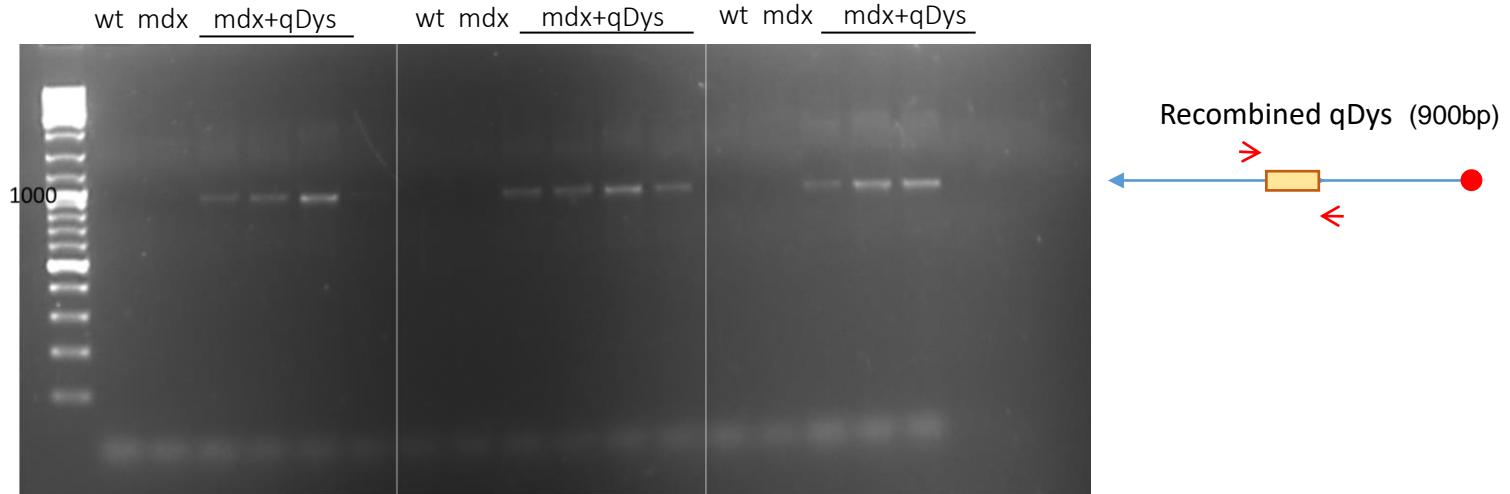


# Figure S1

A



B

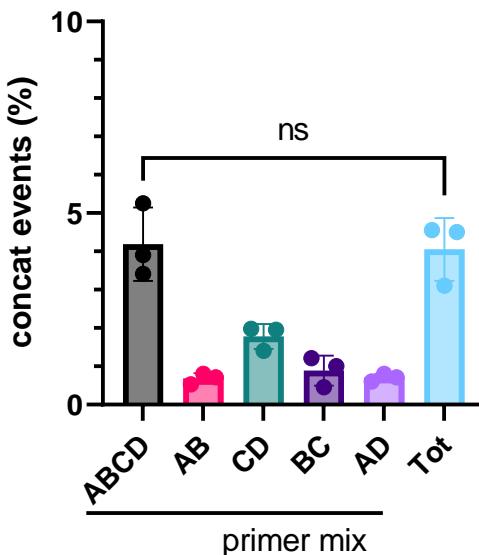
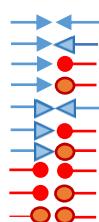
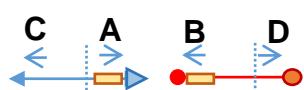


**Figure S1.Verification of identity and specificity of the ddPCR amplified recombinant product.** A) Smal-digested ddPCR amplified product using primers indicated. The 900 bp amplicon was recovered from droplet reaction and loaded onto electrophoretic gel to check the size. B) Sequencing of the amplicon purified from gel. The box shows identity % with the recombinant expected sequence.

