**Hantelys et al, Supplementary File 3**

**A/ Kinetics of FGF1 IRES activity in hypoxia (30 min to 24 h)**

**Normoxia Hypoxia**

** **

**IRES activity**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Hypoxia time (h) | LucF/LucR  Mean | SD | **IRES activity: LucF/LucR \*100** | SD | Mann-Whitney  test  P Value | Significance |
| 0 (n=18) | 0.1726 | 0.0131 | 17.26 | 1.31 |  |  |
| 0.5 (n=3) | 0.1604 | 0.0020 | 16.04 | 0.20 | 0.0444 | \* |
| 1 (n=3) | 0.1474 | 0.0038 | 14.74 | 0.38 | 0.0098 | \*\* |
| 2 (n=3) | 0.1541 | 0.0067 | 15.41 | 0.67 | 0.0158 | \*\* |
| 4 (n=3) | 0.1761 | 0.0118 | 17.61 | 1.18 | 0.7308 |  |
| 6 (n=3) | 0.2049 | 0.0263 | 20.49 | 2.63 | 0.0165 | \* |
| 8 (n=3) | 0.2204 | 0.0167 | 22.04 | 1.67 | 0.0015 | \*\* |
| 16 (n=3) | 0.1515 | 0.0025 | 15.15 | 0.25 | 0.0098 | \*\* |
| 24 (n=3) | 0.1283 | 0.0112 | 12.83 | 1.12 | 0.0015 | \*\* |
|  |  |  |  |  |  | \*= p<0.05 |
|  |  |  |  |  |  | \*\*=p<0.01 |
|  |  |  |  |  |  |

**B/ FGF1 IRES**

**LucF**

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  |  | **Biological replicates** | | | | | | | | | | | | | |
| Time | Condition | 1 | 2 | | 3 | 4 | | 5 | | 6 | | 7 | 8 | 9 | |
| 4h | Normoxia | 77 310 | 91 175 | | 63 445 | 52 728 | | 58 279 | | 56 285 | | 59 485 | 53 665 | 53 223 | |
| Hypoxia | 248 088 | 192 876 | | 303 300 | 138 626 | | 171 997 | | 152 394 | | 110 552 | 157 006 | 149 316 | |
| 8h | Normoxia | 3 721 | 5 041 | | 2 401 | 72 622 | | 72 505 | | 73 578 | | 121 992 | 112 068 | 132 044 | |
| Hypoxia | 10 760 | 12 896 | | 8 624 | 70 529 | | 72 640 | | 33 059 | | 118 486 | 109 078 | 112 198 | |
| 24h | Normoxia | 231 071 | 198 199 | | 263 943 | 208 454 | | 220 996 | | 210 670 | | 226 971 | 199 587 | 262 787 | |
| Hypoxia | 138 279 | 153 534 | | 123 025 | 100 770 | | 113 450 | | 99 785 | | 105 268 | 70 597 | 90 603 | |
| **LucR** | | | |  | | |  | |  | |  | | | |  | |  |  |  |  |  |
|  |  | **Biological replicates** | | | | | | | | | | | | | |
| Time | Condition | 1 | 2 | | 3 | 4 | | 5 | | 6 | | 7 | 8 | 9 | |
| 4h | Normoxia | 604 174 | 507 891 | | 700 457 | 452 118 | | 345 763 | | 472 356 | | 495 500 | 529 252 | 547 526 | |
| Hypoxia | 996 143 | 899 215 | | 1 093 070 | 714 978 | | 915 991 | | 757 182 | | 754 720 | 974 680 | 917 680 | |
| 8h | Normoxia | 12 580 | 13 628 | | 11 532 | 700 579 | | 710 733 | | 712 400 | | 912 286 | 860 603 | 855 579 | |
| Hypoxia | 35 746 | 46 304 | | 25 188 | 631 364 | | 634 140 | | 633 140 | | 826 664 | 827 015 | 822 262 | |
| 24h | Normoxia | 1 010 280 | 1 134 589 | | 885 971 | 1 021 135 | | 1 036 550 | | 1 020 270 | | 956 252 | 896 252 | 789 825 | |
| Hypoxia | 662 414 | 716 741 | | 608 087 | 519 324 | | 514 720 | | 769 988 | | 455 239 | 386 245 | 351 256 | |

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF/LucR** | |  | | | |  | | |  |  |  | |
|  |  | | **Biological replicates** | | | | | | | | | | | | | |
| Time | Condition | | 1 | 2 | 3 | | 4 | 5 | | | | 6 | | 7 | 8 | 9 |
| 4h | Normoxia | | 0.1280 | 0.1795 | 0.0906 | | 0.1166 | 0.1686 | | | | 0.1192 | | 0.1201 | 0.1014 | 0.0972 |
| Hypoxia | | 0.2490 | 0.2145 | 0.2775 | | 0.1939 | 0.1878 | | | | 0.2013 | | 0.1465 | 0.1611 | 0.1627 |
| 8h | Normoxia | | 0.2958 | 0.3699 | 0.2082 | | 0.1037 | 0.1020 | | | | 0.1033 | | 0.1337 | 0.1302 | 0.1543 |
| Hypoxia | | 0.3010 | 0.2785 | 0.3424 | | 0.1117 | 0.1145 | | | | 0.0522 | | 0.1433 | 0.1319 | 0.1365 |
| 24h | Normoxia | | 0.2287 | 0.1747 | 0.2979 | | 0.2041 | 0.2132 | | | | 0.2065 | | 0.2374 | 0.2227 | 0.3327 |
| Hypoxia | | 0.2338 | 0.1478 | 0.1675 | | 0.1974 | 0.2435 | | | | 0.1885 | | 0.2312 | 0.1828 | 0.2579 |

