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Effect of exercise on energy expenditure and body composition in astronauts onboard the International Space Station: Considerations for interplanetary travel

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1 **Original research article**

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3 **Effect of exercise on energy expenditure and body composition in astronauts onboard the International**
4 **Space Station: Considerations for interplanetary travel**

5
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34

35 **Running heading** (84 characters)

36 Body composition and energy requirements in astronauts onboard the International Space Station

37

38 **Abstract** (277 words)

39 *Objective:* Body mass (BM) loss and body composition (BC) changes threaten astronauts' health and mission
40 success. However, the energetics contribution of the exercise countermeasure to these changes has never been
41 investigated during long-term missions. We studied energy balance and BC in astronauts during 6-month missions
42 onboard the International Space Station.

43

44 *Methods:* Before and after at least 3 months in space, BM, BC, total and activity energy expenditure (TEE and
45 AEE) were measured using the doubly labeled water method in 11 astronauts (2011- 2017). Physical activity (PA)
46 was assessed by the SensewearPro® activity-device.

47

48 *Results:* Three-month spaceflight decreased BM (-1.20kg (SE 0.5); $P=0.04$), mainly due to non-significant fat-free
49 mass loss (FFM; -0.94kg (0.59)). The decrease in walking time (-63.2min/day (11.5); $P<0.001$) from preflight was
50 compensated by increases in non-ambulatory activities (+64.8min/day (18.8); $P<0.01$). Average TEE was
51 unaffected but a large interindividual variability was noted. Astronauts were stratified into those who maintained
52 (stable_TEE; $n=6$) and those who decreased (decreased_TEE $n=5$) TEE and AEE compared to preflight data.
53 Although both groups lost similar BM, FFM was maintained and FM reduced in stable_TEE astronauts, while
54 FFM decreased and FM increased in decreased_TEE astronauts (estimated between-group-difference (EGD) in
55 $\Delta\text{FFMindex}$ [FFMI] 0.87kg/m^2 , 95% CI +0.32 to +1.41; $P=0.01$, $\Delta\text{FMindex}$ [FMI] -1.09kg/m^2 , 95% CI -2.06 to -
56 0.11kg/m^2 ; $P=0.03$). The stable_TEE group had higher baseline FFMI, and greater baseline and inflight vigorous
57 PA than the decreased_TEE group ($P<0.05$ for all). ΔFMI and ΔFFMI were respectively negatively and positively
58 associated with both ΔTEE and ΔAEE .

59

60 *Conclusion:* Both ground fitness and inflight overall PA are associated with spaceflight-induced TEE and BC
61 changes and thus energy requirements. New instruments are needed to measure real-time individual changes in
62 inflight energy balance components.

63

64 **Word counts:** 5486

65 **Key points**
66

- Space agencies ranked energy balance control to critical level for human space exploration. Although exercise is a cornerstone of countermeasure programs to prevent microgravity-induced physiological alterations, the energy cost of physical activity during long-term spaceflights has never been studied.
- Six-month missions on the ISS led to large inter-individual variability in body composition changes. Astronauts maintaining pre-flight total and activity energy expenditures (possibly due to both physical training and unexpected non-exercise activity), maintained fat-free mass but lost fat mass. Conversely those who decreased energy expenditures lost fat-free mass but gained fat mass. Fat mass changes reflects unmatched energy intake adaptation to expenditures during flight.
- On average, astronauts who maintained energy expenditures during flight were also fitter on the ground.
- The large between-astronauts variability suggests that energy requirements cannot be derived from general population equations. Methods to track inflight changes in body composition, energy intake and energy expenditure are needed to determine individual energy requirements to ensure astronaut's performance and mission success.

67
68

69 **Supplementary Information** – The online version contains supplementary material available at xxxx.

70

71 **Acknowledgments**

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73 Aerospace Company (former DAMEC, Odense Denmark) and CADMOS Center (Toulouse, France) for the
74 preparation, operational support during baseline data collection on the different training sites (Houston, TX, USA;
75 Koln, Germany) but also during the ground support of the inflight sessions. We are thanking Tanya Halliday for
76 her support in the energy intake assessment.

77

78 **Declarations**

79 *Funding* – This study was funded by the French Space Agency (CNES, French Space Agency) and the European
80 Space agency which provided financial and operational supports for the Energy experiment.

81 *Conflicts of interest* – PB, AZ, DSA, IC, ELR, CT, AM, MG, GGK, AB, CS and SB declare that they have no
82 potential conflicts of interest that might be relevant to the contents of this manuscript.

83 *Availability of data and material* – Data are accessible upon reasonable request to the authors after validations by
84 space agencies.

85 *Ethics approval* – The study was yearly approved by the NASA Institutional Review Board under NASA
86 7116301606HR. The ESA Medical Board and the JAXA Institutional Review Board for human experiment also
87 approved the experiment.

88 *Consent to participate* – The study was conducted in conformity with the policy statement regarding the use of
89 human participants as outlined in the Declaration of Helsinki of 1964. All astronauts received a detailed
90 presentation of the experiment before enrolling the study and signed a written informed consent.

91 *Consent for publication* – A global authorization from the European Space Agency was obtained to use and publish
92 the photos (ESA copyright).

93

94 **Author contribution** – SB, DAS, GGK and AM designed the study. PB, AB, CS and SB drafted the manuscript.
95 AZ, CT, AM, GGK, CS and SB collected data. PB, ELR, AZ, DAS, IC, MG, AB, CS and SB analyzed the data.
96 CS and PB realized the statistical analysis. All the authors read and approved the final version of the manuscript.

97

98 **1 INTRODUCTION**

99 The human spaceflight program has entered a new phase of space exploration directed towards the Moon and
100 Mars. Space Agencies have developed roadmaps that define research priorities enabling planetary exploration.
101 Among those, understanding inflight energy balance regulation has been placed as a top priority, as energy
102 homeostasis is vital for both medical and operational reasons. Any energy deficit will not only impact fat mass
103 (FM), but it can also increase loss of bone and of muscle mass and strength, favor cardiovascular deconditioning
104 and impair numerous other physiological functions. Overall it conditions astronaut's performance and health, and
105 ultimately may affect mission success [1].

106 A loss of body mass (BM) is a hallmark of spaceflights. On Mir (4 months)[2, 3], Shuttle (4 to 19 days)[4, 5] and
107 early International Space Station (ISS) missions (128 to 195 days)[6, 7], astronauts lost more than 5% of their
108 preflight BM despite sufficient food onboard with no clear relationship with mission duration [8]. In some cases,
109 this loss even exceeded 10% of preflight BM, which is clinically significant. Conversely, BM was successfully
110 maintained in few missions, such as SpaceLab Life Sciences Space Shuttle missions SLS1 and SLS2 in the 1990s
111 [9] or, more recently, on the ISS [7, 10]. Yet, recent reports from the ISS show that astronauts still lose from 2 to
112 5% of their initial BM with a large between-subject variability [6, 11]. We recently hypothesized that an energy
113 balance dysregulation, i.e. an uncoupling between energy intake (EI) and expenditure, occurs in space [8]. Very
114 little data, however, exist to support this hypothesis.

