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| **Supplementary material 1.**  |
|  | **Middle-aged** **50-59****n= 27** | **Older****60+****n=27** |  |
| **Females %** | 37 | 22 |  |
| **Variables** |  |  |  |
| Age, years | 52.0 (54.3 to 53.1) | 64.6 (63.0 to 66.4) |  |
| Height, m | 1.71 (1.67 to 1.75) | 1.71 (1.67 to 1.75) | 0.926 |
| Weight, kg | 74.7 (68.5 to 80.9)\_ | 74.5 (69.2 to 80.3) | 0.937 |
| BMI, kg‧m2 | 25.4 (23.4 to 27.3) | 25.7 (24.0 to 27.4) | 0.788 |
| **Exercise volume**  |  |  |  |
| Ex volume, min/week | 228.8 (183.7 to 274.0) | 227.9 (172.0 to 283.8) | 0.979 |
| BGL, mmol/l | 4.9 (4.5 to 5.4) | 5.0 (4.8 to 5.3) | 0.195 |
| Insulin, ulU/mL | 7.5 (4.7 to 9.5) | 6.7 (5.8 to 8.0) | 0.450.1871 |
| IGF-1, pg‧mL-1 | 72.3 (25.6 to 118.9) | 224.1 (-61.4 to 509.5) | 0.366 |
| Testosterone, ng‧mL-1 | 1.0 (0.45 to 1.6) | 1.9 (1.2 to 2.6) | 0.457 |
| Estradiol, pg‧mL-1 | 30.5 (5.3 to 55.6) | 26.6 (3.6 to 49.5) | 0.683 |
| Cortisol, ng‧mL-1 | 361.0 (284.6 to 438.0) | 393.0 ( 297.8 to 488.8) | 0.615 |
| WBC, pg‧mL-1 | 4.85 (4.3 to 5.3) | 5.6 (5.0 to 6.2) | T(2.1) 49=0.037 |
| Neutrophils, pg‧mL-1 | 2.5 (2.2 to 3.0) | 3.0 (2.6 to 3.2) | 0.168 |
| Lymphocytes, pg‧mL-1 | 2.0 (1.7 to 2.2) | 2.7 (2.0 to 2.6) | 0.160 |
| Monocytes, pg‧mL-1 | 0.35 (0.25 to 0.44) | 0.33 (3.0 to 4.0) | 0.294 |
| IL2, pg‧mL-1 | 4.6 (2.9 to 5.8) | 3.7 (2.8 to 4.6) | 0.531 |
| IL6, pg‧mL-1 | 5.0 (0.87 to 8.3) | 6.7 (2.5 to 11.4) | 0.557 |
| IL8, pg‧mL-1 | 5.7 (1.9 to 7.5) | 6.6 ( 3.3 to 9.9) | 0.742 |
| IL10, pg‧mL-1 | 17.6 (11.0 to 25.6) | 19.4 (13.7 to 25.8) | 0.545 |
| TNF-α, pg‧mL-1 | 2.2 (1.3 to 2.7) | 2.3 (2.0 to 2.8) | 0.545 |
| Mean (95% CI) are shown for continuous variablesAbbreviations: ADM, Appendicular muscle mass, BGL, blood glucose levels, BM, body mass, BMI, body mass index, BMD, bone mineral density, FFM, Fat free mass, FM, fat mass, IGF, insulin-like growth factor, IL, interleukin, PRO, protein, RMR, resting metabolic rate, RQ, respiratory quotient, TLM, Total lean mass, WBC, white blood cellsns, not significant † *P*<0.05, †† *P*<0.01, †† *P*<0.001 significant differences between groups |

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| **Supplementary material 2. .**  |
