**S1 File**

**Table A Clinical and biochemical data from healthy horses and those with laminitis. (Vessels were cultured in Matrigel for quantification of angiogenic response).**

|  |  |  |
| --- | --- | --- |
|  | **Healthy (Matrigel)****n = 10** | **Laminitis (Matrigel)****n = 6** |
| **Age (years)** | 19.1 ± 1.4 | 21.5 ± 2.3 |
| **Insulin (mIU/L)**  | 4.1 ± 1.4 | 57.1 ± 29.0\* |
| **ACTH (pg/mL)** | 39.6± 19.5 | 156.1± 23.9\* |
| **Cortisol (nmol/L)** | 152.6 ± 12.5 | 161.4 ± 22.8 |

Data are mean ± SEM and the groups were compared using a Student’s t-test or a Mann-Whitney U test (Insulin). \* denotes a significant difference between the groups (P>0.05).

**Table B Clinical and biochemical data from healthy horses and those with laminitis (Vessels were cultured in collagen for next generation sequencing).**

|  |  |  |
| --- | --- | --- |
|  | **Healthy (Collagen)****n=3** | **Laminitis (Collagen)****n=3** |
| **Age (years)** | 18.9 ± 2.4 | 19.8 ± 2.7 |
| **Insulin (mIU/L)**  | 5.8 ± 1.2 | 32.0 ± 12.4 |
| **ACTH (pg/mL)** | 28.2 ± 6.5 | 75.4 ± 19.6 |
| **Cortisol**  | 146.2 ± 21.3 | 157.9 ± 16.3 |

Data are mean ± SEM and the groups were compared using a Student’s t-test or a Mann-Whitney U test (Insulin). \* denotes a significant difference between the groups (P>0.05).

|  |  |  |  |
| --- | --- | --- | --- |
| **Gene symbol, full name**  | **Forward Primer**  | **Reverse Primer** | **UPL**  |
| *Actβ* (Actin, beta)  | accagaggcatacagggaca  | ctaaggccaaccgtgaaaag | 64  |
| *Col4a1* (Collagen, type IV, alpha 1)  | agttggaggaatgggcttg  | ccagggacaccctgtgag  | 80  |
| *Cxcl5*(C-X-C motif chemokine ligand 5)  | cagtgggtttgagaacaccata  | ctggaggctcattgtggac | 25  |
| *Fkbp51* (FK506 binding protein 5)  | ccttcttgctccgagcttt  | tgttcaagaagttcgcagagc | 69  |
| *Mmp9*(Matrix metallopeptidase 9)  | cagaggtaacccacgtcagc  | gggatccaccttctgagactt | 7  |
| *Per1*(Period circadian clock 1)  | acagcagccacggttctc  | ggacccaggagtgcacag | 71  |

**Table C Murine primer sequences for PCR**

**Table D New vessel outgrowths from laminar vessels of healthy horses and those with laminitis cultured in Matrigel or Type 1 Collagen at day 3.**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | **Healthy:****Matrigel (n=10)** | **Healthy:****Collagen (n=3)** | **Laminitis:****Matrigel (n=6)** | **Laminitis:****Collagen (n=3)** |
| **DMEM** | 5.5 ± 2.3 | 7.3 ± 3.3 | 9.0 ± 5.4 | 2.3 ± 6.7 |
| **FBS** | 11.5 ± 4.3 | 10.0 ± 5.2 | 13.5 ± 6.1 | 17.5 ± 4.9 |
| **Cortisol**  | 56.3 ± 15.0 | 25.0 ± 9.5 | 43.0 ± 21.7 | 44.5 ± 17.3 |
| **Cortisol + FBS** | 108.0 ± 54.5 | 44.0 ± 21.3 | 36.6 ± 17.0 | 40.0 ± 10.6 |

Data are mean number of outgrowths ± SEM. Data were analysed by two way ANOVA and Bonferroni post-hoc test. There was no effect of growth matrix or disease group on response to treatment. n = number of horses.

**Table E New vessel outgrowths from laminar vessels of healthy horses and those with laminitis cultured in Matrigel or Type 1 Collagen at day 7.**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | **Healthy:****Matrigel (n=10)** | **Healthy:****Collagen (n=3)** | **Laminitis:****Matrigel (n=6)** | **Laminitis:****Collagen (n=3)** |
| **DMEM** | 17.0 ± 4.9 | 16.2 ± 8.5 | 14.9 ± 6.5 | 13.0 ± 7.3 |
| **FBS** | 28.2 ± 9.2 | 27.5 ± 4.6 | 27.3 ± 6.9 | 30.8 ± 14.3 |
| **Cortisol**  | 188.5 ± 43.6 | 82.5 ± 22.3 | 105.6 ± 38.9 | 73.3 ± 34.7 |
| **Cortisol + FBS** | 249.5 ± 78.5 | 172.5 ± 32.7 | 117.5 ± 45.3 | 130.0 ± 34.8 |

Data are mean number of outgrowths ± SEM. Data were analysed by two way ANOVA and Bonferroni post-hoc test. There was no effect of growth matrix or disease group on response to treatment. n = number of horses.

**Table F New vessel outgrowths from laminar arteries and laminar veins of healthy horses and those with laminitis cultured in Matrigel at day 3.**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | **Healthy:****Artery (n=4)** | **Healthy:****Vein (n=6)** | **Laminitis:****Artery (n=3)** | **Laminitis:****Vein (n=3)** |
| **DMEM** | 21.0 ± 4.9 | 16.2 ± 8.0 | 12.4 ± 3.4 | 19.9 ± 6.3 |
| **FBS** | 18.2 ± 8.7 | 26.3 ± 7.5 | 21.5 ± 8.5 | 30.0 ± 10.6 |
| **Cortisol**  | 48.3 ± 15.9 | 65.2 ± 30.4 | 58.4 ± 19.4 | 61.7 ± 17.9 |
| **Cortisol + FBS** | 99.4 ± 34.5 | 82.5 ± 29.3 | 36.6 ± 18.5 | 72.0 ± 11.6 |

Data are mean number of outgrowths ± SEM. Data were analysed by two-way ANOVA and Bonferroni post-hoc test. There was no effect of vessel type or disease group on response to treatment. (n= number of horses).

**Table G New vessel outgrowths from laminar arteries and laminar veins of healthy horses and those with laminitis cultured in Matrigel at day 7.**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | **Healthy:****Artery (n=4)** | **Healthy:****Vein (n=6)** | **Laminitis:****Artery (n=3)** | **Laminitis:****Vein (n=3)** |
| **DMEM** | 15.0 ± 7.9 | 18.2 ± 7.5 | 15.4 ± 9.2 | 13.0 ± 7.3 |
| **FBS** | 37.2 ± 14.2 | 24.5 ± 7.6 | 18.5 ± 3.5 | 33.0 ± 12.6 |
| **Cortisol**  | 167.5 ± 35.6 | 142.5 ± 52.3 | 110.6 ± 33.9 | 93.3 ± 24.1 |
| **Cortisol + FBS** | 221.5 ± 86.5 | 182.5 ± 72.4 | 207.5 ± 43.6 | 183.0 ± 44.3 |

Data are mean number of outgrowths ± SEM. Data were analysed by two way ANOVA and Bonferroni post-hoc test. There was no effect of vessel type or disease group on response to treatment. n = number of horses. (n= number of horses).