115 The regulation of energy balance was measured with objective doubly labeled water (DLW) methods in three
116 studies during short term spaceflights only [4, 9, 12]. The results of two of these studies indicate that energy
117 balance is not optimal in space when exercise is performed, i.e. compensatory changes in EI are insufficient to
118 match increases in energy expenditure [13]. During the 17-day Life and Microgravity Science (LMS) Space Shuttle
119 mission, Stein et al. [4] observed in four astronauts an energy deficit as large as 5.7 MJ/day that was associated
120 with the loss of up to 2 kg of fat mass (FM). Concomitantly, even though heavy exercise training was specifically
121 prescribed during this mission to prevent loss of muscle mass and bone, astronauts presented a negative nitrogen
122 balance indicating protein loss related to a maladjustment of energy and protein intakes to high requirements. By
123 contrast, during the SLS1 and SLS2 missions, during which no exercise was prescribed, the astronauts maintained
124 stable energy balance with only a moderate negative nitrogen balance likely thanks to the maintenance of protein

125 intakes despite an adapted reduction in energy intakes. [9]. Because exercise is the cornerstone of the
126 countermeasure program to prevent unloading-induced loss of skeletal muscle mass and strength, bone, aerobic
127 capacity and other health-related outcomes [14, 15], these observations suggest that understanding its role in
128 energy balance regulation is critical, notably for long-duration missions.

129 This exploratory study was therefore designed to measure total energy expenditure (TEE) and activity energy
130 expenditure (AEE), using the gold standard DLW method, in relation to body composition in 11 astronauts after
131 at least 3 months onboard the ISS. Inferences to evaluate energy requirements in space were derived. We
132 hypothesized that astronauts' individual BM and body composition changes are explained by inflight AEE.

133

134 **2 MATERIALS AND METHODS**

135 Protocols, methodological validation and related assumptions of the calculations used are fully described in **online**
136 **supplemental data**.

137

138 **2.1 Participants**

139 Eighteen astronauts (16 men and 2 women) voluntarily took part in the study between 2011 and 2017. All were
140 subjected to extensive physical and medical examination prior to the flight. None had history of chronic disease,
141 and all of them were healthy throughout the mission. Out of the eighteen astronauts, three (2 men and 1 woman)
142 performed preflight measurements only, due to rescheduling priorities during the flight, and four (3 men and 1
143 woman) served as controls to correct for background isotopic changes on the ISS during an experimental session
144 (see **supplemental methods** and **supplemental Figs 1 to 3**). Data presented here were therefore collected in 11
145 men.

146 The study was yearly approved by NASA Institutional Review Board (IRB) under NASA 7116301606HR.
147 European Space Agency (ESA) Medical Board and Japanese Space Agency (JAXA) IRB for human experiments
148 also approved the protocol. Written informed consent was obtained from all astronauts.

149

150 2.2 Protocol

151 For the 11 remaining astronauts, each completed two research sessions of 10 days, one on Earth (ground) and one
152 onboard the ISS (flight). The ground session was conducted within the year before flying, while astronauts were
153 at the European Astronaut Center (EAC) in Koln, Germany. On average, the mean delay between ground
154 measurements and the flight was 99 days (SD 78). The astronauts were deemed in energy balance the year before
155 launch (see **Fig 1A** and **supplemental methods**). The flight session was conducted after at least three months in
156 space and before the last month onboard the ISS. This three-to-five-month window was selected to provide data
157 for long-term spaceflights while avoiding stress associated with the preparation for return to Earth. On average,
158 flight sessions were conducted after 108 days (19) in space.

159 Ground and flight experiments, strictly similar, were realized under the supervision of ESA and French Space
160 Agency (CNES) science officers in charge of the experiment and the investigators from Toulouse Space Center
161 (CADMOS, France), and were preceded by dry runs of the experiments conducted at EAC. BM and body
162 composition, TEE and its components (resting metabolic rate (RMR) and AEE) and physical activity (PA) were
163 measured. Additionally, post-flight BM and composition were measured on the ground after landing.

164

165 2.3 Total energy expenditure and its components

166 TEE was measured over 10 days using the doubly-labeled water (DLW) method as previously described [16]. The
167 DLW is the gold standard to assess TEE in free living conditions. The technique is fully described in **supplemental**
168 **methods**. Briefly, it is based on the exponential elimination of the stable isotopes ^2H and ^{18}O after a bolus dose of
169 water labeled with both isotopes. The ^2H are lost as water, whereas the ^{18}O are lost both as water and as CO_2 . Thus,
170 the excess disappearance rate of ^{18}O relative to ^2H is a measure of the CO_2 production rate. This latter is converted
171 to TEE using the food quotient or the respiratory quotient and the classic indirect calorimetry equations. On the
172 DLW dosing day, RMR was measured in fasting state, at rest for 45 min using the Pulmonary Function System
173 (PFS, manufactured by the Danish Aerospace Company, former DAMEC)[17]. AEE was calculated from TEE
174 and RMR assuming a diet-induced thermogenesis of 10% of TEE. PA level (PAL) was calculated as the ratio
175 between TEE and RMR.

176

177 **2.4 Body composition**

178 Pre- and post-flight ground BM measures were obtained during NASA medical operations with a calibrated scale
179 at EAC, and fat-free mass (FFM) and FM by dual X-ray energy absorptiometry (DXA, Hologic, Marlborough,
180 MA, USA; software version 12.7.3.1 in 2011 to 15.5.3 in 2016). Inflight BM was measured using the
181 SLAMMD (Space Linear Acceleration Mass Measurement Device) within one day of the DLW dosing day, and
182 FFM and FM were obtained from the DLW method. BM, FFM and FM indexes (BMI, FFMI, FMI), were
183 calculated by dividing BM, FFM and FM (in kg) by squared height (in squared m).

184

185 **2.5 Energy intake**

186 Inflight mean daily EI, expressed in MJ/day, was calculated from the changes in body composition and TEE
187 between the preflight and inflight DLW sessions deeming astronauts were in stable energy balance on ground
188 measurements [18], and compared with theoretical EI calculated from the 2001 Dietary Reference Intakes (DRI)
189 [19]. Details are provided in **supplemental methods**.