|  | **Low training (≤149-90 minutes, n=16)**  | **Moderate (≥150-299, n=24)**  | **High training 300+ (n=14)** | ***P*** |
| **Variables**  | 50% female  | 21% female | 35% female  |  |
| Age (years)  | 60.1 (56.06 to 64) | 58.4 (55.6 to 61) | 58.8 (57.0 to 60.7) | 0.720 |
| Height (cm) | 1.68 (1.63 to 1.74) | 1.75 (1.71 to 1.78) | 1.71 (1.68 to 1.74) | 0.065 |
| Weight (kg) | 78.0 (69.9 to 86.1) | 75.8 (69.9 (81.8)  | 69.5 (60.3 to 78.9) | 0.285 |
| BMI (kg/m2) | 28.0 (25.0 to 30.8) | 24.7 (23.0 to 26.8) | 24.3 (22.2 to 26.8) | 0.044 |
| **Self-reported exercise**  |  |  |  |  |
| Ex volume  | 101.7 (92.3 to 111) | 213.3 (196.5 to 229.9)  | 394.0 (329.1 to 461.3) |  |
| **Biochemistry**  |  |  |  |  |
| BGL, mmol/L | 5.2 (4.8 to 5.6) | 4.9 (4.5 to 5.1) | 4.8 (4.5 to 5.2) | 0.251 |
| Insulin, mIU/L  | 6.6 (4.0 to 9.2) | 7.2 (5.2 to 9.2) | 7.1 (6.1 to 8.1) | 0.456 |
| IGF-1, pg‧mL- | 7.0 (5.2 to 8.7)  | 7.4 (5.7 to 9.2) | 6.7 (5.7 to 7.7) | 0.425 |
| Testosterone, ng/dL  | 0.96 (0.27 to 1.7) | 1.82 (1.3 to 2.3) | 1.5 (0.55 to 2.2) | 0.184 |
| Estradiol, ng/dL | 32.7 (-28.3 to 93.7) | 40.6 (7.4 to 73.9) | 24.7 (0.71 to 48.8) | 0.682 |
| Cortisol, ng‧mL-1 | 353.1 (266.1 to 440.1) | 400.7 (294.1 to 507.4) | 364.4 (257.9 to 470.9) | 0.884 |
| WBC pg‧mL-1 | 5.4 (4.4 to 6.5) | 5.0 (4.4 to5.5) | 5.2 (4.6 to 5.7) | 0.380 |
| Neutrophils, pg‧mL-1 | 2.8 (2.1 to 3.4) | 2.7 (2.3 to 3.1) | 2.5 (1.9 to 3.0) | 0.508 |
| Lymphocytes, pg‧mL-1 | 2.1 (1.7 to 2.3) | 1.9 (1.6 to 2.2) | 2.2 (1.7 to 2.6) | 0.864 |
| Monocytes, pg‧mL-1 | 0.3 (0.23 to 0.36) | 0.33 (0.26 to 0.40) | 0.32 (0.26 to 0.38) | 0.519 |
| IL2, pg‧mL-1 | 3.8 (1.9 to 5.7) | 4.4 (3.0 to 5.0) | 4.3 (3.0 to 5.7) | 0.864 |
| IL6, pg‧mL-1 | 5.9 (-0.16 to 12.06) | 5.32 (1.2 to 9.45)  | 6.7 (2.6 to 10.9) | 0.895 |
| IL8, pg‧mL-1 | 5.9 (2.02 to 9.8) | 4.2 (2.0 to 6.1) | 9.0 (2.8 to 15.1) | 0.164 |
| IL10, pg‧mL-1 | 20.6 (12.6 to 28,6) | 19.4 (13.2 to 25.7) | 14.3 (7.4 to 21.3) | 0.462 |
| TNF, pg‧mL-1 | 2.6 (1.7 to 3.5) | 2.2 (1.6 to 2.7) | 2.1 (1.5 to 2.7) | 0.464 |
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| Mean (95% CI) are shown for continuous variablesAbbreviations: ADM, Appendicular muscle mass, BGL, blood glucose levels, BM, body mass, BMI, body mass index, BMD, bone mineral density, FFM, Fat free mass, FM, fat mass, IGF, insulin-like growth factor, IL, interleukin, PRO, protein, RMR, resting metabolic rate, RQ, respiratory quotient, TLM, Total lean mass, WBC, white blood cellsns, not significant † *P*<0.05, †† *P*<0.01, †† *P*<0.001significant differences between groups |

