

**Additional file 1a. Genetic testing results for dysferlinopathy cases**

| Age (years) | Sex (M/F) | <i>DYSF</i> variants      |                                | Western blot result | IF <sup>1</sup> staining result |
|-------------|-----------|---------------------------|--------------------------------|---------------------|---------------------------------|
|             |           | Allele 1                  | Allele 2                       |                     |                                 |
| 14          | M         | c.2643+1G>A (splice site) | c.1948delC p.L650Yfs*6         | 0                   | 0                               |
| 15          | M         | deletion of exons 3-4     | deletion of exons 3-4          | 0                   | 0                               |
| 15          | M         | c.2643+1G>A (splice site) | c.865T>C p.S289P               | ++                  | ++                              |
| 17          | M         | c.3444T>A p.Y1148*        | c.4497delT p.F1499Lfs*4        | 0                   | 0                               |
| 20          | M         | c.5979dupA p.E1994Rfs*3   | c.5979dupA p.E1994Rfs*3        | 0                   | 0                               |
| 20          | M         | c.3444T>A p.Y1148*        | c.5713C>T p.R1905*             | 0                   | 0                               |
| 21          | M         | c.5077C>T p.R1693W        | c.5698_5699delAG p.S1900Efs*14 | 0                   | 0                               |
| 21          | M         | c.4253G>A p.G1418D        | None identified                | 0                   | 0                               |
| 22          | F         | c.5429G>A p.R1810K        | c.5429G>A p.R1810K             | 0                   | 0                               |
| 22          | M         | c.2779del p.A927Lfs*21    | c.3478C>T p.Q1160*             | 0                   | 0                               |
| 23          | M         | c.5105T>C p.L1702P        | c.5105T>C p.L1702P             | ND                  | 0                               |
| 23          | M         | c.855+1delG (splice site) | c.3036G>A p.W1012*             | 0                   | 0                               |
| 25          | F         | c.5022delT p.F1674Lfs*48  | c.5022delT p.F1674Lfs*48       | +                   | 0                               |
| 26          | F         | c.2643+1G>A (splice site) | c.2643+1G>A (splice site)      | ND                  | 0                               |
| 27          | M         | c.755C>T p.T252M          | c.5444G>T p.C1815F             | 0                   | 0                               |

|    |   |                                  |                           |    |   |
|----|---|----------------------------------|---------------------------|----|---|
| 27 | M | c.2372C>G p.P791R                | c.2372C>G p.P791R         | 0  | 0 |
| 28 | M | c.895G>T p.G299W                 | c.895G>T p.G299W          | ND | 0 |
| 30 | M | c.3167G>A p.R1058Q               | c.3167G>A p.R1058Q        | 0  | 0 |
| 31 | M | c.755C>T p.T252M                 | c.4685dupT p.M1562Ifs*39  | ND | 0 |
| 32 | F | c.4081T>C p.C1361R               | deletion of exons 25-29   | 0  | 0 |
| 32 | F | c.2643+1G>A (splice site)        | c.2643+1G>A (splice site) | 0  | 0 |
| 33 | M | c.1368C>A p.C456*                | c.1368C>A p.C456*         | 0  | 0 |
| 40 | M | c.2643+1G>A (splice site)        | c.4577A>C p.K1526T        | ND | 0 |
| 42 | M | c.3892A>G p.I1298V               | c.1878G>A p.M626I         | ND | 0 |
| 45 | F | c.997_998delGC p.A333Qfs*25      | c.1053+1G>A (splice site) | ND | 0 |
| 46 | M | c.1834C>T p.Q612*                | c.4833A>G p.S1611S        | 0  | 0 |
| 48 | M | c.855+1delG (splice site)        | c.3036G>A p.W1012*        | ND | 0 |
| 49 | M | c.254_256delAGG p.L85_V86delinsT | c.4795T>C p.C1599R        | 0  | 0 |
| 53 | M | c.937+4A>T (splice site)         | c.5979dupA p.E1994Rfs*3   | 0  | 0 |
| 54 | M | c.2643+1G>A (splice site)        | c.4577A>C p.K1526T        | 0  | 0 |
| 58 | F | c.1663C>T p.R555W                | c.1663C>T p.R555W         | 0  | 0 |
| 68 | F | c.757C>T p.R253W                 | c.865T>C p.S289P          | +  | 0 |