190

191 **2.6 Physical exercise and activities**

192 Physical training sessions were composed of both aerobic and resistive exercises and prescribed 6 days/week, as
193 detailed in **supplemental methods**. Briefly, 30-45min of aerobic exercise per session were prescribed on the cycle
194 ergometer (CEVIS) or on a treadmill (T2) with vibration isolation system (**Supplemental Fig 4**) [20, 21]. During
195 treadmill exercise, an external vertical load (60-100% of BM) was applied to the astronauts to partially compensate
196 for the absence of body weight in microgravity environment. The resistance exercise program consisted of 2 to 3
197 workouts per session that were performed on the advanced resistive exercise device (ARED) (**Supplemental Fig**
198 **4**). Each workout was composed of 5 to 7 exercises primarily focused on the lower body using 8 to 15 repetitions
199 for 3 to 4 sets. Loads were individually adapted by the personal exercise trainers throughout the flight. Parameters
200 of the exercise training actually performed during the spaceflight were obtained from the exercise dairy logs.
201 Overall PA was further evaluated from a Sensewear Pro (SWP) activity monitor (Body media Inc®, Pittsburgh,
202 PA USA).

203 Self-reported exercise logs were provided by NASA. These logs included date and duration of each aerobic
204 exercise session, the average speed and external load for T2 and average power and revolution per minute for
205 CEVIS. For ARED, they included date and detailed exercise workouts of each resistive exercise session, with
206 number of sets and repetitions, and loads. Mean daily and weekly exercise parameters were calculated over a
207 period beginning 2 months before the experimental flight session (**details in supplemental files**). An estimation
208 of T2 power was derived from speed and external load from the American College of Sports Medicine (ACSM)
209 formulas [22], and aerobic exercise workloads were calculated as power*duration. Resistive exercise workloads
210 were calculated as sets*repetitions*loads [23]. Because the duration for the resistive exercise sessions was not
211 provided, muscular time under tension was estimated from the number of repetitions, assuming a 2-to-5 s count
212 for each repetition (i.e. including both concentric and eccentric phases), according to movement complexity from
213 online video featuring this kind of exercise [24]. An additional 1-min resting time by set was added to approximate
214 time involved with exercise on ARED. The goal of this calculation was to allow a comparison with the SWP-
215 derived data.

216 The SWP, an activity multi-sensor armband that includes a bi-axial accelerometer, was worn on the non-dominant
217 arm throughout the two 10-day DLW sessions. Its companion software (professional version 8.0) incorporates a
218 proprietary machine-learning activity classification algorithm based on heat flux, galvanic skin response, skin and
219 near-body ambient temperature and accelerometry measures patterns [25]. It was used to identify non-wear periods
220 and, after exclusion of non-valid days, to obtain steps and categorize each minute in 4 classes: inactivity, walking,
221 running and all other detected activities groups as non-ambulatory PA. Inflight valid data were obtained for 9
222 astronauts only. The norm of the 1-min acceleration signal mean amplitude deviation (MAD) was used as an
223 additional proxy of PA workload and intensity [26]. MAD removes the static component due to gravity from the
224 acceleration signal to keep only the dynamic component due to body movements and changes in velocity. It is
225 therefore poorly influenced by microgravity. Aggregation of 1-min MAD during activities, a composite variable
226 that corresponds to the product of duration by the intensity of a given effort/exercise bout or of overall daily PA,
227 are presented in milli-g (i.e., 0.001 g). Vigorous PA (VPA) was estimated using a MAD-cutpoint of 16 milli-g/min.
228 Details are provided in **supplemental methods**.

229

230 2.7 Statistical analyses

231 Data were first analyzed for the entire group of astronauts (n=11). To understand the inter-individual variability,
232 astronauts were then stratified according to inflight TEE, i.e. either maintained close to preflight values
233 (stable_TEE; n=6) or decreased (decreased_TEE n=5).

234 Differences between the two groups stratified for inflight TEE changes at baseline and for inflight physical training
235 were examined using unpaired Student's t-tests. Linear mixed-effects models accounting for repeated
236 measurements, with subjects as random effect and indicator for flight as fixed factor, were used to test the overall
237 effect of spaceflight on the anthropometric, energetic and PA outcomes. The group effect (stable_TEE vs
238 decreased_TEE) on the flight-induced changes was tested by adding a group x flight interaction term. Our
239 statistical inference was respectively on the overall outcome net changes and their between-group differences
240 estimated (EGD) with their 95% Confidence Interval (CI). Effect size was estimated using Hedges'g, calculated
241 as the EGD divided by the estimation of the groups' weighted pooled standard deviation, with a Hedges'g greater
242 than 0.2, 0.5, and 0.8 indicating small, medium, and large effect size, respectively [27]. TEE, AEE, EI and EB
243 were further adjusted for mean BM, and RMR for both mean FM and mean FFM (whole group mean baseline
244 values were used as reference values).

245 General linear models were used to examine the associations of 1) FFMI and FMI changes (inflight-preflight) with
246 TEE and AEE changes (inflight-preflight), and 2) inflight FFMI and FMI with different inflight PA variables.

247 Baseline values and inflight physical training variables are presented as means (standard deviation, SD). Unless
248 otherwise noted, results of the linear mixed-models are presented as least square means (standard error, SE).

249 Statistical analyses were performed using SAS 9.4 (SAS Institute, Cary, North Carolina) with a significance level
250 at 5%. Figures were realized with Prism 9 (GraphPad, San Diego, California).

251

252 3 RESULTS

253 3.1 Few spaceflight changes for the whole group of astronauts

254 At baseline (**Table 1**), astronauts had a mean age of 45.7 years (SD 7.7), a BMI of 24.3 kg/m² (2.1) with a normal-
255 to-high FFMI of 19.6 kg/m² (1.9). They were quite active, as indicated by a TEE of 13.2 MJ/day (1.9), an AEE of

256 4.9 MJ/day (1.1) and a PAL of 1.90 (0.20). SWP-derived daily overall PA time was 161.8 min/day (56.3). This
257 included 66.8 min/day (34.1) walking and 8.1 min/day (10.5) running, corresponding to a total of 10077 steps/day
258 (2834) and 7.4 min/day (9.2) of SWP-derived VPA.

259 - Please insert Table 1 near here -

260 **Fig 1a** presents the changes in BM and composition throughout the whole mission and **Table 2** details the effect
261 of at least 3 months in space on the main outcomes. Astronauts were in energy balance the year before launch and
262 spaceflight had modest impact on BM, FM and FFM. Compared to preflight values, BM decreased by 1.20 kg (SE
263 0.50; $P=0.04$) mainly due to a non-significant reduction in FFM of 0.94 kg (0.59; $P=0.14$).