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| **Supplementary material 3.**  |
|  | **LPI ≤0.8g.kg.day****N=7** | **MPI****>0.8-1.19g/kg****N=12** | **HPI ≥1.2.g.kg.day***N=34* |  **p-value** |
| **% female** **Variables**  |  |  |  |  |
| Age, years  | 61.8 (54.1 to 69.5) | 56.1 (53.2 to 59.0) | 56.6 (52.1 to 61.9) | 0.170 |
| Height, cm | 1.73 (1.64 to 1.8) | 1.71 (1.67 to 1.80) | 1.17 (1.67 to 1.7) | 0.699 |
| Weight, kg | 86.1 (70.1 to 102.1) | 82.3 (72.6 to 92.0) | 69.1 (65.0 to 73.3) | 0.002 |
| BMI, kg/m2 | 28.5 (24.8 to 32.1) | 27.2 (24.1 to 32.1) | 23.5 (22.6 to 24.5) | 0.001 |
|  |  |  |  |  |
| **Training volume**  |  |  |  |  |
| Ex volume (mins/day) | 200 (119.0 to 280.9) | 162.7 (108.4 to 217.1) | 258.2 (210 to 305.5) | 0.091 |
| **Biochemistry**  |  |  |  |  |
| BGL, mmol/L | 5.3 (4.7 to 5.9) | 4.8 (4.5 to 5.2) | 5.0 (4.8 to 5.2) | 0.402 |
| Insulin, mIU/L  | 7.6 (5.3 to10) | 7.7 (4.8 to 10.5) | 6.7 (5.8 to 7.9) | 0.722 |
| IGF-1, pg‧mL- | 138.2 (47.8 to 228.5) | 136.7 [43.8 to 229.5) | 154.3 (74.7 to 410.1) | 0.976 |
| Testosterone,ng/dL  | 1.5 (0.3 to 2.8) | 1.1 (0.4 to 1.7) | 1.7 (1.1 to 2.2) | 0.330 |
| Estradiol, ng/dL | 14.0 (1.8 to 26.1) | 21.8 (15.7 to 59.5) | 35.5 (12.7 to 58.4) | 0.560 |
| Cortisol, ng‧mL-1  | 313.1 (143.4 to 482.9) | 380.8 (277.7 to 483.8) | 374.0 (298.3 to 449.6) | 0.752 |
| WBC, pg‧mL-1Neutrophils, pg‧mL-1 | 4.8 (3.4 to 5.5)2.9 (2.5 to 3.4) | 4.6 (3.7 to 5.6)2.8 (2.3 to 2.9) | 5.0 (4.5 to 5.4)2.6 (2.3 to 2.9) | 0.8320.553 |
| Lymphocytes, pg‧mL-1 | 2.3 (1.6 to 2.9) | 2.3 (1.9 to 2.7) | 2.0 (1.8 to 2.3) | 0.238 |
| Monocytes, pg‧mL-1 | 0.35 (0.23 to 0.47) | 0.38 (0.24 to 0.52) | 0.32 (0.28 to 0.37) | 0.547 |
| IL2, pg‧mL-1 | 3.2 (0.7 to 5.7) | 3.9 (2.6 to 5.2) | 4.4 (3.4 to 5.4) | 0.580 |
| IL6, pg‧mL-1 | 1.5 (-0.1 to 3.1) | 3.8 (0.81 to 6.8) | 7.5 (3.7 to 11.3) | 0.258 |
| IL8, pg‧mL-1 | 1.91 (0.89 to 2.9) | 5.2 (0.77 to 9.5) | 7.1 (4.2 to 10.1) | 0.150 |
| IL10, pg‧mL-1 | 17.0 (2.8 to 31.1) | 18.3( [11.8 to 24.8) | 18.8 (13.3 to 24.4) | 0.872 |
