**SUPPLEMENTARY INFORMATION**

TITLE:

LIPIDOMICS REVEALS SEASONAL SHIFTS IN A LARGE-BODIED HIBERNATOR, THE BROWN BEAR

AUTHORS:

Sylvain Giroud, Isabelle Chery, Fabrice Bertile, Justine Bertrand-Michel, Georg Tascher, Guillemette Gauquelin-Koch, Jon M Arnemo, Jon E. Swenson, Navinder J. Singh, Etienne Lefai, Alina L. Evans, Chantal Simon, Stéphane Blanc

Table S1: Arithmetic means (‘Means’) standard errors (‘SE’) of concentrations (in mmol l-1) of specific fatty acids among total fatty acids in white adipose tissue (‘WAT’), muscle tissue (‘Muscle’) and blood plasma (‘Plasma’) of bears during the summer active period (‘Summer’) and in winter hibernation (‘Winter’). Sample sizes used in the linear mixed-effects models are presented in Table 1. Significant p-values are highlighted in bold. ‘ND’ refers to non-detectable.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Tissues | Fatty acids | Means ± SE | |  | P-values |
|  |  | **Summer** | **Winter** |  |  |
| WAT |  |  |  |  |  |
|  | C10:0 | ND | ND |  |  |
|  | C12:0 | ND | ND |  |  |
|  | C14:0 | 0.01 ± 0.01 | 1.39 ± 0.26 |  | **<0.01** |
|  | C15:0 | ND | ND |  |  |
|  | C16:0 | 0.47 ± 0.20 | 37.65 ± 9.05 |  | **0.01** |
|  | C18:0 | 0.26 ± 0.09 | 11.89 ± 2.34 |  | **<0.01** |
|  | C20:0 | 0.01 ± 0.01 | 0.29 ± 0.02 |  | **<0.001** |
|  | C21:0 | ND | ND |  |  |
|  | C22:0 | 0.00 ± 0.01 | 0.07 ± 0.07 |  | 0.328 |
|  | C23:0 | ND | ND |  |  |
|  | C24:0 | ND | ND |  |  |
|  | C14:1 ω5 | ND | ND |  |  |
|  | C15:1 ω5 | ND | ND |  |  |
|  | C16:1 ω7 | 0.01 ± 0.01 | 0.37 ± 0.09 |  | **0.011** |
|  | C16:1 ω9 | 0.06 ± 0.03 | 8.50 ± 2.08 |  | **0.01** |
|  | C17:1 ω7 | ND | ND |  |  |
|  | C18:1 ω7 | 0.06 ± 0.03 | 3.57 ± 0.93 |  | **0.013** |
|  | C18:1 ω9 | 0.81 ± 0.42 | 67.14 ± 10.95 |  | **<0.01** |
|  | C20:1 ω9 | 0.01 ± 0.01 | 1.03 ± 0.24 |  | **<0.01** |
|  | C22:1 ω9 | ND | ND |  |  |
|  | C24:1 ω9 | ND | ND |  |  |
|  | C18:2 ω6 | 0.07 ± 0.03 | 4.13 ± 1.23 |  | 0.02 |
|  | C20:2 ω6 | 0.00 ± 0.00 | 0.26 ± 0.12 |  | 0.071 |
|  | C22:2 ω6 | ND | ND |  |  |
|  | C18:3 ω3 | 0.02 ± 0.01 | 1.42 ± 0.56 |  | 0.048 |
|  | C18:3 ω6 | ND | ND |  |  |
|  | C20:3 ω3 | 0.00 ± 0.00 | 0.03 ± 0.03 |  | 0.328 |
|  | C20:3 ω6 | 0.00 ± 0.00 | 0.09 ± 0.02 |  | **<0.01** |
