two baseline probe days and

165   the two post-intervention probe days to provide a single measure of EI and subjective appetite

166   sensations at both time points. Data were averaged in this way because, as part of a wider

167   project, the two probe days involved the consumption of a novel yoghurt or a calorie and

168   energy matched control yoghurt immediately after breakfast. As the two different yoghurts

169   had no effect on any of the outcome measures in this study, we included it as part of the total

170   breakfast intake and averaged the probe days at baseline and post intervention to give a more

171   robust pre and post intervention measure.

172   **Free-living physical activity, sedentary behaviour and energy expenditure**

173   Free-living PA, SB and EE were measured using the SenseWear Armband mini (SWA;

174   BodyMedia, Inc., Pittsburgh, PA), as has previously been described (3). Measures were

175   completed before the exercise intervention (week -1), week 1 and week 10 of the exercise

176   intervention and post-intervention (after the exercise intervention was complete; week 13).

177   Participants wore the SWA at all times apart from when showering, bathing or swimming, this

178   included wearing the SWA during structured exercise sessions. Participants wore the SWA on

179   the posterior surface of their upper non-dominant arm for a minimum of 22 hours/d for 7-8

180   days. The SWA measures motion (triaxial accelerometer), galvanic skin response, skin

181   temperature and heat flux. Proprietary algorithms available in the accompanying software

182   (SenseWear Professional software version 8.0, algorithm v5.2) calculate EE and classify the

183   intensity of activity. SB was classified as <1.5 METs, light 1.5-2.9 METs, moderate 3-5.9

184   METs and vigorous >6 METs (16). Moderate and vigorous PA was grouped together to form

185   one MVPA category to correspond with the guidelines for PA. Activity EE was calculated by

186   summing the energy expended in activities >1.5 METs. PA and SB variables were expressed

187   as average min/d and activity EE was expressed as average kcal/d by dividing the total min/d

188   or kcal/d recorded during the whole wear period by the number of days participants wore the

189   SWA. For a wear period to be valid there had to be ≥5 days of valid data (≥22 hours/d)

190   including ≥1 weekend day (17). The SWA has been shown to accurately estimate time spent

191   in sedentary, light and moderate activities, total EE, EE at rest and EE during free-living light

192   and moderate intensity PA (18-21).

193   **Non-exercise physical activity**

194   The duration of weekly prescribed exercise was averaged over 7 days for week 1 (M = 47.30

195   min/d, SD = 6.96) and week 10 (M = 40.16 min/d, SD = 5.83) of the exercise intervention.

196   Average structured exercise minutes per day was then subtracted from time spent in MVPA

197 per day measured using the SWA during week 1 and week 10 of the exercise intervention to

198 determine NEPA MVPA. Similarly, the five day exercise-induced EE (2500 kcal) was

199 averaged over 7 days (357.14 kcal/d) and subtracted from activity EE measured using the SWA

200 during week 1 and week 10 of the exercise intervention to determine NEPA activity EE.

201 **Exercise intervention**

202 Participants were required to exercise at the laboratory exercise facility five times per week for

203 12-weeks. Each exercise session was individually tailored to expend 500 kcal at 70% of their

204 HR maximum (2500 kcal/wk). Participants completed a maximal treadmill fitness test and

205 expired air was collected and analysed using indirect calorimetry (SensorMedics Vmax29,

206 California, USA) to calculate EE during exercise. Standard stoichiometric equations were used

207 with respiratory data (VO_2/VCO_2) to calculate the energy expended at 70% HR maximum (22).

208 To account for changes in fitness and BM, a further VO_2 max test was performed during week

209 six of the intervention to recalculate the exercise duration required to expend 500 kcal at 70%

210 HR maximum. Compliance with the exercise intervention was monitored and tracked daily

211 using HR monitors (S610, POLAR, Finland) to ensure the correct intensity and duration of

212 exercise was achieved. Participants could choose from a selection of exercise equipment:

213 bicycle ergometers, cross-trainers, rowing ergometers and treadmills. Participants could attend

214 the laboratory exercise facility between 7 am and 7 pm Monday – Friday. The facility could

215 accommodate up to 6 participants exercising at any one time. The target total EE over the 12-

216 week exercise intervention was 29,000 kcal for each participant. If participants missed an

217 exercise session for any reason they were required to make up the time they had missed by

218 exercising for longer on other days or exercising away from the laboratory over the weekend

219 providing they recorded their exercise session with the HR monitor. Participants were excluded

220   from the study on a case by case basis if they repeatedly missed exercise sessions and it was

221   deemed unrealistic to make up the exercise they had missed.

222   **Blood parameters**

223   Venous blood samples were collected into 10ml syringes and then transferred to EDTA-

224   containing Monovette tubes. These tubes contained a mixture of inhibitors (dipeptidyl

225   peptidase IV (DPP4) inhibitor (10µl/ml blood), aprotinin (50µl/ml blood) and pefabloc SC

226   (50µl/ml blood)) to prevent degradation of the peptides to be measured. Samples were drawn

227   at eight time points during the morning of the probe day at 0 min and after breakfast at +15

228   min, +30 min; +60 min; +90 min; +120 min; +180 min and +230 min for the measurement of

229   metabolic and appetite peptide levels. After collection, samples were centrifuged for 10

230   minutes at 4°C and 4000 rpm. Samples were immediately pipetted into Eppendorf tubes and

231   stored at -80°C awaiting analysis. Insulin, acylated ghrelin, peptide YY (PYY) and glucagon-

232   like peptide 1 (GLP-1) were analysed in this study. Total PYY was measured due to feasibility.