| TNF-α, pg‧mL-1 | 1.8 (1.7 to 2.6) | 2.6 (1.8 to 3.3) | 2.2 (1.7 to 2.7) | 0.475 |
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| **Supplementary table 4**  |
|  | **Males (n= 36)** | **Females (n= 18)** | *P* |
| **Variables**  |  |  |  |
| Age, years  | 59.1 (56.8 to 61.4) | 58.3 (54.8 to 61.9) | ns |
| Height, cm | 1.76 (56.8 to 61.42)  | 1.61 (1.58 to 1.65)  | P<0.01 |
| Weight, kg | 80.2 (75.5 to 84.9) | 64.1 (54.8 to 62.0) | P<0.001 |
| BMI, kg/m2 | 25.77 (24.4 to 27.1) | 25.1 (22.2 to 27.9) | ns  |
| RMR, kcals.kg.day | 18.7 (17.6 to 19.7) | 18.98 (17.2 to 20.6) | 0.810 |
|  |  |  |  |
| **Training volume**  |  |  |  |
| Ex volume (mins/day) | 229.7 (193.4 to 265.6) | 213.1 (134.6 to 291.7) | ns  |
| **Biochemistry**  |  |  |  |
| BGL, mmol/L | 5.1 (4.8 to5.3) | 4.7 (4.4 to 5.1) | P=0.034 (41) 2.2 |
| Insulin, mIU/L  | 8.0 (6.6 to 9.2) | 5.6 (4.8 to 6.3) | 0.013 |
| IGF-1, pg‧mL- | 174.9 (-27.87 to 377.7) | 108.2 (30.8 to 185.6) | 0.549 |
| Testosterone,ng/dL  | 2.06 (1.65 to 2.465) | 0.25 (0 to 0.60) | P<0.001 |
| Estradiol, ng/dL | 31.3 (9.0 to 53.5) | 23.2 (-1.61 to 48.2) | 0.659 |
| Cortisol, ng‧mL-1  | 368.8 (294.2) | 366.6 (473.5 to 360.8) | ns |
| WBC, pg‧mL-1Neutrophils, pg‧mL-1 | 5.3 (4.9 to 5.7)2.8 (2.5 to 3.1) | 5.3 (4.6 to 6.1)2.5 (1.9 to 3.0) | 0.05950.140 |
| Lymphocytes, pg‧mL-1 | 2.1 (1.8 to 2.4) | 2.1 (1.7 to 2.5) | 0.826 |
| Monocytes, pg‧mL-1 | 0.37 (0.3 to 0.44) | 0.27 (0.22 to 0.32) | P=0.034 |
| IL2, pg‧mL-1IL6, pg‧mL-1IL8, pg‧mL-1IL10, pg‧mL-1TNF-α, pg‧mL-1 | 4.1 (3.2 to 5.0)5.8 (2.5 to 9.2)5.6 (3.0 to 8.3) 18.0 (13.1 to 23.0)2.3 (1.8 to 2.7) | 4.4 (2.7 to 6.2)5.8 (1.2 to 10.4)6.1 (2.6 to 9.5)22.0 (14.4 to 29.1)2.4 (1.6 to 3.2) | nsnsnsnsns |
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| Mean (95% CI) are shown for continuous variablesAbbreviations: ADM, Appendicular muscle mass, BGL, blood glucose levels, BM, body mass, BMI, body mass index, BMD, bone mineral density, FFM, Fat free mass, FM, fat mass, IGF, insulin-like growth factor, IL, interleukin, PRO, protein, RMR, resting metabolic rate, RQ, respiratory quotient, TLM, Total lean mass, WBC, white blood cellsns, not significant † *P*<0.05, †† *P*<0.01, †† *P*<0.001significant differences between groups |

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