|  | C20:4 ω6 | 0.04 ± 0.01 | 0.20 ± 0.02 |  | **0.011** |
|  | C22:4 ω6 | 0.01 ± 0.01 | 0.13 ± 0.01 |  | 0.049 |
|  | C20:5 ω3 | 0.01 ± 0.01 | 0.12 ± 0.05 |  | 0.071 |
|  | C22:5 ω3 | 0.01 ± 0.01 | 0.35 ± 0.12 |  | 0.027 |
|  | C22:5 ω6 | ND | ND |  |  |
|  | C22:6 ω3 | 0.01 ± 0.01 | 0.25 ± 0.24 |  | 0.308 |
| Muscle |  |  |  |  |  |
|  | C10:0 | ND | ND |  |  |
|  | C12:0 | ND | ND |  |  |
|  | C14:0 | ND | ND |  |  |
|  | C15:0 | ND | ND |  |  |
|  | C16:0 | 0.03 ± 0.01 | 0.33 ± 0.27 |  | 0.388 |
|  | C18:0 | 0.02 ± 0.04 | 0.08 ± 0.06 |  | 0.411 |
|  | C20:0 | ND | ND |  |  |
|  | C21:0 | ND | ND |  |  |
|  | C22:0 | ND | ND |  |  |
|  | C23:0 | ND | ND |  |  |
|  | C24:0 | 0.01 ± 0.01 | 0.00 ± 0.00 |  | 0.284 |
|  | C14:1 ω5 | ND | ND |  |  |
|  | C15:1 ω5 | ND | ND |  |  |
|  | C16:1 ω7 | 0.01 ± 0.00 | 0.25 ± 0.25 |  | 0.422 |
|  | C16:1 ω9 | ND | ND |  |  |
|  | C17:1 ω7 | 0.00 ± 0.00 | 0.01 ± 0.01 |  | 0.443 |
|  | C18:1 ω7 | 0.01 ± 0.01 | 0.03 ± 0.03 |  | 0.397 |
|  | C18:1 ω9 | 0.03 ± 0.09 | 0.54 ± 0.45 |  | 0.372 |
|  | C20:1 ω9 | ND | ND |  |  |
|  | C22:1 ω9 | ND | ND |  |  |
|  | C24:1 ω9 | ND | ND |  |  |
|  | C18:2 ω6 | 0.01 ± 0.01 | 0.08 ± 0.07 |  | 0.432 |
|  | C20:2 ω6 | ND | ND |  |  |
|  | C22:2 ω6 | ND | ND |  |  |
|  | C18:3 ω3 | 0.00 ± 0.00 | 0.11 ± 0.11 |  | 0.428 |
|  | C18:3 ω6 | ND | ND |  |  |
|  | C20:3 ω3 | ND | ND |  |  |
|  | C20:3 ω6 | ND | ND |  |  |
|  | C20:4 ω6 | 0.01 ± 0.01 | 0.02 ± 0.02 |  | 0.401 |
|  | C22:4 ω6 | ND | ND |  |  |
|  | C20:5 ω3 | 0.01 ± 0.01 | 0.01 ± 0.01 |  | 0.458 |
|  | C22:5 ω3 | 0.01 ± 0.01 | 0.01 ± 0.01 |  | 0.443 |
|  | C22:5 ω6 | ND | ND |  |  |
|  | C22:6 ω3 | 0.01 ± 0.01 | 0.01 ± 0.01 |  | 0.454 |
| Plasma |  |  |  |  |  |
|  | C10:0 | ND | ND |  |  |
|  | C12:0 | ND | ND |  |  |
|  | C14:0 | 0.06 ± 0.02 | 0.20 ± 0.24 |  | **<0.01** |
|  | C15:0 | ND | ND |  |  |
|  | C16:0 | 2.26 ± 0.50 | 6.57 ± 0.58 |  | **<0.01** |
|  | C18:0 | 2.67 ± 0.29 | 4.82 ± 0.33 |  | **<0.01** |
|  | C20:0 | ND | ND |  |  |
|  | C21:0 | ND | ND |  |  |
|  | C22:0 | ND | ND |  |  |
|  | C23:0 | ND | ND |  |  |
|  | C24:0 | ND | ND |  |  |
|  | C14:1 ω5 | ND | ND |  |  |
|  | C15:1 ω5 | ND | ND |  |  |
|  | C16:1 ω7 | 0.26 ± 0.06 | 0.55 ± 0.04 |  | **0.01** |