**IRES activity (AU : LucF/LuR x 100)**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Time | Condition | Total mean | SD | H/N ratio | M-W test  P value | Significance |
| 4h | Normoxia | 12.46 | 3.07 |  | 0.0012 | \*\* |
| Hypoxia | 19.94 | 4.27 | **1.60** |
| 8h | Normoxia | 17.79 | 9.57 |  | 0.8633 |  |
| Hypoxia | 17.91 | 10.10 | 1.01 |
| 24h | Normoxia | 23.53 | 4.95 |  | 0.2581 |  |
| Hypoxia | 20.46 | 3.56 | 0.87 |

**C/ FGF2 IRES**

**LucF**

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | | | | | | | | | | | |  |  |  |  |  |  |  |  |  |  |
|  |  | **Biological replicates** | | | | | | | | |
| Time | Condition | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 4h | Normoxia | 89 743 | 98 302 | 91 878 | 104 059 | 102 906 | 102 632 | 80 652 | 78 568 | 70 687 |
| Hypoxia | 109 876 | 111 252 | 98 034 | 109 548 | 111 252 | 120 034 | 85 698 | 90 565 | 84 568 |
| 8h | Normoxia | 146 589 | 149 931 | 129 865 | 84 185 | 90 263 | 75623 | 80 256 | 77 895 | 79 568 |
| Hypoxia | 124 357 | 130 350 | 152 345 | 85 227 | 82 565 | 78 265 | 80 268 | 82 568 | 79 635 |
| 24h | Normoxia | 116 853 | 120 498 | 124 567 | 226 539 | 198 256 | 161 526 | 178 258 | 170 625 | 168 262 |
| Hypoxia | 182 431 | 173 530 | 165 431 | 245 261 | 256 154 | 198 782 | 258 684 | 298 365 | 224 265 |
|  |  |  |  |  |  |  |  |  |  |  |
| **LucR** |  |  |  |  |  |  |  |  |  |  |
|  |  | **Biological replicates** | | | | | | | | |
| Time | Condition | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 4h | Normoxia | 628834 | 603251 | 508251 | 686008 | 676840 | 627525 | 458252 | 398254 | 405258 |
| Hypoxia | 457353 | 502343 | 376543 | 492213 | 456687 | 475340 | 386685 | 356812 | 298568 |
| 8h | Normoxia | 876234 | 1012834 | 786352 | 647158 | 709937 | 655582 | 478584 | 489656 | 505362 |
| Hypoxia | 508675 | 642576 | 756243 | 592213 | 556687 | 604257 | 425542 | 398568 | 405268 |
| 24h | Normoxia | 576834 | 715157 | 496642 | 686695 | 677527 | 628212 | 578568 | 550268 | 562454 |
| Hypoxia | 780284 | 1173705 | 987564 | 1242151 | 1051822 | 1054542 | 978587 | 857258 | 895635 |

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF/LucR** | |  | | | |  | | |  |  |  | |
|  |  | | **Biological replicates** | | | | | | | | | | | | | |
| Time | Condition | | 1 | 2 | 3 | | 4 | 5 | | | | 6 | | 7 | 8 | 9 |
| 4h | Normoxia | | 0.1427 | 0.1630 | 0.1808 | | 0.1517 | 0.1520 | | | | 0.1636 | | 0.1760 | 0.1973 | 0.1744 |
| Hypoxia | | 0.2402 | 0.2215 | 0.2604 | | 0.2226 | 0.2436 | | | | 0.2525 | | 0.2216 | 0.2538 | 0.2832 |
| 8h | Normoxia | | 0.1673 | 0.1480 | 0.1651 | | 0.1301 | 0.1271 | | | | 0.1154 | | 0.1677 | 0.1591 | 0.1574 |
| Hypoxia | | 0.2445 | 0.2029 | 0.2014 | | 0.1439 | 0.1483 | | | | 0.1295 | | 0.1886 | 0.2072 | 0.1965 |
| 24h | Normoxia | | 0.2026 | 0.1685 | 0.2508 | | 0.3299 | 0.2926 | | | | 0.2571 | | 0.3081 | 0.3101 | 0.2992 |
| Hypoxia | | 0.2338 | 0.1478 | 0.1675 | | 0.1974 | 0.2435 | | | | 0.1885 | | 0.2643 | 0.3480 | 0.2504 |

**IRES activity (AU : LucF/LuR x 100)**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Time | Condition | Total mean | SD | H/N ratio | M-W test  P value | Significance |
| 4h | Normoxia | 16.68 | 1.70 |  | <0.0001 | \*\*\*\* |
| Hypoxia | 24.44 | 2.08 | **1.46** |
| 8h | Normoxia | 14.86 | 1 .96 |  | 0.0503 | ns |
| Hypoxia | 18.48 | 3.69 | 1.24 |
| 24h | Normoxia | 26.88 | 5.41 |  | 0.0939 | ns |
| Hypoxia | 22.68 | 6.03 | 0.84 |