233   Because the overwhelming composition of circulating total PYY is known to be PYY3–36, the

234   present PYY (total) assay effectively measured PYY3–36. A previous study showed an

235   essentially perfect correlation between this PYY (total) assay and a PYY3–36 selective

236   radioimunoassay. The relevant antibodies for PYY (total) used in the present study (originally

237   from Linco, St. Charles, Missouri), have been used by others to demonstrate the effects of

238   PYY3–36 (23). The inter- and intra- assay coefficients of variations were 6.35% and 6.2% for

239   insulin, 3.81% and 5.3% for leptin, 4.24% and 4.05% for GLP-1, 4.91% and 5.9% for PYY

240   (total) and 5.12% and 4.45% for acylated ghrelin, respectively.

241   Only a subset of participants completed the postprandial blood samples. Reasons for missing

242   peptide data included unsuccessful cannulation, and participants' unwillingness to take part in

243   this part of the study. All samples that were drawn, were analysed and have been included in

244   the manuscript.

245   **Statistical analysis**

246   Data are reported as mean ± SD throughout, unless otherwise stated. Statistical analysis was

247   performed using IBM SPSS for Windows (Chicago, Illinois, Version 21) and significance was

248   set at $p < .05$. All variables were checked for outliers and normality was assessed using the

249   Shapiro-Wilk test. Changes in anthropometrics, body composition and RMR from baseline to

250   post-intervention were assessed using paired sample t-tests. To examine changes in EI, free-

251   living PA, SB, NEPA and activity EE in response to structured aerobic exercise, one-way

252   repeated measures ANCOVA were performed with baseline BMI entered as a covariate and

253   reported where significant. Change in subjective appetite sensations and appetite hormones

254   from baseline to post-intervention were assessed using two-way ANCOVA (Week*Time) with

255   effects of baseline BMI reported where significant. Where appropriate Greenhouse-Geisser

256   probability levels were used to adjust for sphericity. Post hoc comparisons using Bonferroni

257   adjustments were used if statistical significance was detected. Because of the large individual

258   variations in fasting levels of metabolic and appetite hormones, the change from baseline was

259   computed at each time point for each individual for all of the variables. Simple linear regression

260   was also performed to identify whether differences in exercise-induced EE or change in total

261   EI explained the variation in body composition change between participants. The last

262   observation carried forward (LOCF) method was used to account for missing data for the eight

263   participants who dropped out of the study. The analyses that were conducted on the completer

264   dataset were repeated on the LOCF dataset. Results were reported only when LOCF analyses

265   differed from completer analyses.

266 **RESULTS**

267 The prescribed total EE over the 12-week exercise intervention was 29,000 kcal for each

268 participant. The mean total measured exercise-induced EE was 28,792.3 kcal (SD = 872.96),

269 which was 99.3% of the prescribed EE.

270 **Change in body composition, anthropometrics and resting metabolism**

271 Paired sample t-tests revealed there was a significant reduction in BM [t(23) = 2.18, p = .04],

272 BMI [t(23) = 2.25, p = .035], WC [t(23) = 4.60, p < .001] and FM [t(23) = 3.36, p = .003] and

273 a significant increase in FFM [t(23) = 3.35, p = .003], see **Table 1**.

274 Assuming 1 kg of BM (70:30 fat/lean tissue) is equivalent to 7,700 kcal (24), the predicted

275 sample average weight loss resulting from the total exercise-induced energy deficit (28,792.29

276 kcal) was 3.74 kg. The observed weight loss was less than the predicted weight loss (22.19%

277 of predicted) indicating compensation for the exercise-induced energy deficit occurred. There

278 was no significant change in RMR from baseline to week 12 [p = .304], see **Table 1**.

279   **Table 1 around here**

280 There was considerable variability in weight loss and body composition change between

281 participants. Seventeen participants lost weight, one participant remained the same and six

282 participants gained weight following the 12-week supervised aerobic exercise intervention.

283 Changes in BM ranged from -4.3 kg to +3.1 kg (see figure 1). Of the 24 participants, 20 reduced

284 their FM, one remained the same and three gained FM with changes ranging from -4.4 kg to

285 +4.9 kg. Two participants had unfavourable changes in both FM (increased) and FFM

286   (decreased). Total exercise-induced EE did not explain the variation in BM change $[F_{(1, 22)} =$

287   1.259, p = .274, R^2 = .054], FM change $[F_{(1, 22)} = 2.418, p = .134, R^2 = .099]$ or FFM change

288    $[F_{(1, 22)} = 1.475, p = .237, R^2 = .063]$.

289   **Energy intake**

290   Paired sample t-tests revealed participants total EI during week 12 probe days was significantly

291   higher compared with total EI during baseline probe days $[t_{(23)} = 2.35, p = 0.028]$.

292   Furthermore, *ad libitum* EI (lunch, dinner and snack box EI combined) $[t_{(23)} = 2.31, p = .03]$

293   and snack box EI $[t_{(23)} = 2.09, p = .048]$ were also higher at week 12. However, there was no

294   significant difference in lunch [p = .998] or dinner [p = .194] EI, see **Table 2.** When these

295   analyses were adjusted for baseline BMI (ANCOVA), there was no effect of BMI and no

296   interaction between BMI and the intervention.

297   **Table 2 around here**

298   As with body composition change, there was considerable variability in total EI change from

299   baseline to week 12 between participants. Ten participants decreased their EI, whereas 14

300   participants increased their EI. Change in total EI ranged from -581.5 kcal/d to +763.9 kcal/d.