1 = immunofluorescence

|                                                 |
|-------------------------------------------------|
| Western Blot and IF <sup>1</sup> Interpretation |
| 0 = absent or nearly absent                     |
| + = reduced by ~75%                             |
| ++ = reduced by ~50%                            |
| ND = not done                                   |

**Additional file 1b. Genetic testing results for Becker muscular dystrophy cases**

| Age (years) | Sex (M/F) | DMD variants                        | IF <sup>1</sup> staining result                                                            |
|-------------|-----------|-------------------------------------|--------------------------------------------------------------------------------------------|
| 4           | M         | c.2380G>A<br>p.E794K                | Reduced to absent staining with all antibodies                                             |
| 4           | M         | deletion exons 17-44                | Reduced to absent staining with all antibodies                                             |
| 4           | M         | c.2330T>C<br>p.L777P                | Reduced with all antibodies; absent staining with actin binding domain, exons 7/8 antibody |
| 5           | M         | deletion exons 19-28                | Reduced to absent staining with rod domain antibody                                        |
| 6           | M         | deletion exons 45-57                | Reduced to absent staining with all antibodies                                             |
| 8           | M         | c.7189C>T<br>p.Q2397*               | Predicts in-frame deletion involving exon 50                                               |
| 8           | M         | c.6290+3076 A>G<br>p.Thr3055Serfs*1 | Reduced to absent staining with all antibodies                                             |
| 9           | M         | IVS48 splice site mutation          | Reduced with all antibodies; absent staining with actin binding domain, exons 7/8 antibody |
| 10          | M         | duplication exons 3-30              | Reduced staining with all antibodies                                                       |
| 10          | M         | duplication exons 5-9               | Reduced with all antibodies; absent staining with actin binding domain, exons 7/8 antibody |
| 11          | M         | c.961-5831C>T<br>(splice site)      | Reduced with all antibodies; absent staining with actin binding domain, exons 7/8 antibody |
| 11          | M         | c.76A>C<br>p.N26H                   | Reduced with all antibodies; absent staining with actin binding domain, exons 7/8 antibody |
| 12          | M         | c.6438+47818G>T<br>p.Lys2146Valfs*1 | Reduced staining with actin binding domain, exons 7/8 antibody                             |
| 14          | M         | deletion exons 45-51                | Predicts in-frame deletion involving exons 45-50                                           |

|    |   |                                     |                                                                                                     |
|----|---|-------------------------------------|-----------------------------------------------------------------------------------------------------|
| 14 | M | stop exon 41                        | Reduced staining with all antibodies; absent staining with actin binding domain, exons 7/8 antibody |
| 14 | M | duplication exons 2-25              | Reduced to absent staining with all antibodies                                                      |
| 19 | M | c.2949+909 C>T<br>p.Ala984Thrfs*33  | Reduced staining with carboxy terminus and with actin binding domain, exons 7/8 antibodies          |
| 19 | M | deletion exons 46-52                | Reduced to absent staining with all antibodies                                                      |
| 21 | M | deletion exons 45-48                | Predicts in-frame deletion involving exons 45-49                                                    |
| 22 | M | c.239C>A<br>p.A80E                  | Reduced staining with rod domain antibody                                                           |
| 23 | M | c.1724T>C<br>p.L575P                | Reduced staining with actin binding domain, exons 7/8 antibody                                      |
| 25 | M | deletion exons 13-44                | Predicts a large, in-frame deletion of at least exons 20-43                                         |
| 28 | M | c.10753C>T<br>p.Q3585*              | Absent staining with carboxy terminus antibody                                                      |
| 31 | M | c.9G>A<br>p.W3*                     | Reduced staining with all antibodies; absent staining with actin binding domain, exons 7/8 antibody |
| 31 | M | deletion exons 45-48                | Predicts in-frame deletion involving exons 45-46                                                    |
| 38 | M | deletion exons 45-53                | Predicts in-frame deletion involving exons 45-50                                                    |
| 42 | M | None identified                     | Predicts in-frame deletion involving exon 50                                                        |
| 42 | M | c.3780_3786+3del10<br>p.L1261Kfs*20 | Reduced staining with carboxy terminus antibody                                                     |
| 51 | M | None identified                     | Reduced staining with all antibodies                                                                |
| 84 | M | duplication exons 45-48             | Reduced to absent staining with all antibodies                                                      |