264 - Please insert Figure 1 near here -

265 No significant overall changes in TEE, RMR and AEE were noted between preflight and flight (**Table 2**).
266 However, inflight SWP-derived ambulatory activities dramatically dropped with daily steps decreasing by 6583
267 steps/day (SE 730; $P<0.001$) and walking time by 63.2 min/day (SE 11.5; $P<0.001$). While SWP-derived running
268 time remained stable at 14.4 min/day (SD 6.4), in agreement with the 13.3 min/day (SD 4.6) of self-reported time
269 spent exercising on the T2, this reduction in ambulatory activities was almost fully compensated by an increase in
270 SWP-derived non-ambulatory activities of 64.8 min/day (SE 18.8; $P<0.01$). This explained the absence of
271 significant net changes in SWP-derived overall active time and accelerometry MAD, a proxy of overall PA
272 workload. Daily SWP-derived-VPA was not significantly different from ground values. The one-hour increase in
273 non-ambulatory activities, leading to a total of 151.8 min/day (SE 18.9; range 119.7-194.5), was only partially
274 explained by the 10.7 min/day (SD 4.4) spent on the CEVIS and the about 29.0 min/day (SD 14.3) on the ARED
275 as reported by the astronauts (**Supplemental Table 1**). This suggested an increase in PA not related to physical
276 training. Overall, the astronauts reported 6.2 aerobic exercise sessions/week (SD 1.4; range 3.6-8.6) corresponding
277 to a total duration of 167.8 min/week (SD 43.9; range 86.9-257.8). They reported 4.5 resistance training
278 sessions/week (SD 1.8; range 1.8-6.6), consisting of 5.9-10.2 exercises using 8.8 to 15.1 repetitions for 2.2-3.2
279 sets. This led to a total of 1481 repetitions/week (SD 834; range 468-3044) and a muscular time under tension of
280 81.8 min/week (SD 43.5; range 28.8-161.2). These numbers exhibit a great inter-individual variability in the
281 adherence to the exercise prescriptions. They are to be compared to the 180-270 min/week aerobic, and 6-resistive
282 exercise sessions/week (corresponding to about 1500 repetitions/week) recommendations [21].

283 The astronauts were stratified into two groups, according to their inflight TEE changes, i.e. no change in TEE or
284 decrease in TEE compared to preflight values. Fortuitously, the two groups ended up having almost equal number
285 of subjects.

286 - Please insert Table 2 near here -

287 **3.2 Baseline Characteristics of inflight-TEE-changes groups**

288 At baseline (**Table 1**), both groups were of the same age and had similar FMI but, compared to the decreased_TEE
289 group, the stable_TEE astronauts had higher FFMI (20.6 kg/m² [SD 1.5] vs 18.3 kg/m² [1.6]; $P=0.03$) and
290 presented a more active profile. TEE, AEE and daily SWP-derived overall active time did not differ between the
291 two groups. However, the stable_TEE astronauts spent more time on ground in SWP-derived VPA (12.4 min/day
292 [9.8] vs 1.3 min/day [2.6]; $P=0.04$) and running ($P=0.04$), and had higher running-related MAD ($P=0.04$) resulting
293 in greater SWP-derived mean activity intensity (4.9 milli-g/min [0.9] vs 3.5 milli-g/min [0.9]; $P=0.03$) than the
294 decreased_TEE astronauts.

295

296 **3.3 Anthropometric changes by inflight-TEE-changes groups**

297 Compared to preflight, both groups lost equivalent BM, but contrasted changes in body composition were noted
298 (**Table 2** and **Fig 1a and 1b**). While the stable_TEE group maintained FFMI and displayed a slight FMI loss, their
299 counterparts lost FFMI and gained FMI. The EGD for FFMI changes (stable_TEE compared to decreased_TEE
300 group) was +0.87 kg/m² (95%CI 0.32 to 1.41; effect size=1.7; $P<0.01$); and the EGD for FMI changes -1.09 kg/m²
301 (95%CI -2.06 to -0.11; effect size=1.2; $P=0.03$).

302

303 **3.4 Energy expenditures changes by inflight-TEE-changes groups**

304 The EGD for TEE changes adjusted for BM was +2.98 MJ/day (95%CI 1.75 to 4.22; effect size=2.6; $P<0.001$),
305 mainly related to differences in AEE changes (BM-adjusted EGD +3.02 MJ/day, 95%CI 1.81 to 4.23; effect
306 size=2.7; $P<0.001$) while RMR remained stable in both groups (**Table 2** and **Fig 2a**). As illustrated in **Fig 2b**, the
307 individual FMI net changes were negatively associated with net changes in TEE ($R^2=0.58$; $P<0.01$) and AEE
308 ($R^2=0.54$; $P=0.01$) while net changes in FFMI were positively associated with net changes in TEE ($R^2=0.64$;
309 $P<0.01$) and, to a lesser degree, net changes in AEE ($R^2=0.47$; $P=0.02$).

310

- Please insert Figure 2 near here -

311

312 **3.5 Physical activity changes by inflight-TEE-changes groups**

313 Net changes in SWP and accelerometry-derived activity parameters (**Table 2** and **Fig 3a**) were globally similar
314 across groups. However, like what was observed on the ground, the stable_TEE astronauts still spent more time
315 inflight in SWP-derived VPA (16.7 min/day [SE 3.1] vs 4.6 min/day [3.4]; $P=0.02$), had higher overall-activity
316 accelerometry MAD (884 milli-g/day [125] vs 505 milli-g/day [140]; $P=0.06$), and SWP-derived mean activity
317 intensity (4.60 milli-g/min [0.41] vs 3.35 milli-g/min [0.45]; $P=0.06$) than the decreased_TEE group. Reported
318 inflight resistance training characteristics and time spent performing CEVIS were not significantly different
319 between groups (**Supplemental Table 1**). The stable_TEE group reported non-significant higher inflight aerobic
320 workloads than the decreased_TEE group ($P=0.10$), due to more time spent on T2 (15.7 min/day [SD 3.0] vs 10.4
321 min/day [4.6]; $P=0.05$) but also to higher average speeds (11.2 [0.9] vs 8.5 [2.1] km/h; $P=0.02$). This was in good
322 agreement with higher SWP-derived running-times (18.0 min/day [SE 3.3] vs 9.9 min/day [3.7]; $P=0.12$) and
323 running-related accelerometry MAD (461 milli-g/day [SE 87] vs 154 milli-g/day [98]; $P=0.03$)

324 As illustrated in **Fig 3b**, after at least 3 months on the ISS individuals' inflight FMI were inversely associated with
325 SWP-derived overall activity-related accelerometry MAD ($R^2=0.73$; $P<0.01$), time spent in SWP-derived VPA
326 ($R^2=0.56$; $P=0.02$) and self-reported T2 relative workload ($R^2=0.71$; $P=0.001$). Conversely FFMI values were
327 positively associated with SWP-derived overall activity-related accelerometry MAD ($R^2=0.48$; $P=0.04$), time
328 spent in SWP-derived VPA ($R^2=0.62$; $P=0.01$) and self-reported T2 relative workload ($R^2=0.40$; $P=0.04$). Practice
329 of ARED and CEVIS was not associated with any body composition parameter.