301   Change in total EI did not explain the variation in BM change $[F_{(1, 22)} = 0.583, p = .453, R^2$

302   = .026], FM change $[F_{(1, 22)} = 1.336, p = .260, R^2 = .057]$ or FFM change $[F_{(1, 22)} = 1.065, p$

303   = .313, R^2 = .046].

304   **Subjective appetite sensations**

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305   There was no significant difference between baseline and week 12 fasting hunger ratings [$t(23)$

306    $= 1.64$, $p = .12$]. There was a main effect of week [$F(1, 23) = 7.82$, $p = .01$] with hunger being

307   higher (when measured over the whole day) at week 12 (M = 25.58 mm, SD = 16.49) compared

308   with baseline (M = 21.68 mm, SD = 17.11). Pairwise comparisons with Bonferroni adjustments

309   revealed VAS hunger ratings were significantly higher during the post-intervention probe days

310   compared with baseline immediately post-breakfast [$t(23) = 2.08$, $p = .049$], 15 min [$t(23) =$

311   2.65, $p = .014$], 30 min [$t(23) = 2.63$, $p = .015$], 90 min [$t(23) = 2.20$, $p = .038$], immediately

312   post-lunch [$t(23) = 2.33$, $p = .029$], immediately post-dinner [$t(23) = 2.63$, $p = .015$] and 600

313   min [$t(23) = 3.01$, $p = .006$]. There was also a main effect of time [$F(2.69, 61.95) = 66.99$, $p <$

314    $.001$) but no week*time interaction [$F(6.12, 140.70) = 0.73$, $p = .63$], see **Figure 2a**. Paired

315   sample t-tests revealed there was a significant increase in AUC for hunger [$t(23) = 2.61$, $p =$

316    $.016$] throughout the whole day from baseline to week 12.

317   There was no significant difference between baseline and week 12 fasting fullness ratings [$t(23)$

318    $= 1.03$, $p = .32$]. There was a main effect of week [$F(1, 23) = 5.55$, $p = .03$], with fullness being

319   lower (when measured over the whole day) at week 12 [M = 56.12 mm, SD = 19.54] compared

320   with baseline [M = 60.06 mm, SD = 19.71]. Pairwise comparisons with Bonferroni adjustments

321   revealed VAS fullness ratings were significantly lower during the week 12 probe days

322   compared with baseline at 30 min [$t(23) = 2.17$, $p = .040$], 180 min [$t(23) = 2.65$, $p = .014$],

323   immediately post-lunch [$t(23) = 2. 78$, $p = .011$], immediately post-dinner [$t(23) = 2.49$, $p =$

324    $.021$] and at 600 min [$t(23) = 2.41$, $p = .024$]. There was also a main effect of time [$F(4.26,$

325    $97.99) = 75.28$, $p < .001$) but no week*time interaction [$F(7.54, 173.32) = 0.58$, $p = .78$], see

326   **Figure 2b**. Paired sample t-tests revealed there was a significant decrease in AUC for fullness

327   [$t(23) = 2.18$, $p = .04$] throughout the whole day from baseline to week 12. The results of these

328   analyses did not change when controlling for baseline BMI (ANCOVA).

Change in free-living physical activity, sedentary behaviour and non-exercise physical activity

When the structured exercise sessions were included in the SWA data during the week 1 and 10 measurement period, the amount of time spent in MVPA was significantly different between the four different time points [$F(3, 66) = 18.57$, $p < .001$]. Post hoc tests revealed MVPA was significantly higher during the first and tenth week of the exercise intervention compared to baseline and post-intervention [$p < .05$], see **Figure 3a**. Similarly, activity EE differed significantly between the different time points [$F(3, 66) = 17.16$, $p < .001$]. Post hoc tests revealed activity EE was also significantly higher during the first and tenth week of the exercise intervention compared with baseline and post-intervention [$p < .05$], see **Figure 3a**.

A repeated measures ANCOVA revealed that there was a significant difference in mean sedentary time between the different time points [$F(3, 66) = 3.32$, $p = .03$]. Post hoc tests revealed that there was a significant increase in sedentary time between the first week of exercise and the week following the completion of the exercise intervention [$p = .02$]. When the repeated measures ANCOVA was conducted on the LOCF dataset [$F(3, 93) = 5.11$, $p = .002$], there was a significant decrease in SB from baseline to week 1 [$p = .043$] and baseline to week 10 [$p = .047$] of the exercise intervention. The increase in sedentary time between the first week of exercise and the week following the completion of the exercise intervention remained significant [$p = .02$]. There was no covariate effect of baseline BMI and no interaction between BMI and the intervention.

Sleep, sedentary time, light PA and MVPA are collinear which means an increase in one category of activity would lead to a decrease in at least one other. The sum of the change in sleep, sedentary time and light PA (all categories excluding MVPA) between baseline and

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352   week 1 and baseline and week 10 was calculated to identify whether the increase in structured

353   MVPA displaced these activities rather than displacing MVPA that participants already

354   performed as part of their daily routines. The sum of all the activity categories other than

355   MVPA between baseline and week 1 was -59.61 min/d (SD = 43.89) and between baseline and

356   week 10 was -41.19 min/d (SD = 51.70). Change in MVPA from baseline to week 1 was +50.20

357   min/d (SD = 37.96) and from baseline to week 10 was +42.63 min/d (SD = 49.87). Structured

358   MVPA appears to displace sleep, SB and light PA but not NEPA MVPA.

359   When the structured exercise was removed from the SWA data during week 1 and week 10 of

360   the exercise intervention there was no significant difference between baseline, week 1, week

361   10 and post-intervention NEPA MVPA [$F(3, 66) = 0.05$, $p = .99$] or NEPA activity EE [$F(3,$

362    $66) = 0.87$, $p = .46$], see **Figure 3b**. NEPA MVPA ranged from 85.8 min/d to 88.7 min/d and

363   NEPA activity EE ranged from 864.4 kcal/d to 760.1 kcal/d.

364   **Change in fasting and postprandial appetite-related peptide response**

365   There was a significant decrease in fasting insulin levels from baseline to post-intervention, as

366   shown in **Table 3**. There was no significant difference in fasting acylated ghrelin, PYY or GLP-

367   1 between baseline and post-intervention [$p > .05$].