**D/ VEGFA IRES a**

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF** |  |  | | | | |  | |  | |  | |  |  |  | |  |  | |
|  |  |  | | | | |  | |  | |  | |  |  |  | |  |  | |
|  |  | **Biological replicates** | | | | | | | | | | | | | | | | |
| Time | Condition | 1 | 2 | 3 | 4 | 5 | | 6 | | 7 | | 8 | | | | 9 | | |
| 4h | Normoxia | 106 987 | 7 470 | 80 384 | 80 568 | 75 356 | | 78 658 | | 68 256 | | 60 658 | | | | 58 698 | | |
| Hypoxia | 198 708 | 10 543 | 112 982 | 187 525 | 175 862 | | 168 568 | | 105 364 | | 121 231 | | | | 135 682 | | |
| 8h | Normoxia | 140 987 | 13 381 | 229 805 | 100 578 | 96 235 | | 97268 | | 230 264 | | 214 235 | | | | 221 658 | | |
| Hypoxia | 291 799 | 15 874 | 217 091 | 186 568 | 202 525 | | 164 570 | | 402 562 | | 380 256 | | | | 351 214 | | |
| 24h | Normoxia | 95 467 | 11 501 | 66 548 | 102 540 | 110 402 | | 98 758 | | 125 214 | | 123 214 | | | | 110 254 | | |
| Hypoxia | 43 998 | 3 278 | 18 093 | 60 568 | 55 845 | | 68 025 | | 70 658 | | 65 252 | | | | 64 154 | | |
|  |  |  |  |  |  |  | |  | |  | |  | | | |  | | |
| **LucR** |  |  |  |  |  |  | |  | |  | |  | | | |  | | |
|  |  |  |  |  |  |  | |  | |  | |  | | | |  | | |
|  |  | **Biological replicates** | | | | | | | | | | | | | | | | |
| Time | Condition | 1 | 2 | 3 | 4 | 5 | | 6 | | 7 | | 8 | | | | 9 | | |
| 4h | Normoxia | 557 863 | 620 150 | 654 367 | 512 362 | 458 685 | | 398 568 | | 369 568 | | 398 658 | | | | 405 235 | | |
| Hypoxia | 1 387 607 | 1 255 594 | 987 656 | 985 365 | 987 878 | | 865 258 | | 875 698 | | 870 258 | | | | 865 234 | | |
| 8h | Normoxia | 1 098 767 | 1 618 781 | 1 579 760 | 758 652 | 684 594 | | 705635 | | 656 265 | | 652 485 | | | | 698 584 | | |
| Hypoxia | 1 465 480 | 1 285 249 | 1 354 320 | 1 025 268 | 985 698 | | 1 014 254 | | 878 258 | | 852 012 | | | | 878 565 | | |
| 24h | Normoxia | 1 246 587 | 1 620 876 | 1 674 890 | 878 985 | 985 635 | | 868 457 | | 758 587 | | 765 625 | | | | 720 258 | | |
| Hypoxia | 1 054 689 | 879 823 | 987 698 | 1 121 245 | 1 002 568 | | 987 685 | | 975 685 | | 985 625 | | | | 945 252 | | |

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF/LucR** | |  | | | |  | | |  |  |  | |
|  |  | | **Biological replicates** | | | | | | | | | | | | | |
| Time | Condition | | 1 | 2 | 3 | | 4 | 5 | | | | 6 | | 7 | 8 | 9 |
| 4h | Normoxia | | 0.1918 | 0.0120 | 0.1228 | | 0.1572 | 0.1643 | | | | 0.1974 | | 0.1847 | 0.1522 | 0.1448 |
| Hypoxia | | 0.1432 | 0.0084 | 0.1144 | | 0.1903 | 0.1780 | | | | 0.1948 | | 0.1203 | 0.1393 | 0.1568 |
| 8h | Normoxia | | 0.1283 | 0.0083 | 0.1455 | | 0.1326 | 0.1406 | | | | 0.1378 | | 0.3509 | 0.3283 | 0.3173 |
| Hypoxia | | 0.1991 | 0.0124 | 0.1603 | | 0.1820 | 0.2055 | | | | 0.1623 | | 0.4584 | 0.4463 | 0.3998 |
| 24h | Normoxia | | 0.0766 | 0.0071 | 0.0397 | | 0.1167 | 0.1120 | | | | 0.1137 | | 0.1651 | 0.1609 | 0.1531 |
| Hypoxia | | 0.0417 | 0.0037 | 0.0183 | | 0.0540 | 0.0557 | | | | 0.0689 | | 0.0724 | 0.0662 | 0.0679 |

**IRES activity (AU : LucF/LuR x 100)**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Time | Condition | Total mean | SD | H/N ratio | M-W test  P value | Significance |
| 4h | Normoxia | 14.75 | 5.61 |  | 0.4363 | ns |
| Hypoxia | 13.84 | 5.66 | 0.94 |
| 8h | Normoxia | 18.77 | 11.64 |  | 0.1359 | ns |
| Hypoxia | 24.73 | 15.24 | **1.32** |
| 24h | Normoxia | 10.50 | 5.47 |  | 0 .0244 | \* |
| Hypoxia | 4.99 | 2.42 | **0.48** |

