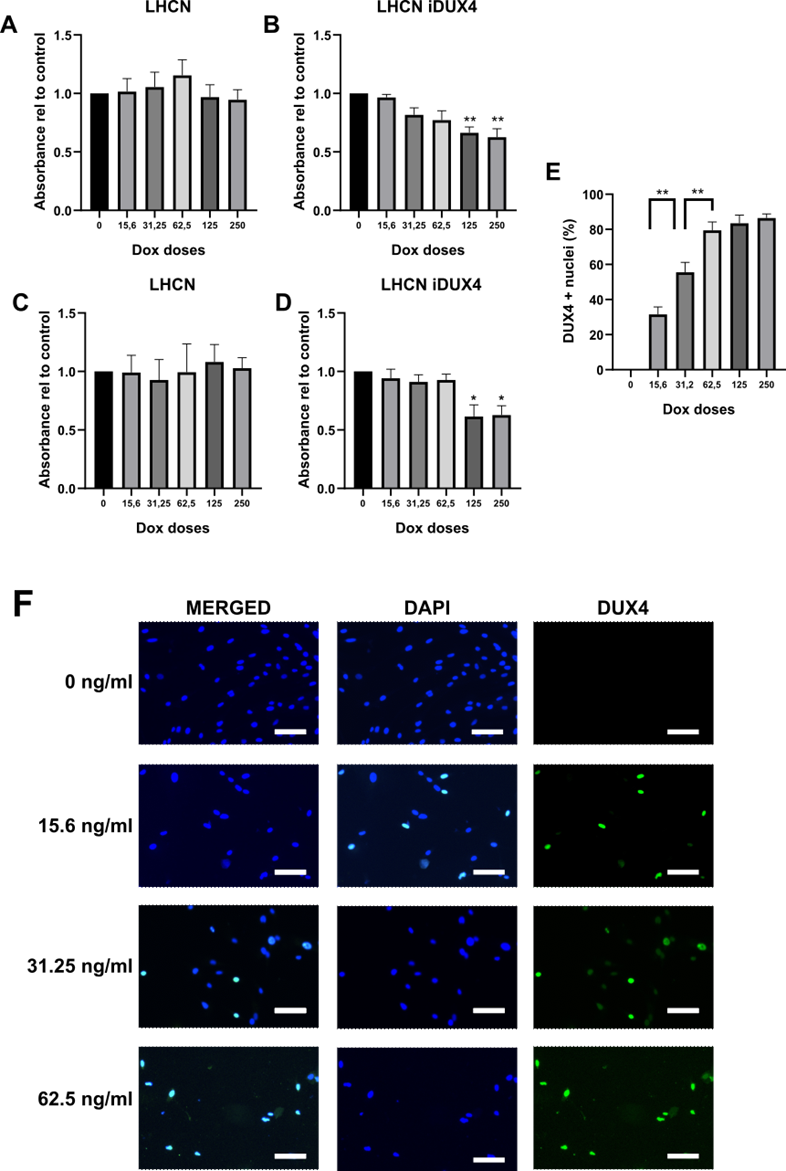
**Supplementary Data**

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**Figure S1. Effect of DUX4 induction on LHCN-M2 and LHCN-M2 iDUX4 cell viability**. **A-D**. MTT (A-B) andCCK8 (C-D) tests were performed 24h after induction of DUX4 expression with increasing doses of doxycycline (DOX, ng/ml). Mean ± SEM, \*p<0.05, \*\*p<0.01, One-way ANOVA with Holm Sidak post hoc test *vs* control (DOX: 0 ng/ml). **E**. Quantification of DUX4 positive (DUX4+) nuclei normalized to the total number of nuclei (DAPI) 24h after induction of DUX4 expression with increasing doses of DOX. Mean ± SEM, \*p<0.05, \*\*p<0.01, One-way ANOVA with Holm Sidak post hoc test *vs* control (DOX: 0 ng/ml). **F.** Representative field showing DUX4+ nuclei detected by immunofluorescence (green) as described in Figure 1. DAPI was used to stain nuclei (blue). Scale = 100µm. Experiments were performed on 3 independent cultures, each in triplicate.

Une image contenant capture d’écran, Caractère coloré, ligne, fenêtre

Description générée automatiquement

**Figure S2. No effect of DUX4 expression on the *Hif1α* pathway in the DUX4 IMEP murine model with a low dose of DUX4 expression.** **A**. Experiment time courses. **B**. Representative sections of TA electroporated with 5 µg of *pCIneo* (left column)or *pCIneo-DUX4* (right column) plasmids at 7-, 14- and 21-days post-injection. èFibrosis, èCentral nuclei, èAtrophic fibers. Scale = 100µm for 20-X and 50 µm for 40-X magnification. **C**. Dose–response of plasmid amount *vs* muscle lesion area in mouse TA, 1-week post-IMEP procedure. The lesion area percentage was evaluated on total cryosection of medial and proximal part of TA electroporated with different doses of a DUX4 expression plasmid (*pCIneo-DUX4*). The results obtained from saline solution injected groups and *pCIneo* group were pooled into a single control group, as no statistical difference could be highlighted between groups at anytime point. \* p< 0,05, \*\* p< 0,01, \*\*\*p<0,001, Kruskal Wallis followed by Dunn's post hoc test. Control group n=10, 0,5 µg and 1µg n=2, 5µg n=6, 10 µg n=7, 20µg n=8, 40µg n=4. **D**. Effect of DUX4 expression on *Wfdc3* *mRNA* level in the IMEP model. RT-qPCR quantifications were normalized to *Rplp0*. \* p< 0,05, \*\* p< 0,01; Kruskal Wallis followed by a Dunn’s post-hoc test. For 1- and 3-day group: *pCIneo-DUX4* n=4, *pCIneo* n=6, saline n=2. For 7- and 14-day groups: *pCIneo-DUX4* n=3, *pCIneo* n=3, saline n=4. For the 21-day group: *pCIneo-DUX4* n=3, *pCIneo* n=4, saline n=4. The results obtained from saline solution injected groups and *pCIneo* group were pooled into a single control group, as no statistical difference could be highlighted between groups at any time point. **E**. Effect of DUX4 induction on *Hif1α* *mRNA* level in the IMEP model. RT-qPCR normalized to *Rplp0* gene. \*p< 0,05, \*\*p< 0,01; Kruskal Wallis followed by a Dunn’s post-hoc test. For 1- and 3-day group: *pCIneo-DUX4* n=4, *pCIneo* n=6. For 7- and 14-days group: *pCIneo-DUX4* n=3, *pCIneo* n=4. For the 21-day group: *pCIneo-DUX4* n=3, *pCIneo* n=4. Results obtained from saline solution injected groups from all time points, were pooled together, as no statistical difference could be highlighted between groups at any time point for all tested genes, saline group n=14.

Une image contenant horloge

Description générée automatiquement

**Figure S3.** **Efficiency of siRNAs directed against *Hif1α* mRNA (*siHIF*): dose-response analysis.** The TA muscle was electroporated with either saline solution, *siCTL* or *siHIF*. *The Hif1α* *mRNA* level was quantified by RT-qPCR and normalized to *Rplp0*. \*p<0.05, \*\*\*p<0.001, Kruskal Wallis followed by a Dunn’s post-hoc test. Saline : n=10, 0.005 µg, 0.01 µg, 0.05 µg, 0.5 µg and 1 µg : n=4, 2µg : n=10.