368   **Table 3 around here**

369   Postprandial profiles for insulin, acylated ghrelin, PYY, and GLP-1 at baseline and post-

370   intervention are displayed in **Figure 4**. There was a main effect of week for PYY [$F(1, 17) =$

371   9.14, $p = .008$] which was higher post-intervention (M = 51.19 ng/L, SD = 21.93) compared

372   with baseline (M = 35.96 ng/L, SD = 16.36). Post hoc tests using the Bonferroni correction

373 revealed that PYY was significantly higher during the post-intervention probe day at +30 min

374 [p = .002], +60 min [p = .003], and +90 min [p = .041]. There was a main effect of time [$F_{(2.01,}$

375 $_{34.23)}$ = 17.24, p < .001] and a significant week*time interaction [$F_{(3.00, 51.06)}$ = 3.17, p =

376 .032].

377 There was no main effect of week for insulin [$F_{(1, 17)}$ = 1.29, p = .272], acylated ghrelin [$F_{(1,}$

378 $_{16)}$ = 0.21, p = .651] or GLP-1 [$F_{(1, 17)}$ = 0.23, p = .642]. There was a significant main effect

379 of time for insulin [$F_{(1.31, 22.24)}$ = 67.35, p < .001], acylated ghrelin [$F_{(1.98, 31.65)}$ = 64.34,

380 p < .001] and GLP-1 [$F_{(2.01, 34.19)}$ = 34.50, p < .001], however there was no week*time

381 interaction for insulin [$F_{(2.81, 47.68)}$ = 0.96, p = .417], acylated ghrelin [$F_{(3.23, 51.72)}$ = 1.16,

382 p = .335] or GLP-1 [$F_{(2.80, 47.67)}$ = 1.36, p = .268].

383 **DISCUSSION**

384 The 12-week exercise intervention resulted in a significant reduction in BM and FM, refuting

385 claims from some academics that exercise/PA does not promote weight loss (25). However,

386 weight loss was less than predicted and there was considerable variability in weight change

387 between individuals ranging from -4.3 kg to +3.1 kg. Less than predicted weight loss and large

388 individual variability in weight change have previously been reported in response to increased

389 exercise (1, 7). Total exercise-induced EE throughout the intervention (99.3% of prescribed on

390 average) did not contribute to the variability in weight change, thus ruling out the possibility

391 that the variability was due to adherence to the exercise intervention.

392 It has been suggested that exercise-induced EE will be compensated for through increased EI

393 or decreased NEPA to offset the negative energy balance, rendering exercise futile for weight

394 loss (26, 27). The exercise-induced energy deficit in the current study was not fully

395   compensated for as participants did in fact lose weight on average. However, partial

396   compensation was evident as participants lost less weight than predicted when calculated based

397   on the exercise-induced energy deficit. When calculated the increase in EI between baseline

398   and post-intervention probe days was repeated every day for 12-weeks the accumulated

399   increase in EI would be approximately 15,000 kcal. This is approximately half of the EE due

400   to exercise; thereby effectively reducing the exercise potency by 50%. It is also worth noting

401   that the static Wishnofsky predictive equation (24) for estimating weight loss is simplistic and

402   does not account for adaptations in other components of energy balance as a result of an energy

403   deficit (for example, increased EI, physiological reductions in RMR, an increase in FFM or a

404   decrease in NEPA) and could lead to overestimation of predicted weight loss (28).

405   Furthermore, the 1 kg of BM is equivalent to 7700 kcal rule (1 kg of BM consists of 70% fat

406   and 30% FFM) is based on short-term low-calorie diets and is not directly applicable to the

407   change in body composition induced by exercise. Indeed, in the current study, and others (29),

408   there was in fact a significant increase in FFM.

409   It was hypothesised that EI would increase post-intervention in response to increased exercise

410   as has previously been demonstrated (7, 10). Indeed, there was a significant increase in total,

411   *ad libitum* and snack box EI at week 12. While some studies show no change in EI, these are

412   often unsupervised and rely on self-report measures of EI (30). When calculated as a

413   proportion of the energy expended per exercise session, the increase in EI represented

414   compensation of 36%, which is similar to the 30% compensation observed by Whybrow et al.

415   (10). The participants in the Whybrow study were lean men and women and would be

416   expected to compensate for a negative energy balance more readily as they have less of a

417   'buffer' (FM) than overweight or obese individuals. That could explain why the degree of

418   compensation is similar in both studies despite the present study being considerably longer.

419 Participants had more FM in the current study and therefore compensation may not occur as

420 quickly as would be expected in lean individuals. It has previously been noted that BM

421 regulation is asymmetrical; a positive energy balance (and weight gain) is well tolerated

422 whereas a negative energy balance (and weight loss) is strongly defended against (31). This

423 study, together with previous research (32), provides further support for the asymmetry of

424 BM regulation evidenced by the compensatory increase in EI to defend against weight loss in

425 response to a prolonged period of increased exercise-induced EE. A strength of this study is

426 the objective measurement of 24 hour EI, however, it is acknowledged that using episodic

427 test meal intake to infer changes in habitual intake has limitations (33). Rather, probe day

428 measures of EI can be viewed as assays for eating behaviour and give an indication of

429 compensatory appetite responses to perturbations in energy balance that are free from

430 external influences (34). Similar test meals and probe day procedures to those reported in the

431 current study have previously been shown to detect exercise-induced compensation in eating

432 behaviour (7).