**E/ VEGFA IRES b**

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF** | | |  | | |  | |  |  | |  | |  | |  | |  | |  | |  | |
|  | | |  | | |  | |  |  | |  | |  | |  | |  | |  | |  | |
|  |  | | | **Biological replicates** | | | | | | | | | | | | | | | | | |
| Time | | Condition | | 1 | 2 | | 3 | | | 4 | | 5 | | 6 | | 7 | | 8 | | 9 | |
| 4h | | Normoxia | | 140 874 | 98 149 | | 100 854 | | | 102 368 | | 112 327 | | 106 258 | | 104 241 | | 124 251 | | 114 212 | |
| Hypoxia | | 100 976 | 91 883 | | 93 743 | | | 86 358 | | 87 652 | | 85 365 | | 80 242 | | 78 658 | | 81 542 | |
| 8h | | Normoxia | | 159 873 | 208 754 | | 170 983 | | | 130 254 | | 112 584 | | 112 547 | | 124 212 | | 110 226 | | 102 265 | |
| Hypoxia | | 378 983 | 513 871 | | 389 750 | | | 298 654 | | 305 265 | | 300 245 | | 310 245 | | 302 142 | | 298 265 | |
| 24h | | Normoxia | | 499 864 | 537 099 | | 556 098 | | | 498 567 | | 465 856 | | 475 554 | | 487 568 | | 452 028 | | 435 982 | |
| Hypoxia | | 350 980 | 439 788 | | 345 230 | | | 345 268 | | 344 568 | | 324 265 | | 285 475 | | 284 658 | | 235 268 | |
|  | |  | |  |  | |  | | |  | |  | |  | |  | |  | |  | |
| **LucR** | |  | |  |  | |  | | |  | |  | |  | |  | |  | |  | |
|  | |  | |  |  | |  | | |  | |  | |  | |  | |  | |  | |
|  |  | | | **Biological replicates** | | | | | | | | | | | | | | | | | |
| Time | | Condition | | 1 | 2 | | 3 | | | 4 | | 5 | | 6 | | 7 | | 8 | | 9 | |
| 4h | | Normoxia | | 1 087 650 | 2 019 783 | | 1 876 580 | | | 986 256 | | 968 586 | | 987 584 | | 876 548 | | 857 625 | | 985 562 | |
| Hypoxia | | 2 098 750 | 2 993 873 | | 3 542 712 | | | 1 987 584 | | 1 876 258 | | 1 785 625 | | 1 685 265 | | 1 457 235 | | 1 471 457 | |
| 8h | | Normoxia | | 2 178 659 | 2 269 092 | | 2 157 890 | | | 1 587 568 | | 1 658 258 | | 1 785 547 | | 1 325 328 | | 1 245 258 | | 1 475 682 | |
| Hypoxia | | 2 765 909 | 3 127 386 | | 2 987 652 | | | 2 258 586 | | 2 457 268 | | 2 358 625 | | 2 425 241 | | 2 574 625 | | 2 145 258 | |
| 24h | | Normoxia | | 3 987 600 | 3 794 801 | | 3 654 780 | | | 3 685 658 | | 3 545 658 | | 3 365 245 | | 3 258 654 | | 3 214 651 | | 3 254 478 | |
| Hypoxia | | 3 265 890 | 3 614 913 | | 2 764 579 | | | 2 998 586 | | 3 058 248 | | 3 124 025 | | 3 021 214 | | 2 875 625 | | 2 874 246 | |

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF/LucR** | |  | | | |  | | |  |  |  | |
|  |  | | **Biological replicates** | | | | | | | | | | | | | |
| Time | Condition | | 1 | 2 | 3 | | 4 | 5 | | | | 6 | | 7 | 8 | 9 |
| 4h | Normoxia | | 0.1295 | 0.0486 | 0.0537 | | 0.1038 | 0.1160 | | | | 0.1076 | | 0.1189 | 0.1449 | 0.1159 |
| Hypoxia | | 0.0481 | 0.0307 | 0.0265 | | 0.0434 | 0.0467 | | | | 0.0478 | | 0.0476 | 0.0540 | 0.0554 |
| 8h | Normoxia | | 0.0734 | 0.0920 | 0.0792 | | 0.0820 | 0.0679 | | | | 0.0630 | | 0.0937 | 0.0885 | 0.0693 |
| Hypoxia | | 0.1370 | 0.1643 | 0.1305 | | 0.1322 | 0.1242 | | | | 0.1273 | | 0.1279 | 0.1174 | 0.1390 |
| 24h | Normoxia | | 0.1254 | 0.1415 | 0.1522 | | 0.1353 | 0.1314 | | | | 0.1413 | | 0.1496 | 0.1406 | 0.1340 |
| Hypoxia | | 0.1075 | 0.1217 | 0.1249 | | 0.1151 | 0.1127 | | | | 0.1038 | | 0.0945 | 0.0990 | 0.0819 |

**IRES activity (AU : LucF/LuR x 100)**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Time | Condition | Total mean | SD | H/N ratio | M-W test  P value | Significance |
| 4h | Normoxia | 10.43 | 3.25 |  | 0.0005 | \*\*\* |
| Hypoxia | 4.45 | 0.98 | **0.43** |
| 8h | Normoxia | 7.88 | 1.11 |  | <0.0001 | \*\*\*\* |
| Hypoxia | 13.33 | 1.33 | **1.69** |
| 24h | Normoxia | 13.90 | 0.85 |  | <0.0001 | \*\*\*\* |
| Hypoxia | 10.68 | 1.37 | **0.77** |