|  | C16:1 ω9 | ND | ND |  |  |
|  | C17:1 ω7 | ND | ND |  |  |
|  | C18:1 ω7 | 0.33 ± 0.07 | 0.85 ± 0.08 |  | **<0.01** |
|  | C18:1 ω9 | 4.30 ± 0.45 | 6.77 ± 0.53 |  | **0.015** |
|  | C20:1 ω9 | ND | ND |  |  |
|  | C22:1 ω9 | ND | ND |  |  |
|  | C24:1 ω9 | ND | ND |  |  |
|  | C18:2 ω6 | 2.79 ± 0.43 | 4.48 ± 0.34 |  | **<0.01** |
|  | C20:2 ω6 | 0.11 ± 0.02 | 01.9 ± 0.03 |  | 0.068 |
|  | C22:2 ω6 | ND | ND |  |  |
|  | C18:3 ω3 | 0.44 ± 0.08 | 0.16 ± 0.04 |  | 0.017 |
|  | C18:3 ω6 | 0.01 ± 0.01 | 0.01 ± 0.01 |  | 0.632 |
|  | C20:3 ω3 | ND | ND |  |  |
|  | C20:3 ω6 | 0.07 ± 0.02 | 0.14 ± 0.02 |  | 0.021 |
|  | C20:4 ω6 | 1.28 ± 0.16 | 1.74 ± 0.14 |  | 0.057 |
|  | C22:4 ω6 | 0.07 ± 0.01 | 0.13 ± 0.02 |  | 0.02 |
|  | C20:5 ω3 | 0.25 ± 0.04 | 0.07 ± 0.03 |  | 0.019 |
|  | C22:5 ω3 | 0.14 ± 0.02 | 0.30 ± 0.04 |  | 0.018 |
|  | C22:5 ω6 | 0.01 ± 0.01 | 0.03 ± 0.05 |  | **<0.01** |
|  | C22:6 ω3 | 0.06 ± 0.02 | 0.31 ± 0.12 |  | 0.068 |

Table S2: Arithmetic means (‘Means’) and standard errors (‘SE’) of proportions of specific fatty acids (‘FA’) among saturated FA, monounsaturated FA or polyunsaturated FA in white adipose tissue (‘WAT’), muscle tissue (‘Muscle’) and blood plasma (‘Plasma’) of bears during the summer active period (‘Summer’) and in winter hibernation (‘Winter’). Differences of least square means (‘Lsmeans’) between seasons and p-values result from linear-mixed effects models (LMM). Sample sizes used in the LMM are presented in Table 1. Significant p-values are highlighted in bold. ‘ND’ refers to non-detectable.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Tissues | Fatty acids | Means ± SE | | Winter-summer differences (%FA) | |
|  |  | **Summer** | **Winter** | **Lsmeans**± **SE** | **P-values** |
| WAT |  |  |  |  |  |
|  | C10:0 | ND | ND |  |  |
|  | C12:0 | ND | ND |  |  |
|  | C14:0 | 1.64 ± 0.80 | 2.80 ± 0.90 | 1.16 ± 0.99 | 0.300 |
|  | C15:0 | ND | ND |  |  |
|  | C16:0 | 59.95 ± 3.55 | 70.82 ± 3.95 | 10.87 ± 4.30 | 0.057 |
|  | C18:0 | 37.67 ± 3.43 | 25.78 ± 3.88 | -11.88 ± 5.75 | 0.063 |
|  | C20:0 | 0.74 ± 0.30 | 0.77 ± 0.33 | 0.03 ± 0.31 | 0.935 |
|  | C21:0 | ND | ND |  |  |
|  | C22:0 | 0.00 ± 0.06 | 0.10 ± 0.07 | 0.10 ± 0.09 | 0.321 |
|  | C23:0 | ND | ND |  |  |
|  | C24:0 | ND | ND |  |  |