433 The increase in EI was accompanied by an increase in hunger throughout the day (mainly

434 during the morning) and decreased fullness reflected in AUC for hunger and fullness. The

435 results of the current study are similar to those observed in 'non-responders' in the study by

436 King et al. (6) with respect to change in BM (-0.9 kg), FM (-1.2 kg), EI (+164 kcal) and AUC

437 for hunger and fullness. A possible explanation is that the majority of the participants in the

438 current study are 'non-responders'; they do not achieve the predicted change in body

439 composition calculated from their exercise-induced EE. When the current sample are

440 categorised as 'responders' and 'non-responders' using the method described by King et al.

441 (6), two thirds are classified as 'non-responders'. Participants in the current study had a lower

442 BMI at the start of the study (27.94 kg/m^2 vs. 31.80 kg/m^2) which could explain why their

443   weight loss response was less pronounced than that observed in a previous study (6).

444   Furthermore, the study by King et al. (6) included men and men have been shown to exhibit a

445   greater weight loss in response to exercise than women (35, 36). However, this is not a

446   universal finding (37). The current findings in women should not be assumed to generalise to

447   men and further research is required to verify this.

448   Greater compensation in NEPA, rather than changes in EI, have previously been reported in

449   response to increased exercise (38). In the current study, SWA data was initially analysed

450   with structured exercise included in the data collected during week 1 and 10 of the exercise

451   intervention. When MVPA and activity EE were compared across the four time points

452   (baseline, week 1, week 10 and post-intervention) participants spent significantly more time

453   in MVPA and had significantly higher activity EE during week 1 and week 10 compared with

454   baseline and post-intervention. Total compensation in NEPA would be apparent if, for

455   example, MVPA and activity EE did not increase during the exercise intervention. MVPA

456   and activity EE returned to baseline values when PA was measured post-intervention. This

457   demonstrates that participants did not maintain their increased PA levels once the

458   intervention ended. Post-interventions PA levels similar to baseline have previously been

459   highlighted (39-42).

460   There was no evidence for a compensatory increase in SB. In fact, SB was lower in the weeks

461   during the exercise intervention, but only the difference between week 1 of the exercise

462   intervention and post-intervention reached statistical significance. This suggests that the

463   structured exercise displaced some sedentary time. This is in contrast with previous research

464   that suggests that interventions need to specifically target reductions in SB to change

465   sedentary time (12). Indeed, the magnitude of the reduction in SB may have been greater with

466   a specific component of the intervention to target reduced SB in the current study. Further

467   examination of activity monitor data suggests structured exercise also displaces some sleep

468   time and light PA, but the difference in sleep and light PA at the different time points

469   throughout the intervention were not significant. The sum of the difference in sleep, SB and

470   light PA between baseline and week 1 and baseline and week 10 was greater than the change

471   in MVPA (in the opposite direction) at the same time points. Furthermore, when the

472   prescribed exercise was removed from SWA data during week 1 and 10, the remaining

473   NEPA MVPA was remarkably similar to baseline and post-intervention values (<3 minutes

474   difference between all four time points) and there was no significant difference in NEPA

475   activity EE across the four time points. Taken together, these findings suggest that increasing

476   MVPA through a structure exercise intervention displaces time spent sleeping, sedentary and

477   in light PA but not NEPA MVPA. This is in agreement with previous studies (40, 42) and a

478   recent systematic review that concluded no statistically or clinically significant mean change

479   in NEPA occurs during exercise training (11).

480   Appetite-related peptides were measured in this study in order to determine if any exercise-

481   induced changes could be related to adjustments in fasting or postprandial gastrointestinal

482   signalling. However, the peptides did not account for changes in subjective appetite

483   sensations or in EI. PYY was higher on average during post-intervention probe days,

484   however this was not coupled with a decrease in hunger or an increase in fullness as might be

485   expected. In fact, there was a significant increase in hunger and decrease in fullness post-

486   intervention. There was no change in postprandial profiles for insulin, acylated ghrelin or

487   GLP-1 in the present study. Acute studies suggest an exercise intensity of at least 65% $\dot{V}O_2$ is

488   required to induce changes in appetite related peptides (43, 44). However, the present

489   findings are not comparable due to the assessment of longer-term exercise training. There

490   was a significant decrease in fasting insulin from baseline to post-intervention. As insulin

491   levels are proportional to FM it is likely the reduction in insulin was driven by the reduction

492   in FM following the exercise intervention. However, some studies have demonstrated

493   improved insulin sensitivity following exercise interventions independent of weight loss/body

494   composition changes whilst others have demonstrated improvements only occur with weight

495   loss (45). The relative importance of exercise and weight loss remains unclear and it is

496   possible both contributed to the reduction in fasting insulin levels in the present study. These

497   findings, while novel in this context, suggest that the changes in appetite are more likely due

498   to changes in body composition rather than changes in appetite peptides, as has previously

499   been proposed (46). It is possible that a greater change in body composition would be

500   required to see concomitant changes in appetite peptides.

501

502   The quasi-experimental design used in the present study allows certain inferences to be made

503   from the presence or lack of changes in compensatory EI and EE behaviours before and after

504   medium-term exercise training. However due to the single non-randomised sample it is not

505   possible to rule out that the effects reported here would not have been seen after 12 weeks of

506   rest (with the two conditions randomised). Future confirmation of these findings using a

507   randomised controlled trial design would be valuable.