**F/ VEGFC IRES**

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF** | |  |  |  |  |  |  |  |  |  |  |
|  | |  |  |  |  |  |  |  |  |  |  |
|  |  | | **Biological replicates** | | | | | | | | |
| Time | | Condition | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 4h | | Normoxia | 17 645 | 16 237 | 15 672 | 15 245 | 14 582 | 13 258 | 18 750 | 16 985 | 19 754 |
| Hypoxia | 21 673 | 18 377 | 17 834 | 19 875 | 18 625 | 17 485 | 17 258 | 16 238 | 14 587 |
| 8h | | Normoxia | 21 673 | 18 377 | 17 834 | 20 235 | 20 146 | 19 826 | 16 987 | 17 258 | 16 784 |
| Hypoxia | 27 742 | 18 374 | 19 875 | 26 587 | 21 457 | 20 358 | 30 268 | 31 245 | 30 216 |
| 24h | | Normoxia | 23 451 | 21 422 | 22 765 | 21 568 | 20 148 | 18 759 | 19 867 | 20 235 | 28 220 |
| Hypoxia | 30 194 | 22 544 | 26 876 | 28 568 | 27 532 | 26 352 | 19 287 | 24 568 | 22 586 |
|  | |  |  |  |  |  |  |  |  |  |  |
| **LucR** | |  |  |  |  |  |  |  |  |  |  |
|  | |  |  |  |  |  |  |  |  |  |  |
|  |  | | **Biological replicates** | | | | | | | | |
| Time | | Condition | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 4h | | Normoxia | 287 654 | 316 145 | 365 786 | 195 685 | 185 247 | 182 457 | 205 236 | 204 325 | 210 214 |
| Hypoxia | 302 234 | 354 485 | 345 290 | 298 538 | 290 265 | 284 242 | 280 142 | 270 268 | 262 874 |
| 8h | | Normoxia | 425 763 | 455 482 | 480 013 | 390 584 | 382 546 | 367 856 | 356 258 | 321 252 | 384 276 |
| Hypoxia | 403 294 | 341 909 | 375 634 | 390 268 | 352 632 | 342 268 | 410 226 | 401 236 | 410 256 |
| 24h | | Normoxia | 420 632 | 391 071 | 375 100 | 410 214 | 402 158 | 402 387 | 398 268 | 396 216 | 381 568 |
| Hypoxia | 452 093 | 342 612 | 410 973 | 402 358 | 412 586 | 398 574 | 398 265 | 396 246 | 362 142 |

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF/LucR** | |  | | | |  | | |  |  |  | |
|  |  | | **Biological replicates** | | | | | | | | | | | | | |
| Time | Condition | | 1 | 2 | 3 | | 4 | 5 | | | | 6 | | 7 | 8 | 9 |
| 4h | Normoxia | | 0.0613 | 0.0514 | 0.0428 | | 0.0779 | 0.0787 | | | | 0.0727 | | 0.0914 | 0.0831 | 0.0940 |
| Hypoxia | | 0.0717 | 0.0518 | 0.0516 | | 0.0666 | 0.0642 | | | | 0.0615 | | 0.0616 | 0.0601 | 0.0555 |
| 8h | Normoxia | | 0.0509 | 0.0403 | 0.0372 | | 0.0518 | 0.0527 | | | | 0.0539 | | 0.0477 | 0.0537 | 0.0437 |
| Hypoxia | | 0.0688 | 0.0537 | 0.0529 | | 0.0681 | 0.0608 | | | | 0.0595 | | 0.0738 | 0.0779 | 0.0737 |
| 24h | Normoxia | | 0.0558 | 0.0548 | 0.0607 | | 0.0526 | 0.0501 | | | | 0.0466 | | 0.0499 | 0.0511 | 0.0740 |
| Hypoxia | | 0.0668 | 0.0658 | 0.0654 | | 0.0710 | 0.0667 | | | | 0.0661 | | 0.0484 | 0.0620 | 0.0624 |

**IRES activity (AU : LucF/LuR x 100)**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Time | Condition | Total mean | SD | H/N ratio | M-W test  P value | Significance |
| 4h | Normoxia | 7.26 | 1.75 |  | 0.1359 | ns |
| Hypoxia | 6.05 | 0.67 | 0.83 |
| 8h | Normoxia | 4.80 | 0.62 |  | 0.0003 | \*\*\* |
| Hypoxia | 6.55 | 0.91 | **1.36** |
| 24h | Normoxia | 5.50 | 0.82 |  | 0.0315 | \* |
| Hypoxia | 6.38 | 0.64 | **1.16** |