|  | C14:1 ω5 | ND | ND |  |  |
|  | C15:1 ω5 | ND | ND |  |  |
|  | C16:1 ω7 | 0.62 ± 0.32 | 0.57 ± 0.34 | -0.05 ± 0.30 | 0.865 |
|  | C16:1 ω9 | 6.05 ± 0.92 | 9.65 ± 1.01 | 3.60 ± 1.05 | 0.020 |
|  | C17:1 ω7 | ND | ND |  |  |
|  | C18:1 ω7 | 5.62 ± 0.41 | 4.29 ± 0.47 | -1.33 ± 0.59 | 0.080 |
|  | C18:1 ω9 | 87.04 ± 1.05 | 84.12 ± 1.15 | -2.92 ± 1.10 | 0.050 |
|  | C20:1 ω9 | 0.67 ± 0.28 | 1.21 ± 0.32 | 0.54 ± 0.41 | 0.250 |
|  | C22:1 ω9 | ND | ND |  |  |
|  | C24:1 ω9 | ND | ND |  |  |
|  | C18:2 ω6 | 50.23 ± 3.43 | 58.03 ± 3.86 | 7.80 ± 4.64 | 0.154 |
|  | C20:2 ω6 | 0.00 ± 1.15 | 4.54 ± 1.35 | 4.54 ± 1.77 | 0.061 |
|  | C22:2 ω6 | ND | ND |  |  |
|  | C18:3 ω3 | 10.49 ± 2.54 | 18.78 ± 2.72 | 8.29 ± 2.08 | 0.023 |
|  | C18:3 ω6 | ND | ND |  |  |
|  | C20:3 ω3 | 0.00 ± 0.00 | 0.20 ± 0.13 | 0.20 ± 0.18 | 0.321 |
|  | C20:3 ω6 | 0.30 ± 0.26 | 1.45 ± 0.30 | 1.15 ± 0.37 | 0.028 |
|  | C20:4 ω6 | 30.56 ± 4.48 | 3.23 ± 5.26 | -27.24 ± 6.45 | **0.015** |
|  | C22:4 ω6 | 1.28 ± 0.85 | 2.39 ± 0.95 | 1.11 ± 1.03 | 0.333 |
|  | C20:5 ω3 | 1.89 ± 0.64 | 2.05 ± 0.66 | 0.17 ± 0.39 | 0.693 |
|  | C22:5 ω3 | 4.33 ± 1.24 | 5.11 ± 1.36 | 0.77 ± 1.84 | 0.684 |
|  | C22:5 ω6 | ND | ND |  |  |
|  | C22:6 ω3 | 3.00 ± 1.07 | 2.20 ± 1.13 | -0.80 ± 0.89 | 0.411 |
| Muscle |  |  |  |  |  |
|  | C10:0 | ND | ND |  |  |
|  | C12:0 | ND | ND |  |  |
|  | C14:0 | ND | ND |  |  |
|  | C15:0 | ND | ND |  |  |
|  | C16:0 | 62.44 ± 2.02 | 71.83 ± 1.78 | 9.39 ± 2.69 | **<0.01** |
|  | C18:0 | 37.55 ± 2.07 | 26.62 ± 1.66 | -10.94 ± 2.54 | 0.041 |
|  | C20:0 | ND | ND |  |  |
|  | C21:0 | ND | ND |  |  |
|  | C22:0 | ND | ND |  |  |
|  | C23:0 | ND | ND |  |  |
|  | C24:0 | 1.84 ± 1.29 | 0.00 ± 1.11 | -1.85 ± 1.60 | 0.273 |
|  | C14:1 ω5 | ND | ND |  |  |
|  | C15:1 ω5 | ND | ND |  |  |
|  | C16:1 ω7 | 2.92 ± 2.87 | 7.55 ± 2.53 | 4.63 ± 3.82 | 0.246 |
|  | C16:1 ω9 | ND | ND |  |  |
|  | C17:1 ω7 | 0.00 ± 0.00 | 0.59 ± 0.59 | 0.59 ± 0.59 | 0.443 |
|  | C18:1 ω7 | 14.15 ± 1.28 | 6.44 ± 1.09 | -7.71 ± 1.79 | **<0.01** |
|  | C18:1 ω9 | 83.25 ± 2.78 | 84.90 ±2.41 | 1.66 ± 3.29 | 0.625 |
|  | C20:1 ω9 | ND | ND |  |  |
|  | C22:1 ω9 | ND | ND |  |  |
|  | C24:1 ω9 | ND | ND |  |  |
|  | C18:2 ω6 | 73.81 ± 7.18 | 71.77 ± 6.19 | -2.04 ± 8.43 | 0.813 |