508   On average there was a significant increase in EI from baseline to post-intervention providing

509   a plausible explanation for the less than predicted weight loss. However, change in total EI

510   did not explain the variation in BM change. Laboratory measures of EI do not reflect the

511   turbulence of the free-living environment in which eating behaviour is more haphazard and

512   cannot be captured. Indeed, it is possible that the measure of EI obtained from the probe days

513   may not reflect participants eating habits in the free-living environment.

514   It must also be acknowledged that participants' menstrual cycle was not recorded and

515   therefore could not be included as a covariate in analyses. Since there does not seem to be

23

516 any discernible differences between sexes in the appetite and eating behaviour response to

517 acute and longer-term exercise interventions (37, 47), the authors think it is unlikely that the

518 menstrual cycle had a major impact on the study outcomes.

519 Finally, it is worth emphasising that exercise alone is clearly not the most effective way to

520 lose weight, particularly when compared to standard behavioural interventions in which

521 participants may lose 5-10% of weight. The present study demonstrates that exercise can

522 produce modest fat loss without additional dietary assistance. However, the compensatory

523 increase in energy intake observed suggests that an additional dietary intervention would

524 support an even greater weight (fat) loss.

525 **CONCLUSIONS**

526 Overweight women took part in an exercise intervention which comprised five mandatory

527 sessions of aerobic exercise per week for 12-weeks. No constraint was placed on other free-

528 living behaviour (activity or eating) during the 12-weeks. Therefore, participants were able to

529 demonstrate compensation for the energy expended in exercise by an adjustment of their food

530 intake or the amount of SB or free-living PA. At the end of 12-weeks there was a significant

531 decrease in FM and an increase in FFM indicating that the exercise regime had been effective

532 and had generated a significant impact on body composition. However, there was considerable

533 individual variability and the changes in body composition were smaller than could have been

534 expected on the basis of the total energy expended through exercise (actual weight loss was

535 22.19% of predicted). Compensation for the exercise induced EE was detected in a significant

536 increase in EI but no increase in SB or decrease in free-living PA. In fact, the exercise actually

537 displaced SB. The effect of exercise on FM could be amplified by the addition of a dietary

538 strategy designed to prevent a compensatory increase in EI.

24

539 Despite finding a short-term increase in EI during laboratory probe days, the magnitude of this

540 effect was not sufficient to fully explain the difference between predicted and observed weight

541 loss. While food intake in the laboratory setting provides a plausible objective marker of

542 changes in free-living intake, it may not reflect absolute levels of energy consumed during the

543 intervention. Therefore it is not possible to decisively conclude from the present findings that

544 compensation for the exercise was due to EI alone. Future studies using other comprehensive

545 measures of EI and EE are needed to corroborate the present results. Moreover, future studies

546 should investigate how weight status (lean, overweight, obese), the amount of exercise applied

547 (volume, intensity) and the periodicity of exercise (frequent small bouts or fewer large bouts)

548 effect the relationship between exercise and behavioural consequences. Considering an effect

549 on EI, it is known that this end point is influenced by body composition (FM and FFM). These

550 variables are also influenced by exercise, therefore any effect of exercise may be mediated

551 indirectly via changes in body composition or directly through some mechanism involved in

552 cellular metabolism.

553 **LIST OF ABBREVIATIONS**

554 ANCOVA, analysis of covariance; BM, body mass; BMI, body mass index; EE, energy

555 expenditure; EI, energy intake; FFM, fat-free mass; FM, fat mass; HR, heart rate; LOCF, last

556 observation carried forward; MVPA, moderate-to-vigorous physical activity; NEPA, non-

557 exercise physical activity; PA, physical activity; RMR, resting metabolic rate; SB, sedentary

558 behaviour; SD, standard deviation; SWA, SenseWear Armband mini; VAS, visual analogue

559 scale; WC, waist circumference.

560 **DECLARATIONS**

561 Ethics approval and consent to participate

562 All participants provided written informed consent before taking part in the study. The study

563 procedures and all study materials were reviewed and approved by the National Research

564 Ethics Service Committee Yorkshire & the Humber (ref: 09/H1307/7).

565 Consent for publication

566 Not applicable

567 Availability of data and material

568 The datasets used and/or analysed during the current study are available from the corresponding

569 author on reasonable request.

570 Competing interests

571 The authors declare that they have no competing interests.

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575 Authors' contributions

576 AM, MD, CG, GF and JB designed research; AM, MD and CG conducted research; AM

577 analysed data; AM, CG, GF and JB discussed data analysis and interpretation of the data; AM

578 and JB wrote manuscript. All authors approved the final manuscript.

581

582 **REFERENCES**

583 1.   Thomas D, Bouchard C, Church T et al. Why do individuals not lose more
584   weight from an exercise intervention at a defined dose? An energy balance
585   analysis. *Obes. Rev.* 2012;13(10):835-47.

586 2.   Gray R. Health warning - Exercise makes you fat. In. *Daily Telegraph* 2009.

587 3.   Myers A, Gibbons C, Finlayson G, Blundell J. Associations among sedentary
588   and active behaviours, body fat and appetite dysregulation: investigating the
589   myth of physical inactivity and obesity. *Br. J. Sports Med.* 2016:bjsports-2015-
590   095640.

591 4.   Shook RP, Hand GA, Drenowatz C et al. Low levels of physical activity are
592   associated with dysregulation of energy intake and fat mass gain over 1 year.
593   *Am J Clin Nutr.* 2015;102(6):1332-8.

594 5.   Shaw K, Gennat H, O'Rourke P, Del Mar C. Exercise for overweight or obesity.
595   *Cochrane Database Syst Rev.* 2006;4(4).