**G/ VEGFD IRES**

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF** | |  |  |  |  |  |  |  |  |  |  |
|  | |  |  |  |  |  |  |  |  |  |  |
|  |  | | **Biological replicates** | | | | | | | | |
| Time | | Condition | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 4h | | Normoxia | 1 123 453 | 1 092 536 | 1 242 342 | 125 682 | 123 122 | 120 268 | 1 252 025 | 985 986 | 1 025 254 |
| Hypoxia | 1 087 648 | 1 129 996 | 1 250 434 | 110 236 | 112 025 | 99 856 | 1 452 638 | 1 258 326 | 1 425 325 |
| 8h | | Normoxia | 899 766 | 837 544 | 657 889 | 89 526 | 75 862 | 78 588 | 1 418 588 | 1 325 682 | 1 312 022 |
| Hypoxia | 1 141 352 | 977 778 | 855 973 | 112 252 | 85 211 | 92 252 | 1 912 212 | 1 858 562 | 1 757 552 |
| 24h | | Normoxia | 1 187 460 | 1 382 536 | 1 289 532 | 147 214 | 132 250 | 145 258 | 1 547 252 | 1 457 258 | 1 325 206 |
| Hypoxia | 521 596 | 706 269 | 506 468 | 50 231 | 48 215 | 47 528 | 485 625 | 465 258 | 475 528 |
|  | |  |  |  |  |  |  |  |  |  |  |
| **LucR** | |  |  |  |  |  |  |  |  |  |  |
|  | |  |  |  |  |  |  |  |  |  |  |
|  |  | | **Biological replicates** | | | | | | | | |
| Time | | Condition | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 4h | | Normoxia | 11 987 690 | 12 069 833 | 12 069 833 | 1 265 226 | 1 125 268 | 1 125 363 | 10 252 682 | 9 253 258 | 11 220 214 |
| Hypoxia | 10 657 845 | 12 059 630 | 11 656 849 | 1 025 236 | 989 265 | 1 125 236 | 11 212 241 | 13 258 233 | 12 258 225 |
| 8h | | Normoxia | 13 799 872 | 14 349 216 | 12 764 891 | 1 325 662 | 1 685 236 | 1 256 364 | 12 452 120 | 11 572 582 | 11 457 582 |
| Hypoxia | 11 678 964 | 10 969 410 | 12 345 654 | 1 152 635 | 1 232 568 | 1 152 332 | 10 214 785 | 9 865 236 | 11 525 852 |
| 24h | | Normoxia | 14 908 768 | 16 101 138 | 15 779 780 | 1 658 235 | 1 751 820 | 1 256 250 | 10 475 685 | 11 452 232 | 10 254 362 |
| Hypoxia | 8 765 789 | 9 428 743 | 9 076 890 | 895 652 | 952 325 | 875 252 | 9 258 352 | 8 562 265 | 8 956 825 |

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF/LucR** | |  | | | |  | | |  |  |  | |
|  |  | | **Biological replicates** | | | | | | | | | | | | | |
| Time | Condition | | 1 | 2 | 3 | | 4 | 5 | | | | 6 | | 7 | 8 | 9 |
| 4h | Normoxia | | 0.0937 | 0.0905 | 0.1029 | | 0.0993 | 0.1094 | | | | 0.1069 | | 0.1221 | 0.1066 | 0.0914 |
| Hypoxia | | 0.1021 | 0.0937 | 0.1073 | | 0.1075 | 0.1132 | | | | 0.0887 | | 0.1296 | 0.0949 | 0.1163 |
| 8h | Normoxia | | 0.0652 | 0.0584 | 0.0515 | | 0.0675 | 0.0450 | | | | 0.0626 | | 0.1139 | 0.1146 | 0.1145 |
| Hypoxia | | 0.0977 | 0.0891 | 0.0693 | | 0.0974 | 0.0691 | | | | 0.0801 | | 0.1872 | 0.1884 | 0.1525 |
| 24h | Normoxia | | 0.0796 | 0.0859 | 0.0817 | | 0.0888 | 0.0755 | | | | 0.1156 | | 0.1477 | 0.1272 | 0.1292 |
| Hypoxia | | 0.0595 | 0.0749 | 0.0558 | | 0.0561 | 0.0506 | | | | 0.0543 | | 0.0525 | 0.0543 | 0.0531 |

**IRES activity (AU : LucF/LuR x 100)**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Time | Condition | Total mean | SD | H/N ratio | M-W test  P value | Significance |
| 4h | Normoxia | 10.25 | 1.01 |  | 0.5043 | ns |
| Hypoxia | 10.59 | 1.28 | **1.03** |
| 8h | Normoxia | 7.70 | 2.88 |  | 0.0503 | ns |
| Hypoxia | 11.45 | 4.83 | **1.49** |
| 24h | Normoxia | 10.35 | 2.67 |  | <0.0001 | \*\*\*\* |
| Hypoxia | 5.68 | 0.72 | **0.55** |

**H/ c-myc IRES**

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF** | |  |  |  |  |  |  |  |  |  |  |
|  | |  |  |  |  |  |  |  |  |  |  |
|  |  | | **Biological replicates** | | | | | | | | |
| Time | | Condition | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 4h | | Normoxia | 2 567 872 | 3 651 734 | 3 454 332 | 356 812 | 375 210 | 351 278 | 360 212 | 410 251 | 420 215 |
| Hypoxia | 2 029 809 | 3 147 437 | 3 256 210 | 275 895 | 265 826 | 245 628 | 310 215 | 325 230 | 312 020 |
| 8h | | Normoxia | 3 546 872 | 3 161 720 | 3 478 284 | 368 258 | 450 268 | 452 025 | 415 231 | 398 652 | 350 652 |
| Hypoxia | 3 508 730 | 2 749 546 | 2 876 921 | 325 120 | 301 245 | 298 547 | 350 655 | 316 522 | 310 222 |
| 24h | | Normoxia | 7 768 312 | 8 684 097 | 6 987 312 | 358 582 | 258 625 | 247 583 | 362 025 | 352 014 | 342 062 |
| Hypoxia | 7 513 771 | 5 782 962 | 7 981 903 | 486 250 | 460 258 | 410 258 | 658 250 | 568 260 | 487 210 |
|  | |  |  |  |  |  |  |  |  |  |  |
| **LucR** | |  |  |  |  |  |  |  |  |  |  |
|  | |  |  |  |  |  |  |  |  |  |  |
|  |  | | **Biological replicates** | | | | | | | | |
| Time | | Condition | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 4h | | Normoxia | 29 784 420 | 27 508 124 | 26 098 324 | 3 025 325 | 2 758 632 | 2 658 985 | 2 879 258 | 2 586 258 | 2 586 258 |
| Hypoxia | 20 988 822 | 19 862 926 | 21 341 228 | 2 025 128 | 2 212 570 | 2 325 201 | 2 145 251 | 2 014 214 | 2 147 542 |
| 8h | | Normoxia | 24 900 832 | 29 630 539 | 30 983 221 | 2 856 985 | 2 140 236 | 2 562 120 | 3 125 368 | 2 582 321 | 2 836 124 |
| Hypoxia | 23 987 042 | 24 750 406 | 25 987 577 | 2 653 213 | 2 012 251 | 2 120 124 | 2 785 624 | 2 658 325 | 2 625 258 |
| 24h | | Normoxia | 36 987 908 | 35 408 692 | 23 788 902 | 3 856 658 | 4 021 522 | 4 010 698 | 4 025 215 | 4 085 368 | 4 125 214 |
| Hypoxia | 18 898 912 | 13 608 479 | 16 988 904 | 4 582 632 | 4 658 230 | 4 215 120 | 4 658 284 | 4 075 250 | 4 875 251 |