330

- Please insert Figure 3 near here -

331

332 **3.6 Energy intakes by inflight-TEE-changes groups**

333 EI (BM-adjusted) calculated from changes in body composition from the time of beginning of space flight to the
334 time of starting the inflight TEE measurement was 13.6 MJ/day (SE 0.5) in the stable_TEE group and 11.3 MJ/day
335 (0.6) in the decreased_TEE group, representing 93% and 75% of DRIs, respectively (**Fig 4**).

336

- Please insert Figure 4 near here -

337

338 **4 DISCUSSION**

339 The aim of the ENERGY study was to assess the regulation of energy balance and body composition during 6-
340 month missions onboard the ISS by specifically considering the energy cost of PA using the gold standard DLW
341 method. Despite no significant overall changes in body composition and energy expenditures, a slight decrease in
342 BM (-1.5%) was observed in the whole group of astronauts. A large inter-individual variability was however
343 noticed. Astronauts who maintained pre-flight total and activity energy expenditures kept fat-free mass at baseline
344 levels but lost fat mass. Conversely, those who expended less energy during the flight than on the ground lost fat-
345 free mass and gained fat mass. Astronauts who maintained stable TEE during the flight spent more time inflight
346 running and engaged in VPA, and were also the ones with the best fitness on the ground. These results suggest
347 that 1) inflight AEE, possibly due to both physical training and non-exercise PA, drives inflight body composition
348 regulation, 2) inflight energy requirements should be individually evaluated during the whole duration of the
349 spaceflight, 3) baseline participants' characteristics need to be considered for the prescription of both preflight and
350 inflight exercise training; 4) fat mass changes result from unmatched spontaneous EI adaptation to changes in AEE
351 in space that require further investigation.

352

353 **4.1 Ground fitness and PA performance influence changes in body composition during spaceflight**

354 After at least 3 months onboard the ISS, the 11 astronauts presented a slight BM loss, mainly due to a reduction in
355 FFM, despite no significant changes in TEE. The modest effect of spaceflight on BM and body composition in the
356 whole group masked a large interindividual variability with half of the group of astronauts who maintained a stable
357 FFM favoring a slight BM loss (stable_TEE group), while the other half displayed a FFM loss but an increase in
358 FM (decreased_TEE group). This between-subject variability seemed to be associated with heterogeneity in
359 astronauts' ground body composition and engagement in exercise of high intensity [15]. Although all the astronauts
360 were quite fit and active on the ground, those who maintained their FFM during the flight had higher FFMI and
361 spent more time running or in VPA on Earth. These results are in line with the study of Matsumoto et al. [6] that
362 showed habitual PA on Earth was a better predictor of inflight BM loss than PA performed during spaceflight.

363 Whether habitual ground activity levels affect inflight body mass and composition directly or are due to greater
364 engagement of fitter astronauts in exercise during the mission requires further investigation. Of interest, ground
365 differences in SWP-derived VPA and running persisted during the flight with a significant reduction in inflight
366 energy expenditures in the astronauts who were the least active on Earth, but not in the others. We also observed
367 significant associations of inflight self-reported running or SWP and accelerometry proxies of PA workload and
368 intensity with body composition. While improvement of exercise hardware capabilities and prescription have led
369 to better post-flight performances, predicting appropriate inflight exercise loads remains difficult because of the
370 difference in training environment (gravity vs microgravity) and hardware [20, 21]. Beyond their energetics
371 implications, our findings indicate that gaining a better understanding of the associations between ground and
372 inflight exercise characteristics may help designing preflight and inflight prescriptions that favor adherence and
373 enhance inflight performance including for the occupation-related astronaut tasks. Altogether these data suggest
374 that pre-flight astronauts' individual anthropometric and fitness characteristics require more attention for onboard
375 exercise prescriptions and EI recommendations.

376

377 **4.2 Does the exercise countermeasure play a role in the regulation of body composition?**

378 The strong associations of TEE and AEE changes with FFMI and FMI changes suggest that activity-related energy
379 expenditure, in interaction with microgravity exposure, plays a role in inflight body composition changes. In the
380 absence of gravity, TEE is driven by the exercise countermeasure and intra- and extra-vehicular activities while
381 energy expenditure related to standing posture and weight-bearing muscle activity is suppressed. For the first time,
382 SWP and accelerometry used in this study gave a more detailed insight into onboard overall PA patterns. It clearly
383 highlighted the expected decrease in ambulatory activities, except for running activity performed on the T2
384 treadmill. Contrary to our expectations, averaged overall PA time and workload, as estimated by SWP and
385 accelerometry MAD, were not reduced on the ISS; the decrease in ambulatory activities was compensated by non-
386 ambulatory activities. Of interest, accelerometry MAD removes from accelerometry signal any static gravitation
387 component [26] and SWP utilizes the signature of several physiological systems (temperature, sweat, and heat flux
388 and dissipation) in addition to accelerometry, which allows for the disambiguation of activities and contexts (e.g.
389 microgravity) that may confuse a single sensor [25]. Inflight non-ambulatory activities (almost 2.5 hours/day) can
390 only be partially explained by the CEVIS and ARED exercises, which represent about 40 min/day altogether. This

391 suggests that routine and off-nominal mission tasks may be another important component of overall PA.
392 Unfortunately, the identification of specific onboard activities based on the SWP other than walking and running,
393 was not possible due to the arm-placement of the device and the absence of activity learning studies in
394 microgravitational conditions.

395 Mean self-reported training exercises were slightly lower than the recommendations. This was in agreement with
396 previous observations made on the ISS [28]. The lower levels of aerobic exercise reflect a recent tendency to favor
397 resistive exercise for its effect on muscle and bone mass and strength [29, 30]. More importantly a large inter-
398 individual variability in exercise adherence was observed with some astronauts reporting no more than 87
399 min/week of aerobic exercise and less than 500 repetitions/week in resistive exercise with a muscular time under
400 tension lower than 30 min/week.

401 Even if efficiency is lower in space than on the ground, AEE does not seem to be fully explained by exercise
402 training inflight. Treadmill exercise, characterized by higher weekly duration and speed in the astronauts who
403 maintained their TEE, was undoubtedly a contributor of higher AEE and was inversely associated with astronauts'
404 FM changes. Conversely both inflight SWP-derived VPA and T2 workload were positively associated with FFM
405 confirming the protective role of exercise intensity on muscle mass [30]. Of note, time spent on the treadmill alone
406 did not seem to explain the between-group differences in SWP-derived VPA. This suggests that VPA may also
407 reflect an engagement in energy-demanding mission-related activities. However, any extrapolation from SWP
408 sensors about the exact energy cost of both non-training and specific training activities in the context of
409 spaceflights was not possible. Although resistive exercise is considered important to prevent muscle mass and
410 bone loss, we surprisingly did not find any relationship between ARED practice and FFM.