# Figure S2



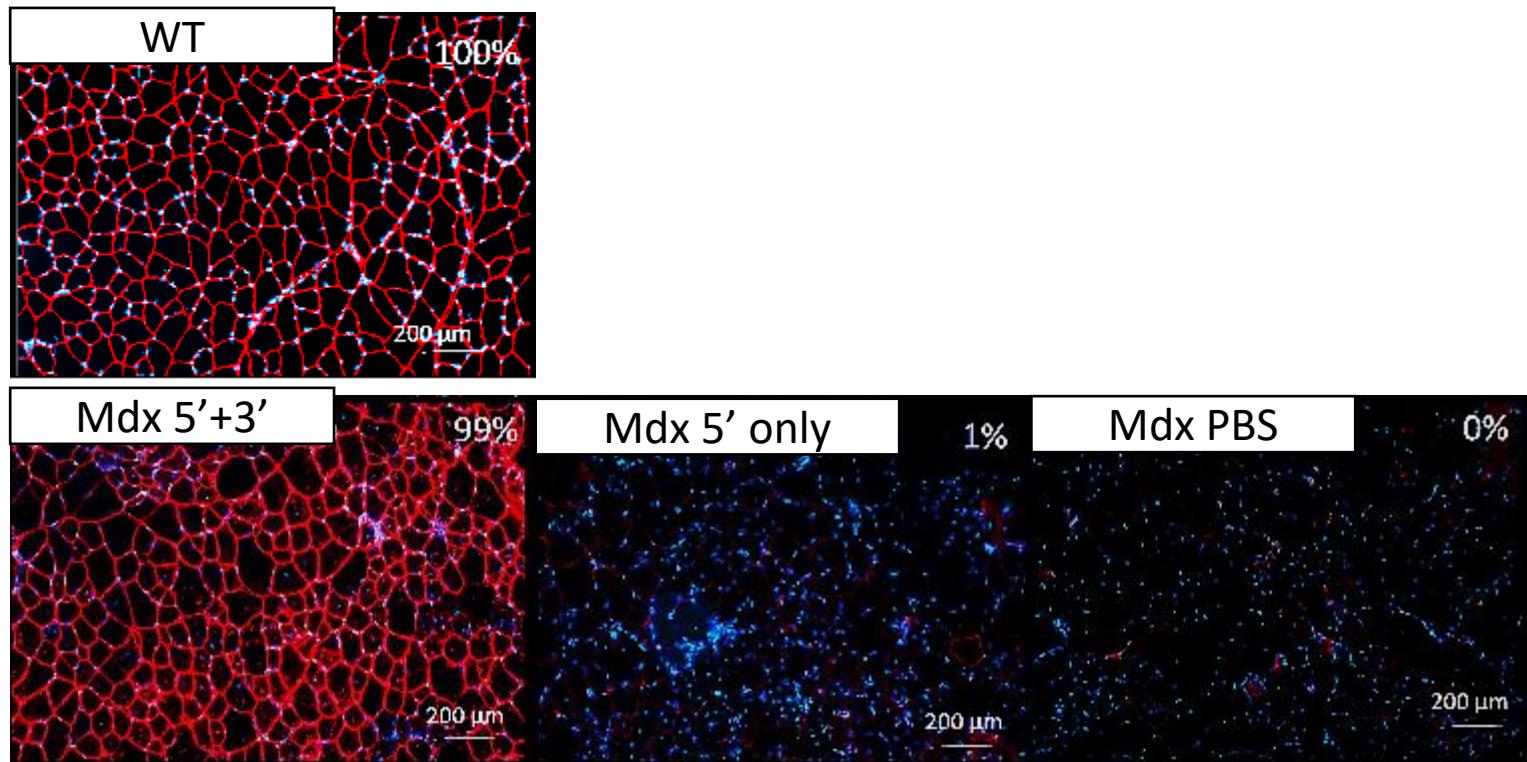
Dual 5'      Dual 3'



**Figure S2. Primer mix optimization for the detection of all concatemerization events.**

Mixing all the primers in the same reaction (ABCD) gives the same result than adding up the concatemerization events (Tot) derived from the single reactions containing single combination of primers (AB, CD, BC, AD). Therefore, the mix of all primers can provide an accurate results in one reaction.

**Figure S3**



**Figure S3. Lack of Dystrophin expression in single vector-injected *mdx* mice.** Representative images of sections of tibialis anterior of DBA2 WT and *mdx* mice after systemic injection of 5' vector alone or in combination with 3'. Indicated in each picture are % of dystrophin positive fibers.

**Table S1.** Primers used in ddPCR multiplex analysis.

Target	Fw primer	Rev primer	Probe
5'	CAGGAGGCCAGGACAATTATTAC	CCATGCTACTTATCTACGTAAAGC	<b>2X FAM</b> TGAGAGGATCCAGAACCACTGGGA
3'	AACCCGCCATGCTACTTATC	TGGAACCTCCTCCACTTCT	<b>VIC</b> AAGATGTACAAGGACAGGCAGGGC
REC	GCCCTGCAGTCTGCTAC	TCCTTCAGCATCTCATTCA	<b>VIC</b> TGAGAGGATCCAGAACCACTGGGA
TTN	TTCAGTCATGCTGCTAGCGC	AAAACGAGCAGTGACGTGAGC	<b>FAM</b> TGCACGGAAGCGTCTCGTCTCAGTC

Mix1 (VCN single vectors)= 5'+3'+TTN

Mix2 (VCN reconstituted vector)= REC+TTN

**Table S2.** Primers used for concatemerization analysis.

<b>A</b>	CAGGATTGAGAGGATCCAGAAC
<b>B</b>	GTTTGACAGGTCA GTGGAGAA
<b>C</b>	CAGGTGTTGGCGCTCTAA
<b>D</b>	TGAGTTGGACAAACCACAAAC