|  | C20:2 ω6 | ND | ND |  |  |
|  | C22:2 ω6 | ND | ND |  |  |
|  | C18:3 ω3 | -0.11 ± 5.63 | 6.02 ± 4.80 | 6.14 ± 6. 49 | 0.396 |
|  | C18:3 ω6 | ND | ND |  |  |
|  | C20:3 ω3 | ND | ND |  |  |
|  | C20:3 ω6 | ND | ND |  |  |
|  | C20:4 ω6 | 12.22 ± 4.01 | 17.72 ± 3.54 | 5.50 ± 5.35 | 0.322 |
|  | C22:4 ω6 | ND | ND |  |  |
|  | C20:5 ω3 | 0.32 ± 0.23 | 0.06 ± 0.20 | -0.26 ± 0.29 | 0.390 |
|  | C22:5 ω3 | 4.11 ± 1.64 | 2.24 ± 1.46 | -1.87 ± 1.63 | 0.296 |
|  | C22:5 ω6 | ND | ND |  |  |
|  | C22:6 ω3 | 10. 07 ± 2.16 | 2.85 ± 1.83 | -7.86 ± 2.70 | 0.020 |
| Plasma |  |  |  |  |  |
|  | C10:0 | ND | ND |  |  |
|  | C12:0 | ND | ND |  |  |
|  | C14:0 | 1.04 ± 0.18 | 1.69 ± 0.18 | 0.66 ± 0.25 | 0.017 |
|  | C15:0 | ND | ND |  |  |
|  | C16:0 | 42.57 ± 2.05 | 55.74 ± 2.05 | 13.18 ± 3.60 | **<0.01** |
|  | C18:0 | 56.46 ± 2.17 | 42.53 ± 2.17 | -13.93 ± 3.80 | **<0.01** |
|  | C20:0 | ND | ND |  |  |
|  | C21:0 | ND | ND |  |  |
|  | C22:0 | ND | ND |  |  |
|  | C23:0 | ND | ND |  |  |
|  | C24:0 | ND | ND |  |  |
|  | C14:1 ω5 | ND | ND |  |  |
|  | C15:1 ω5 | ND | ND |  |  |
|  | C16:1 ω7 | 4.96 ± 0.55 | 6.83 ± 0.55 | 1.88 ± 0.66 | 0.016 |
|  | C16:1 ω9 | ND | ND |  |  |
|  | C17:1 ω7 | ND | ND |  |  |
|  | C18:1 ω7 | 6.27 ± 0.80 | 10.55 ± 0.80 | 4.28 ± 1.20 | **<0.01** |
|  | C18:1 ω9 | 88.77 ± 1.27 | 82.66 ± 1.27 | -6.11 ± 1.75 | **<0.01** |
|  | C20:1 ω9 | ND | ND |  |  |
|  | C22:1 ω9 | ND | ND |  |  |
|  | C24:1 ω9 | ND | ND |  |  |
|  | C18:2 ω6 | 51.96 ± 2.22 | 59.08 ± 2.22 | 7.13 ± 3.38 | 0.064 |
|  | C20:2 ω6 | 2.20 ± 0.35 | 2.59 ± 0.35 | 0.39 ± 0.50 | 0.441 |
|  | C22:2 ω6 | ND | ND |  |  |
|  | C18:3 ω3 | 8.85 ± 1.21 | 2.26 ± 1.21 | -6.58 ± 1.99 | **<0.01** |
|  | C18:3 ω6 | 0.15 ± 0.07 | 0.18 ± 0.07 | 0.03 ± 0.10 | 0.766 |
|  | C20:3 ω3 | ND | ND |  |  |
|  | C20:3 ω6 | ND | ND |  |  |
|  | C20:4 ω6 | 27.34 ± 2.28 | 23.68 ± 2.28 | -3.65 ± 3.23 | 0.272 |
|  | C22:4 ω6 | 1.51 ± 0.22 | 1.75 ± 0.22 | 0.24 ± 0.30 | 0.435 |
|  | C20:5 ω3 | 4.92 ± 0.67 | 1.05 ± 0.67 | -3.87 ± 0.94 | **0.01** |
|  | C22:5 ω3 | 2.58 ± 0.33 | 3.94 ± 0.33 | 1.35 ± 0.47 | **0.01** |
|  | C22:5 ω6 | 0.10 ± 0.06 | 0.40 ± 0.06 | 0.30 ± 0.08 | **<0.01** |
|  | C22:6 ω3 | 1.24 ± 0.81 | 3.66 ± 0.81 | 2.42 ± 1.08 | 0.050 |