411

412 **4.3 Energy intakes during long-term spaceflight**

413 Even if, by reviewing data from past space missions, no association was found between inflight unadjusted EI and
414 BM loss [6], insufficient EI has often been reported during long-term missions [7, 13]. Astronauts in this study
415 consumed about 85% of dietary prescriptions [19], which is similar to self-reported values observed by Smith et
416 al.[7] during past missions onboard the ISS. Hyporexia due to lower food attractivity, altered smell or taste,
417 sickness, or other microgravity-induced disorders [8] are expected to impair the spontaneous adjustment of EI to

418 the changes in energy needs [7, 31]. This may explain why the astronauts who maintained their TEE to preflight
419 values lost FM despite EI close to their theoretical energy requirements. Conversely, the reduction in EI observed
420 in the less active astronauts was expected as their TEE was reduced by about 20%, due to a decrease in AEE.
421 However, despite EI only representing 75% of the dietary recommendations, the astronauts who displayed a
422 decrease in TEE during spaceflight were still in positive energy and fat balance.

423

424 **4.4 How to maintain energy balance during long-term spaceflight?**

425 Exercise countermeasure is the cornerstone of countermeasure programs during human spaceflights. Exercise
426 training is known to benefit several physiological systems, including muscle strength, bone density and aerobic
427 capacity, but effects of the exercise on these health outcomes were beyond the scope of this study. Here we showed
428 that FFM was maintained in the stable_TEE astronauts despite some of them completing less than the prescribed
429 exercise sessions. Our results further suggest that the non-ambulatory non-training activities play a role in the
430 regulation of energy balance. The extent to which these activities, likely related to the occupational tasks on the
431 ISS, influence energy expenditures and body composition remains to be addressed. For example, it is not known
432 how it contributes to the preservation of muscle mass or other critical key performance outcomes. This finding has
433 potential implications for future spaceflights as it calls for considering non-training activities and their associated
434 energy cost when developing the exercise countermeasure [15], especially for long term space missions.

435 This does not preclude the need for improving strategies to help astronauts to comply with the exercise
436 prescriptions. Space agencies must find the best combination between modalities, i.e. duration and intensity for
437 the aerobic exercise, and load and repetitions for the resistive exercise, to benefit most of the physiological systems
438 without affecting others. For example, using high intermittent interval training (HIIT) that efficiently stimulates
439 musculoskeletal and cardiovascular systems with a relatively low impact on energy requirements may be a
440 promising alternative [8, 15, 23, 32]. Other countermeasure such as nutrition or artificial gravity may also be used
441 to optimize the benefits of exercise and PA. In parallel, changes in the agenda of the astronauts could be considered
442 to improve time dedicated to meals for the whole crew together, and food could be further improved to favor a
443 better adjustment to actual energy needs [33, 34].

444 Overall, the results of this study show that both the measurement and regulation of energy balance inflight is
445 challenging. One of the key findings is the large between-subject variability in changes in TEE, AEE and body
446 composition. This variability was previously reported for EI. This has challenging consequences for both exercise
447 and EI prescriptions as a complex tradeoff needs to be assessed. Such a tradeoff would need to consider both the
448 positive impact on health and various physiological functions of high AEE along with adequate EI; but also the
449 negative impact of too low AEE and excessive FM gain. This is why research on both exercise countermeasures
450 and the development of loggers accurately assessing on real-time exercise, energy expenditure and body
451 composition at the individual level is a top priority. Clearly current available devices (along with their algorithms)
452 do not allow to do so and further interdisciplinary studies are needed.

453

454 **4.5 Limitations and strengths of the study**

455 Spaceflight imposes some limitations and strengths that need to be acknowledged. The sample size was relatively
456 low, due to the limited number of inflight experiments. This study was conducted in male astronauts only, and
457 results may not apply to women. Because space flights are limited, this study is one of the only three studies that
458 determined energy requirements and body composition changes inflight using gold standard objective methods
459 such as DLW and activity monitors, but it is the first one to focus on long-duration missions. Utilization of a unique
460 combination of sensors for continuous physiological monitoring related to PA is a strength of the SWP device. As
461 already underlined, the activity classification algorithms were however developed for detecting activities on Earth,
462 limiting their current recognition capabilities and energy extrapolations in the context of spaceflights.
463 Unfortunately, the R&D service of the SenseWearPro company no longer exists and the SWP software is no longer
464 updated. The current version of the software only provides one-min aggregate measures rather than raw signals,
465 which precludes any additional data treatment and validation. Also, heart rate monitoring, an additional valuable
466 physiological signal in the context of exercise and energy evaluation, was available for few astronauts and during
467 the training sessions only. On the other side, making assumptions on exercise performance based on crewmember
468 exercise logs was challenging and not possible for non-training activities. Many of the findings are correlations, which
469 suggests the need for future studies to prove causation.

470

471 **5 CONCLUSION**

472 The between-astronauts variability in body composition and energy expenditure changes observed during
473 spaceflight was related to ground fitness, and inflight practice of activities of high intensity. Importantly, a high
474 inter-individual variability in inflight training was noted with some crewmembers reporting values far below the
475 recommendations, but there was an unexpected engagement in non-ambulatory activities related to the nature of
476 the missions onboard the ISS. These results suggest that energy requirements in space must be individually derived
477 based on real-time measurements of AEE and changes in body composition rather than on current general
478 recommendations and exercise prescriptions. This requires validating in space the use of tri-accelerometry on
479 different body parts, along with other sensors including heart rate monitors, and the development of specific
480 algorithms to detect and quantify all physical activities and derive activity-specific energy expenditures during the
481 space missions. Despite some spontaneous adjustments, we further observed an uncoupling between energy
482 intakes and expenditures that led to energy imbalance. Methodological developments are therefore vital for the
483 control of both sides of the energy balance, i.e., energy intakes and expenditures, or at least body composition
484 evolution during long-duration space missions, which are considered a top priority for exploration by the
485 international space agencies.