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF/LucR** | |  | | | |  | | |  |  |  | |
|  |  | | **Biological replicates** | | | | | | | | | | | | | |
| Time | Condition | | 1 | 2 | 3 | | 4 | 5 | | | | 6 | | 7 | 8 | 9 |
| 4h | Normoxia | | 0.0862 | 0.1328 | 0.1324 | | 0.1179 | 0.1360 | | | | 0.1321 | | 0.1251 | 0.1586 | 0.1625 |
| Hypoxia | | 0.0967 | 0.1585 | 0.1526 | | 0.1362 | 0.1201 | | | | 0.1056 | | 0.1446 | 0.1615 | 0.1453 |
| 8h | Normoxia | | 0.1424 | 0.1067 | 0.1123 | | 0.1289 | 0.2104 | | | | 0.1764 | | 0.1329 | 0.1544 | 0.1236 |
| Hypoxia | | 0.1463 | 0.1111 | 0.1107 | | 0.1225 | 0.1497 | | | | 0.1408 | | 0.1259 | 0.1191 | 0.1182 |
| 24h | Normoxia | | 0.2100 | 0.2453 | 0.2937 | | 0.0930 | 0.0643 | | | | 0.0617 | | 0.0899 | 0.0862 | 0.0829 |
| Hypoxia | | 0.3976 | 0.4250 | 0.4698 | | 0.1061 | 0.0988 | | | | 0.0973 | | 0.1413 | 0.1394 | 0.0999 |

**IRES activity (AU : LucF/LuR x 100)**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Time | Condition | Total mean | SD | H/N ratio | M-W test  P value | Significance |
| 4h | Normoxia | 13.15 | 2.23 |  |  | ns |
| Hypoxia | 13.57 | 2.32 | 1.03 | 0.6048 |
| 8h | Normoxia | 14.31 | 3.30 |  |  | ns |
| Hypoxia | 12.71 | 1.48 | 0.89 | 0.2973 |
| 24h | Normoxia | 13.63 | 8.82 |  |  | ns |
| Hypoxia | 21.95 | 16.04 | **1.61** | 0.0503 |

**I/ EMCV IRES**

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF** | |  |  |  |  |  |  |  |  |  |  |
|  | |  |  |  |  |  |  |  |  |  |  |
|  |  | | **Biological replicates** | | | | | | | | |
| Time | | Condition | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 4h | | Normoxia | 1 698 754 | 1 842 473 | 1 523 420 | 1 852 625 | 1 798 265 | 1 658 985 | 1 985 625 | 1 895 032 | 1 758 320 |
| Hypoxia | 1 768 791 | 1 848 197 | 1 212 320 | 1 658 986 | 1 725 542 | 1 875 258 | 1 254 214 | 1 158 220 | 1 258 210 |
| 8h | | Normoxia | 2 453 998 | 2 217 362 | 2 178 642 | 2 547 240 | 2 580 264 | 1 985 520 | 2 652 124 | 2 758 210 | 2 650 210 |
| Hypoxia | 1 098 422 | 1 388 910 | 1 053 523 | 1 658 250 | 1 752 220 | 1 425 632 | 1 452 025 | 1 252 620 | 1 158 203 |
| 24h | | Normoxia | 2 987 530 | 3 331 188 | 3 097 981 | 2 658 568 | 2 784 240 | 2 645 280 | 2 875 620 | 2 750 215 | 2 458 982 |
| Hypoxia | 3 208 903 | 3 551 010 | 3 254 552 | 2 985 568 | 3 025 106 | 3 685 210 | 3 025 240 | 2 920 213 | 3 857 920 |
|  | |  |  |  |  |  |  |  |  |  |  |
|  | |  |  |  |  |  |  |  |  |  |  |
| **LucR** | |  |  |  |  |  |  |  |  |  |  |
|  | |  |  |  |  |  |  |  |  |  |  |
|  |  | | **Biological replicates** | | | | | | | | |
| Time | | Condition | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 4h | | Normoxia | 1 897 642 | 1 739 558 | 1 676 425 | 1 325 225 | 1 250 265 | 1 325 052 | 2 015 210 | 1 989 570 | 1 997 212 |
| Hypoxia | 1 497 245 | 1 416 380 | 1 123 235 | 998 565 | 865 258 | 1 125 580 | 1 328 620 | 1 285 210 | 1 275 487 |
| 8h | | Normoxia | 2 453 210 | 2 270 622 | 1 987 430 | 2 025 872 | 2 125 672 | 2 110 253 | 2 251 012 | 2 325 026 | 2 452 012 |
| Hypoxia | 1 345 232 | 1 423 859 | 1 125 412 | 1 257 253 | 1 520 252 | 1 427 856 | 1 352 210 | 1 245 253 | 1 124 210 |
| 24h | | Normoxia | 2 234 123 | 2 586 791 | 3 142 423 | 1 987 562 | 2 358 268 | 2 145 214 | 2 454 210 | 2 325 620 | 2 332 210 |
| Hypoxia | 2 076 764 | 1 593 918 | 2 090 845 | 2 125 147 | 2 235 126 | 1 989 987 | 2 452 012 | 2 145 217 | 2 052 982 |

