Expanded View Figures



Figure EV1. Images and muscle histology of Foetuses 4 and 5.

Images and muscle histology of Foetus 4 (22 gw, A-F) and Foetus 5 (13,2 gw, G-K). (A-C) Face and profile pictures showing cystic hygroma, facial dysmorphisms with hypertelorism, high and large forehead and low-set ears. (D) Right arm showing elbow ankylosis. (E) Lower limbs showing bilateral knee ankylosis. (F) Autopsy showing pulmonary hypoplasia and distended bladder. (G, H) Face and profile pictures showing contractures of four limbs with arthrogryposis, multiple pterygia (colli, elbows, knees) and generalised muscular atrophy. (I-K) Histological analysis showing rare muscular fibres of irregular in size, with an excess of conjunctive tissue. Both foetuses present intra-uterine growth retardation and hydrops fetalis.



Figure EV2. Spurious 3' adenylation of 16S rRNA.

(A) Representation of reads beyond the encoded nucleotides for 16S rRNA for Patient 2 and Patient 3 fibroblasts, fibroblast control, HEK PDE12 KO cells and control HEK cells ascertained by MPAT-Seq. (n = 1). (B) Representation of reads beyond the encoded nucleotides for 16S rRNA for RNA extracted from liver of Foetus 4 liver and agematched control liver ascertained by MPAT-Seq. (n = 1). Source data are available online for this figure.



Figure EV3. Mitochondrial respiration in patient fibroblasts.

(A) Mitochondrial oxygen consumption rate (OCR) as measured by a flux analyser on control fibroblasts (grey) and fibroblasts of Patient 2 (red) and Patient 3 (pink). (B) Basal respiration was measured before the addition of Oligomycin. (C) Oligomycin was added to determine ATP production. (D) FCCP was added to determine the maximal respiration and spare respiratory capacity (*p = 0.0263). (E) Extracellular acidification rate. (F) Basal glycolysis. Error bars in (A-F) represent standard error of mean. Each measurement was conducted in at least three replicates (*p = 0.0427, **p = 0.0055). The one-tail t-test was used to calculate the *p* values. Dots show individual measurements (n = 6, technical replicates).



Figure EV4. Gene expression analysis in patient fibroblasts.

(A) System-level analysis of Affymetrix Clariom D transcriptome arrays comparing the RNA levels in Patients 2 and 3 with unaffected controls. Statistical analysis was done using the ANOVA test and FDR-corrected values. The multiple-testing correction is based on the approach of Benjamini, Hochberg, and Yekutieli. (B) Volcano plot showing the differences in gene expression between RNA from Patients 2 and 3 compared with unaffected controls. Red dots represent genes that localise inside the mitochondria, according to MitoCarta, with the most significantly altered genes labelled. Statistical analysis as described for EV4A. (n = 1 biological replicate).