596 6.   King NA, Caudwell PP, Hopkins M, Stubbs JR, Naslund E, Blundell JE. Dual-
597   process action of exercise on appetite control: increase in orexigenic drive but
598   improvement in meal-induced satiety. *Am J Clin Nutr.* 2009;90(4):921-7.

599 7.   King N, Hopkins M, Caudwell P, Stubbs R, Blundell J. Individual variability
600   following 12 weeks of supervised exercise: Identification and characterization
601   of compensation for exercise-induced weight loss. *Int. J. Obes.* 2008;32(1):177-
602   84.

603 8.   Swift DL, Johannsen NM, Lavie CJ, Earnest CP, Church TS. The role of
604   exercise and physical activity in weight loss and maintenance. *Prog.*
605   *Cardiovasc. Dis.* 2014;56(4):441-7.

606 9. Hall KD, Sacks G, Chandramohan D et al. Quantification of the effect of energy
607   imbalance on bodyweight. *The Lancet.* 2011;378(9793):826-37.

608 10. Whybrow S, Hughes DA, Ritz P et al. The effect of an incremental increase in
609   exercise on appetite, eating behaviour and energy balance in lean men and
610   women feeding ad libitum. *Br. J. Nutr.* 2008;100(05):1109-15.

611 11. Fedewa MV, Hathaway ED, Williams TD, Schmidt MD. Effect of Exercise
612   Training on Non-Exercise Physical Activity: A Systematic Review and Meta-
613   Analysis of Randomized Controlled Trials. *Sports Med.* 2016;47(6):1171-82.

614 12. Kozey-Keadle S, Staudenmayer J, Libertine A et al. Changes in sedentary time
615   and physical activity in response to an exercise training and/or lifestyle
616   intervention. *J Phys Act Health.* 2014;11(7):1324-33.

617 13. Washburn R, Lambourne K, Szabo A, Herrmann S, Honas J, Donnelly J. Does
618   increased prescribed exercise alter non-exercise physical activity/energy
619   expenditure in healthy adults? A systematic review. *Clinical Obesity.*
620   2014;4(1):1-20.

621 14. Donnelly JE, Herrmann SD, Lambourne K, Szabo AN, Honas JJ, Washburn
622   RA. Does Increased Exercise or Physical Activity Alter Ad-Libitum Daily Energy
623   Intake or Macronutrient Composition in Healthy Adults? A Systematic Review.
624   *PLoS One.* 2014;9(1):e83498.

625 15. Gibbons C, Caudwell P, Finlayson G, King N, Blundell J. Validation of a new
626   hand-held electronic data capture method for continuous monitoring of
627   subjective appetite sensations. *Int. J. Behav. Nutr. Phys. Act.* 2011;8(1):57-64.

628 16. Ainsworth BE, Haskell WL, Herrmann SD et al. 2011 Compendium of Physical
629   Activities: a second update of codes and MET values. *Med. Sci. Sports Exerc.*
630   2011;43(8):1575-81.

631 17. Scheers T, Philippaerts R, Lefevre J. Variability in physical activity patterns as
632   measured by the SenseWear Armband: how many days are needed? *Eur. J.*
633   *Appl. Physiol.* 2012;112(5):1653-62.

634 18. Berntsen S, Hageberg R, Aandstad A et al. Validity of physical activity monitors
635   in adults participating in free-living activities. *Br. J. Sports Med.* 2010;44(9):657-
636   64.

637 19. Calabró MA, Lee J-M, Saint-Maurice PF, Yoo H, Welk GJ. Validity of physical
638   activity monitors for assessing lower intensity activity in adults. *Int. J. Behav.*
639   *Nutr. Phys. Act.* 2014;11(1):119-28.

640 20. Malavolti M, Pietrobelli A, Dugoni M et al. A new device for measuring resting
641   energy expenditure (REE) in healthy subjects. *Nutr. Metab. Cardiovasc. Dis.*
642   2007;17(5):338-43.

643 21. Johannsen DL, Calabro MA, Stewart J, Franke W, Rood JC, Welk GJ. Accuracy
644   of armband monitors for measuring daily energy expenditure in healthy adults.
645   *Med. Sci. Sports Exerc.* 2010;42(11):2134-40.

646 22. Péronnet F, Massicotte D. Table of nonprotein respiratory quotient: an update.
647   *Can. J. Sport Sci.* 1991;16(1):23-9.

648 23. Degen L, Oesch S, Casanova M et al. Effect of peptide YY3–36 on food intake
649   in humans. *Gastroenterology.* 2005;129(5):1430-6.

650 24. Wishnofsky M. Caloric equivalents of gained or lost weight. *Am J Clin Nutr.*
651   1958;6(5):542-6.

652 25. Malhotra A, Noakes T, Phinney S. It is time to bust the myth of physical inactivity
653   and obesity: you cannot outrun a bad diet. *Br. J. Sports Med.* 2015:bjsports-
654   2015-094911.

655 26. King NA, Caudwell P, Hopkins M et al. Metabolic and behavioral compensatory
656   responses to exercise interventions: barriers to weight loss. *Obesity.*
657   2007;15(6):1373-83.

658 27. Melanson EL. The effect of exercise on non-exercise physical activity and
659   sedentary behavior in adults. *Obes. Rev.* 2017;18:40-9.

660 28. Thomas DM, Gonzalez MC, Pereira AZ, Redman LM, Heymsfield SB. Time to
661   Correctly Predict the Amount of Weight Loss with Dieting. *J. Acad. Nutr. Diet.*
662   2014;114(6):857-61.