486

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574

575 **Table1:** Astronauts' baseline characteristics for the whole group and by inflight-TEE-changes groups

	All Astronauts (n=11)	Astronauts with stable TEE (n=6)	Astronauts with decreased TEE (n=5)	Group difference P Value ^b
Age & anthropometry				
Age (yr)	45.7 (7.7)	43.5 (8.7)	48.4 (6.0)	0.32
Height (cm)	180.3 (6.7)	182.0 (5.7)	178.3 (7.9)	0.39
BM (kg)	79.4 (10.6)	83.9 (8.0)	73.9 (11.6)	0.13
BMI (kg/m ²)	24.3 (2.1)	25.3 (1.4)	23.2 (2.4)	0.10
FFM (kg)	63.8 (8.8)	68.3 (6.0)	58.3 (8.9)	0.05
FFMI (kg/m ²)	19.6 (1.9)	20.6 (1.5)	18.3 (1.6)	0.03
FM (kg)	15.6 (3.9)	15.57 (4.5)	15.6 (3.5)	0.99
FMI (kg/m ²)	4.8 (1.0)	4.7 (1.2)	4.9 (1.0)	0.71
Energetics				
TEE (MJ/day)	13.2 (1.9)	14.0 (1.4)	12.2 (2.1)	0.13
TEE _{BM} (MJ/day) ^a	13.2 (1.0)	13.3 (1.2)	13.0 (1.3)	0.70
RMR (MJ/day)	7.0 (1.2)	7.4 (1.4)	6.6 (0.8)	0.32
RMR _{FFM & FM} (MJ/day) ^a	7.0 (1.0)	7.0 (1.3)	7.0 (1.4)	0.93
AEE (MJ/day)	4.9 (1.1)	5.2 (1.0)	4.4 (1.1)	0.24
AEE _{BM} (MJ/day) ^a	4.9 (0.9)	5.0 (1.1)	4.7 (1.1)	0.71
PAL	1.90 (0.20)	1.94 (0.26)	1.85 (0.09)	0.50
Energy intake (MJ/day)	13.2 (1.9)	14.0 (1.4)	12.2 (2.1)	0.13
Energy intake _{BM} (MJ/day) ^a	13.2 (1.1)	13.3 (1.2)	13.0 (1.3)	0.70
SWP-derived physical activities				
Steps (number/day)	10077 (2834)	1046 (2816)	9614 (3111)	0.65
Overall activity & exercise (min/day)	161.8 (56.3)	164.8 (67.5)	158.3 (47.0)	0.86
Walking (min/day)	66.8 (34.1)	60.2 (27.1)	74.6 (43.1)	0.53
Running (min/day)	8.1 (10.5)	13.7 (11.5)	1.5 (2.8)	0.04
Non-ambulatory activity & exercise (min/day)	87.9 (63.4)	90.9 (61.7)	82.3 (72.4)	0.84
Vigorous physical activity (MAD>16 milli-g; min/day)	7.4 (9.2)	12.4 (9.8)	1.3 (2.6)	0.04
Accelerometry MAD				
Daily total (milli-g/day)	1353 (278)	1487 (309)	1193 (124)	0.08
Overall activity & exercise (milli-g/day)	692 (313)	814 (368)	546 (164)	0.17
Walking (milli-g/day)	286 (171)	250 (104)	329 (235)	0.47
Running (milli-g/day)	188 (242)	322 (259)	27 (54)	0.04
Non-ambulatory activity & exercise (milli-g/day)	218 (141)	241 (151)	190 (140)	0.58

Overall activity & exercise mean intensity (milli-g/min)	4.3 (1.1)	4.9 (0.9)	3.5 (0.9)	0.03
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576 BMI=body mass index; FM=fat mass; FFM=fat-free mass; FMI=fat mass index; FFMI=fat-free mass index;
577 TEE=total energy expenditure, RMR=resting metabolism rate, AEE=activity energy expenditure; EI=energy
578 intake MAD=acceleration mean absolute deviation. Values are means (SD). SWP =Sensewear Pro activity-device.
579 Physical activities were derived from a combination of different physiological signals with an in-built machine-
580 learning activity classification algorithm.
581 ^a BM (TEE, AEE, EI) or FFM and FM (RMR) adjusted lsmeans (SD) (whole group mean baseline values were
582 used as reference values). All astronauts were male.
583 ^b Statistical analyses used Student's t-tests.
584

585 **Table2:** Astronauts' anthropometric, energetic and physical activity changes between preflight and flight for the whole group and by inflight-TEE-changes groups

	All Astronauts (n=11)		Astronauts with stable TEE (n=6)	Astronauts with decreased TEE (n=5)	Linear Mixed Model Analyses		
	Changes from preflight	P value	Changes from preflight	Changes from preflight	EGD in changes from preflight	95%CI	P value
Age & anthropometry							
BM (kg)	-1.20 (0.50)	0.04	-1.59 (0.69)	-0.74 (0.75)	-0.86 (1.02)	-3.17 to 1.46	0.42
BMI (kg/m ²)	-0.39 (0.17)	0.04	-0.49 (0.24)	-0.28 (0.26)	-0.22 (0.35)	-1.01 to 0.57	0.55
FFM (kg)	-0.94 (0.59)	0.14	0.34 (0.56)	-2.48 (0.61)	2.82 (0.83)	0.94 to 4.69	<0.01
FFM index (kg/m ²)	-0.29 (0.18)	0.13	0.10 (0.16)	-0.76 (0.18)	0.87 (0.24)	0.32 to 1.41	<0.01
FM (kg)	-0.26 (0.87)	0.77	-1.93 (0.93)	1.74 (1.02)	-3.67 (1.38)	-6.78 to -0.56	0.03
FM index (kg/m ²)	-0.11 (0.27)	0.70	-0.60 (0.29)	0.49 (0.32)	-1.09 (0.43)	-2.06 to -0.11	0.03
Energetics							
TEE _{BM} (MJ/day) ^a	-0.39 (0.73)	0.60	0.90 (0.37)	-2.08 (0.40)	2.98 (0.54)	1.75 to 4.22	<0.001
RMR _{FFM & FM} (MJ/day) ^a	-0.15 (0.19)	0.43	-0.35 (0.28)	0.11 (0.33)	-0.46 (0.48)	-1.51 to 0.59	0.36
AEE _{BM} (MJ/day) ^a	-0.19 (0.70)	0.79	1.12 (0.36)	-1.90 (0.39)	3.02 (0.53)	1.81 to 4.23	<0.001
PAL	-0.02 (0.10)	0.86	0.23 (0.08)	-0.31 (0.09)	0.54 (0.12)	0.28 to 0.80	0.001
Energy intake (MJ/day)	-0.82 (0.38)	0.06	0.06 (0.34)	-1.86 (0.37)	1.92 (0.50)	0.78 to 3.05	<0.01
Energy intake _{BM} (MJ/day) ^a	-0.61 (0.40)	0.16	0.29 (0.33)	-1.75 (0.36)	2.04 (0.48)	0.96 to 3.13	<0.01
Energy balance (MJ/day)	-0.15 (0.29)	0.61	-0.68 (0.33)	0.48 (0.36)	-1.15 (0.49)	-2.19 to -0.12	0.03
Energy balance _{BM} (MJ/day) ^a	-0.14 (0.30)	0.65	-0.63 (0.30)	0.50 (0.33)	-1.13 (0.44)	-2.06 to -0.20	0.02
SWP-derived physical activities							
Steps (number/day)	-6583 (730)	<0.001	-6277 (1402)	-7231 (1554)	954 (2093)	-3483 to 5391	0.66