1 = immunofluorescence

**Additional file 1c. Genetic testing results for calpainopathy cases**

| Age (years) | Sex (M/F) | CAPN3 variants                              |                                             | Western blot result                                                      |
|-------------|-----------|---------------------------------------------|---------------------------------------------|--------------------------------------------------------------------------|
|             |           | Allele 1                                    | Allele 2                                    |                                                                          |
| 5           | F         | deletion of entire gene (plus part of GANC) | deletion of entire gene (plus part of GANC) | 0                                                                        |
| 9           | M         | c.2462C>T<br>p.A821V                        | None identified                             | 0                                                                        |
| 13          | F         | c.1355-1G>C<br>(splice site)                | c.2115+5G>C<br>(splice site)                | 0                                                                        |
| 13          | F         | c.2306G>A<br>p.R769Q                        | c.2306G>A<br>p.R769Q                        | 0                                                                        |
| 13          | F         | c.2092C>T<br>p.R698C                        | c.2243G>A<br>p.R748Q                        | +                                                                        |
| 15          | M         | c.1981delA<br>p.I661*                       | c.2440-1G>C<br>(splice site)                | 0                                                                        |
| 15          | M         | c.717delT<br>p.Phe239Leufs*14               | c.759_761delGAA<br>p.Lys254del              | 0                                                                        |
| 15          | M         | c.643_663del<br>p.S215_G221del              | c.1746-20C>G<br>(splice site)               | 0                                                                        |
| 17          | M         | c.83_delinsCTT<br>p.D295L_fs*57             | c.1371C>A<br>p.N457K                        | +                                                                        |
| 19          | F         | c.264T>G<br>p.F88L                          | c.2362_2363delinsTCATCT<br>p.R788Sfs*14     | 0                                                                        |
| 22          | M         | c.759_761delGAA<br>p.K254del                | c.1355-1G>C<br>(splice site)                | +                                                                        |
| 24          | M         | c.1327T>C<br>p.S443P                        | c.1505T>C<br>p.1502T                        | +                                                                        |
| 25          | M         | c.481G>A<br>p.G161R                         | c.1746-20C>G<br>(splice site)               | 0                                                                        |
| 26          | F         | c.550delA<br>p.T184Rfs*36                   | c.550delA<br>p.T184Rfs*36                   | 0                                                                        |
| 28          | F         | c.1468C>T<br>p.R490W                        | c.1063C>T<br>p.R355W                        | 94kd calpain-3<br>nearly normal;<br>almost no<br>degradation<br>products |

|    |   |                                             |                               |                                                                          |
|----|---|---------------------------------------------|-------------------------------|--------------------------------------------------------------------------|
| 29 | F | c.2362_2363delinsTCATCT<br>p.R788Sfs*14     | c.533T>C<br>p.I178T           | 0                                                                        |
| 31 | F | c.1063C>T<br>p.R355W                        | c.181T>C<br>p.F61L            | +                                                                        |
| 32 | F | c.245C>T<br>p.P82L                          | None identified               | 0                                                                        |
| 44 | F | c. 245C>T<br>p.P82L                         | c.1468C>T<br>p.R490W          | 94kd calpain-3<br>++; almost no<br>degradation<br>products               |
| 46 | M | c.2362_2363delinsTCATCT<br>p.R788Sfs*14     | None identified               | +                                                                        |
| 48 | F | c.550delA<br>p.T184Rfs*36                   | None identified               | 0                                                                        |
| 53 | M | dominant het c.643_663del<br>p.S215_G221del | N/A                           | +                                                                        |
| 54 | M | dominant het c.643_663del<br>p.S215_G221del | N/A                           | 0                                                                        |
| 56 | M | c.1333G>A<br>p.G445R                        | None identified               | +                                                                        |
| 62 | F | dominant het c.643_663del<br>p.S215_G221del | N/A                           | +                                                                        |
| 63 | M | None identified                             | None identified               | 0                                                                        |
| 64 | F | c.1435A>G<br>p.S479G                        | None identified               | +                                                                        |
| 66 | M | c.1194-9A>G<br>(splice site)                | None identified               | 94kd calpain-3<br>nearly normal;<br>almost no<br>degradation<br>products |
| 68 | M | c.500T>C<br>p.F167S                         | c.1746-20C>G<br>(splice site) | 0                                                                        |
| 71 | M | c.1309C>T<br>p.R437C                        | deletion of exons 17-20       | 0                                                                        |

| Western Blot Interpretation |
|-----------------------------|
| 0 = absent or nearly absent |
| += reduced by ~75%          |

++ = reduced by ~50%