663 29. Stiegler P, Cunliffe A. The role of diet and exercise for the maintenance of fat-
664   free mass and resting metabolic rate during weight loss. *Sports Med.*
665   2006;36(3):239-62.

666 30. Caudwell P, Gibbons C, Finlayson G, Näslund E, Blundell J. Exercise and
667   weight loss: No sex differences in body weight response to exercise. *Exerc.*
668   *Sport Sci. Rev.* 2014;42(3):92-101.

669 31. Blundell JE, Gillett A. Control of food intake in the obese. *Obes. Res.* 2001;9
670   Suppl 4:263S-70S.

671 32. Stubbs RJ, Hughes DA, Johnstone AM, Horgan GW, King N, Blundell JE. A
672   decrease in physical activity affects appetite, energy, and nutrient balance in
673   lean men feeding ad libitum. *Am J Clin Nutr.* 2004;79(1):62-9.

674 33. Hill A, Rogers P, Blundell J. Techniques for the experimental measurement of
675   human eating behaviour and food intake: a practical guide. *International journal*
676   *of obesity and related metabolic disorders: journal of the International*
677   *Association for the Study of Obesity.* 1995;19(6):361-75.

678 34. Gibbons C, Finlayson G, Dalton M, Caudwell P, Blundell JE. Metabolic
679   phenotyping guidelines: studying eating behaviour in humans. *J. Endocrinol.*
680   2014;222(2):G1-G12.

681 35. Ballor D, Keesey RE. A meta-analysis of the factors affecting exercise-induced
682   changes in body mass, fat mass and fat-free mass in males and females. *Int.*
683   *J. Obes.* 1991;15(11):717-26.

684 36. Donnelly JE, Smith BK. Is exercise effective for weight loss with ad libitum diet?
685   Energy balance, compensation, and gender differences. *Exerc. Sport Sci. Rev.*
686   2005;33(4):169-74.

687 37. Caudwell P, Gibbons C, Hopkins M, King NA, Finlayson G, Blundell JE. No sex
688   difference in body fat in response to supervised and measured exercise. *Med.*
689   *Sci. Sports Exerc.* 2013;45(2):351-8.

690 38. Stubbs RJ, Sepp A, Hughes DA, Johnstone AM. The effect of graded levels of
691   exercise on energy intake and balance in free-living men, consuming their
692   normal diet. *Eur. J. Clin. Nutr.* 2002;56(2):129-40.

693 39. Rocha J, Paxman J, Dalton C, Winter E, Broom DR. Effects of a 12-week
694   aerobic exercise intervention on eating behaviour, food cravings, and 7-day
695   energy intake and energy expenditure in inactive men. *Appl Physiol Nutr Metab.*
696   2016;41(11):1129-36.

697 40. Turner JE, Markovitch D, Betts JA, Thompson D. Nonprescribed physical
698   activity energy expenditure is maintained with structured exercise and
699   implicates a compensatory increase in energy intake. *Am J Clin Nutr.*
700   2010;92(5):1009-16.

701 41. Hollowell RP, Willis LH, Slentz CA, Topping JD, Bhakpar M, Kraus WE. Effects
702   of exercise training amount on physical activity energy expenditure. *Med. Sci.*
703   *Sports Exerc.* 2009;41(8):1640-4.

704 42. Church TS, Martin CK, Thompson AM, Earnest CP, Mikus CR, Blair SN.
705   Changes in weight, waist circumference and compensatory responses with
706   different doses of exercise among sedentary, overweight postmenopausal
707   women. *PLoS One.* 2009;4(2):e4515.

708 43. Broom DR, Stensel DJ, Bishop NC, Burns SF, Miyashita M. Exercise-induced
709   suppression of acylated ghrelin in humans. *J. Appl. Physiol.* 2007;102(6):2165-
710   71.

711 44. Deighton K, Batterham RL, Stensel DJ. Appetite and gut peptide responses to
712   exercise and calorie restriction. The effect of modest energy deficits. *Appetite.*
713   2014;81:52-9.

714 45. Bird SR, Hawley JA. Update on the effects of physical activity on insulin
715   sensitivity in humans. *BMJ open sport & exercise medicine.*
716   2017;2(1):e000143.

717 46. Gibbons C, Finlayson G, Caudwell P, Näslund E, Blundell J. 12-weeks
718   Supervised Aerobic Exercise Improves Appetite Regulation, Reduces Fat Mass
719   And Adjusts Gastrointestinal Peptide Biomarkers. *Med. Sci. Sports Exerc.*
720   2015;47(5S):680.

721 47. Thackray AE, Deighton K, King JA, Stensel DJ. Exercise, appetite and weight
722   control: are there differences between men and women? *Nutrients.*
723   2016;8(9):583.

724

725 **FIGURES, TABLES AND ADDITIONAL FILES**

726 Figure 1. Individual variability in BM change between participants.

727   Figure 2. VAS (a) hunger and (b) fullness ratings during baseline (BL) and post-intervention

728   (PI) probe days (error bars are standard error). $*$ = p < .05, indicates significant difference

729   between baseline and post-intervention.

730   Figure 3. Time spent in MVPA and activity EE before (baseline; BL), during the 12-week

731   exercise intervention (week 1 and 10) and after the exercise intervention (post-intervention; PI)

732   measured using the SWA with structured exercise included (a) and removed (b) from the data

733   (n=23), $**$ = p < .01, $***$ = p < .001.

734   Figure 4. Postprandial profiles for insulin (a), acylated ghrelin (b), PYY (c), and GLP-1 (d) at

735   baseline (BL) and post-intervention (PI; n=18), $*$ = p < .05, $**$ = p < .01.