Overall activity & exercise (min/day)	13.2 (23.8)	0.59	24.4 (33.2)	-0.9 (36.8)	25.2 (49.6)	-79.9 to 130.3	0.62
Walking (min/day)	-63.2 (11.5)	<0.001	-57.3 (16.0)	-70.1 (17.7)	12.8 (23.9)	-37.8 to 63.4	0.60
Running (min/day)	6.3 (4.0)	0.13	4.4 (4.5)	8.5 (5.0)	-4.1 (6.7)	-18.3 to 10.1	0.55
Non-ambulatory activity & exercise (min/day)	64.8 (18.8)	<0.01	77.3 (36.3)	60.8 (40.2)	16.5 (54.2)	-98.3 to 131.4	0.76
Vigorous physical activity (MAD>16 milli-g; min/day)	2.8 (2.1)	0.23	4.3 (4.2)	3.3 (4.6)	1.0 (6.2)	-12.2 to 14.1	0.88
Accelerometry MAD							
Daily total (milli-g/day)	-40.8 (74.7)	0.60	12.6 (147.3)	-67.9 (163.1)	80.5 (219.8)	-385.4 to 546.4	0.72
Overall activity & exercise (milli-g/day)	11.1 (93.2)	0.91	70.2 (169.0)	-40.4 (187.2)	110.7 (252.2)	-424.0 to 645.4	0.67
Walking (milli-g/day)	-264.6 (57.3)	<0.001	-235.0 (79.4)	-299.9 (87.9)	64.9 (118.5)	-186.2 to 316.1	0.59
Running (milli-g/day)	113.2 (73.8)	0.16	138.4 (118.3)	127.5 (131.1)	10.9 (176.6)	-363.4 to 385.3	0.95
Non-ambulatory activity & exercise (milli-g/day)	151.8 (60.9)	0.02	166.8 (83.8)	132.0 (92.9)	34.8 (125.1)	-230.4 to 300.0	0.78
Overall activity & exercise mean intensity (milli-g/min)	-0.23 (0.35)	0.53	-0.32 (0.55)	-0.16 (0.61)	-0.16 (0.82)	-1.90 to 1.58	0.84
Activity NASA logs ^c							
T2 (min/day)	13.3 (1.4)	.	15.7 (1.6)	10.4 (1.7)	-5.3 (2.3)	-10.5 to -0.1	0.04
CEVIS (min/day)	10.7 (1.3)	.	10.0 (1.9)	11.4 (2.1)	1.4 (2.8)	-4.9 to 7.7	0.63
ARED (min/day) ^d	29.0 (4.3)	.	30.9 (6.1)	26.8 (6.7)	-4.1 (9.0)	-24.5 to 16.4	0.66

586 BMI=body mass index; FM=fat mass; FFM=fat-free mass; FMI=fat mass index; FFMI=fat-free mass index; TEE=total energy expenditure, RMR=resting metabolism rate,
587 AEE=activity energy expenditure; EI=energy intake; EB=energy balance; MAD=Acceleration mean absolute deviation; T2= treadmill device onboard the ISS; CEVIS= Cycle
588 Ergometer with Vibration Isolation and Stabilization system; ARED= Advanced Resistive Exercise Device. SWP =Sensewear Pro device.

589 Physical activities were derived from a combination of different physiological signals with an in-built machine-learning activity classification algorithm.

590 Values are estimated Lsmeans (SE) from a mixed-effects models for repeated measurements.

591 ^a BM (TEE, AEE, EI, EB) or FFM and FM (RMR) adjusted Models (whole group mean baseline values were used as reference values).

592 ^c Inflight Values

593 ^d ARED duration was approximate from number of repetitions and sets (2-to5 sec/repetition, according to movement complexity, with addition of 1-min recuperation time by
594 set)

595

596 **Legends**

597 **Fig 1:** Body mass and composition changes during flight

598 Changes in body mass and composition throughout the whole mission (a) and during the experimental sessions
599 (b). Values are means (SE) (a), and lsmeans (SE) (b) from mixed-effects models for repeated measurements with
600 estimated group differences (EGD) presented with their 95% confidence interval; * $P < 0.05$; ** $P < 0.01$ (b).

601 BM= body mass; FFM= fat-free mass; FM= fat mass; DXA= dual X-ray absorptiometry; SLAMMD= Space
602 Linear Acceleration Mass Measurement Device; DLW= doubly labeled water

603

604 **Fig 2:** Energy expenditure changes during flight

605 Changes in total energy expenditure and its components during the experimental sessions (a) and scatterplots for
606 the relationship between body composition changes and energy expenditure components changes (b). Values are
607 lsmeans (SE) from mixed-effects models for repeated measurements with estimated group differences (EGD)
608 presented with their 95% confidence interval; *** $P < 0.001$ (a). Least square regression lines are plotted with their
609 95% confidence interval in shaded areas (b).

610 TEE_{BM}= total energy expenditure adjusted for body mass; AEE_{BM}= activity-related energy expenditure adjusted
611 for body mass; RMR_{FFM & FM}= resting metabolic rate adjusted for fat-free mass and fat mass; FFMI= fat-free mass
612 index; FMI= fat mass index.

613

614 **Fig 3:** Physical activity changes during flight

615 Changes in SWP-derived activity duration and acceleration-MAD (as a proxy of activity workload) during the
616 experimental sessions (a) and scatterplots for the relationship between inflight body composition and inflight SWP-
617 derived or reported physical activity (b). Values are lsmeans (SE) from mixed-effects models for repeated
618 measurements (a). Least square regression lines are plotted with their 95% confidence interval in shaded areas;
619 individual values are available for 9 astronauts only (b).

620 FFMI= fat free mass index; FMI= fat mass index; T2= treadmill device onboard the ISS; SWP= Sensewear Pro
621 activity monitor; MAD= acceleration mean amplitude deviation; VPA= vigorous physical activity (MAD>16
622 milli-g)

623

624 **Fig 4:** Estimated energy intakes during flight

625 Percentage of energy intake (EI) estimated from the changes in body composition and total energy expenditure
626 between the preflight and inflight experimental sessions from the 2001 Dietary Reference Intakes (DRIs)^[19] for
627 each groups stratified for inflight TEE changes. Values are individual data and mean (SE)