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF/LucR** | |  | | | |  | | |  |  |  | |
|  |  | | **Biological replicates** | | | | | | | | | | | | | |
| Time | Condition | | 1 | 2 | 3 | | 4 | 5 | | | | 6 | | 7 | 8 | 9 |
| 4h | Normoxia | | 0.8952 | 1.0592 | 0.9087 | | 1.3980 | 1.4383 | | | | 1.2520 | | 0.9853 | 0.9525 | 0.8804 |
| Hypoxia | | 1.1814 | 1.3049 | 1.0793 | | 1.6614 | 1.9943 | | | | 1.6660 | | 0.9440 | 0.9012 | 0.9865 |
| 8h | Normoxia | | 1.0003 | 0.9765 | 1.0962 | | 1.2574 | 1.2139 | | | | 0.9409 | | 1.1782 | 1.1863 | 1.0808 |
| Hypoxia | | 0.8165 | 0.9755 | 0.9361 | | 1.3189 | 1.1526 | | | | 0.9984 | | 1.0738 | 1.0059 | 1.0302 |
| 24h | Normoxia | | 1.3372 | 1.2878 | 0.9859 | | 1.3376 | 1.1806 | | | | 1.2331 | | 1.1717 | 1.1826 | 1.0544 |
| Hypoxia | | 1.5451 | 2.2278 | 1.5566 | | 1.4049 | 1.3534 | | | | 1.8519 | | 1.2338 | 1.3613 | 1.8792 |

**IRES activity (AU : LucF/LuR x 100)**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Time | Condition | Total mean | SD | H/N ratio | M-W test  P value | Significance |
| 4h | Normoxia | 108.55 | 22.02 |  | 0.1903 | ns |
| Hypoxia | 130.21 | 38.62 | **1.20** |
| 8h | Normoxia | 110.34 | 11.29 |  | 0.2224 | ns |
| Hypoxia | 103.42 | 14.11 | 0.94 |
| 24h | Normoxia | 119.68 | 11.99 |  | 0.0003 | \*\*\* |
| Hypoxia | 160.16 | 32.22 | **1.34** |

**J/ Control without IRES (hairpin)**

**LucF**

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  |  | | Biological replicates | | | | | | | |  |  |
| Time | Condition | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 8 h | Normoxia | 2082 | 2490 | 1901 | 4203 | 4273 | 3176 | 5188 | 5900 | 6041 |
| Hypoxia | 1937 | 3592 | 2125 | 5664 | 1600 | 4811 | 3270 | 4499 | ND |
| 24 h | Normoxia | 2262 | 1232 | 1692 |  |  |  |  |  |  |
| Hypoxia | 2063 | 3082 | 4574 |  |  |  |  |  |  |

**LucR**

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  |  | Biological replicates | | | | | | | | | |  |  |
| Time | Condition | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 8 h | Normoxia | 91411 | 99067 | 79850 | 155803 | 142912 | 127454 | 163996 | 161011 | 198604 |
| Hypoxia | 98850 | 86360 | 51747 | 100895 | 73120 | 120118 | 69871 | 75614 | ND |
| 24 h | Normoxia | 95815 | 50314 | 72697 |  |  |  |  |  |  |
| Hypoxia | 49263 | 68572 | 104606 |  |  |  |  |  |  |

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **LucF/ LucR** | | |  | |  | |  | |  |  | |  | |  | |  |
|  |  | Biological replicates | | | | | | | | | | | |  |  |
| Time | Condition | 1 | | 2 | 3 | 4 | | 5 | 6 | 7 | 8 | | 9 |
| 8 h | Normoxia | 0.0228 | | 0.0251 | 0.0238 | 0.0270 | | 0.0299 | 0.0249 | 0.0239 | 0.0239 | | 0.0239 |
| Hypoxia | 0.0236 | | 0.0245 | 0.0233 | 0.0561 | | 0.0219 | 0.0400 | 0.0238 | 0.0238 | | 0.0238 |
| 24 h | Normoxia | 0.0196 | | 0.0416 | 0.0411 |  | |  |  |  |  | |  |
| Hypoxia | 0.0419 | | 0.0449 | 0.0437 |  | |  |  |  |  | |  |

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
|  |  | **AU : LucF/LucR \*100** | |  |  |
| Time | Condition | Total mean (n=9) | SD | M-W test  P value | Significance |
| 8h | Normoxia | 2.80 | 0.45 | 0.4129 | ns |
| Hypoxia | 3.88 | 1.47 |