A mutation-independent approach via transcriptional upregulation of a disease modifier gene rescues muscular dystrophy *in vivo*

Dwi U. Kemaladewi^{1,*}, Prabhpreet S. Bassi^{1,2,*}, Steven Erwood^{1,2}, Dhekra Al-Basha^{3,4}, Kinga I. Gawlik⁵, Kyle Lindsay¹, Ella Hyatt¹, Rebekah Kember¹, Kara M. Place¹, Ryan M. Marks¹, Madeleine Durbeej⁵, Steven A. Prescott^{3,4,6}, Evgueni A. Ivakine^{1,**}, Ronald D. Cohn^{1,2,7,**}

¹Program in Genetics and Genome Biology, the Hospital for Sick Children Research Institute, Toronto, Canada

²Department of Molecular Genetics, University of Toronto, Canada

³Program in Neuroscience and Mental Health, the Hospital for Sick Children Research Institute, Toronto, Canada

⁴Department of Physiology, University of Toronto, Canada

⁵Unit of Muscle Biology, Department of Experimental Medical Science, Lund University, Lund, Sweden

⁶Institute of Biomaterials and Biomedical Engineering, University of Toronto, Canada

⁷Department of Pediatrics, the Hospital for Sick Children, Toronto, Canada

*These authors contributed equally to this work

**These authors contributed equally to this work

Introductory paragraph

Identification of genetic modifiers has provided critically important insights into the pathogenesis and heterogeneity of disease phenotypes in individuals affected by neuromuscular disorders (NMDs). Targeting modifier genes to improve disease phenotypes could be especially beneficial in cases where the causative genes are large, structurally complex and the mutations are heterogeneous. Here, we report a mutation-independent strategy to upregulate expression of a compensatory disease-modifying gene in Congenital Muscular Dystrophy type 1A (MDC1A) using a CRISPR/dCas9-based transcriptional activation system.

MDC1A is caused by nonfunctional Laminin $\alpha 2$, which compromises muscle fibers stability and axon myelination in peripheral nerves¹. Transgenic overexpression of *Lama1*, encoding a structurally similar protein Laminin $\alpha 1$, ameliorates muscle wasting and paralysis in the MDC1A mouse models, demonstrating its important role as a disease modifier². Yet, upregulation of *Lama1* as a postnatal gene therapy is hampered by its large size, which exceeds the current genome packaging capacity of clinically relevant delivery vehicles such as adeno-associated viral vectors (AAVs).

In this study, we use the CRISPR/dCas9-based transcriptional activation system to upregulate *Lama1*. This system is comprised of catalytically inactive *S. aureus* Cas9 (dCas9) fused to VP64 transactivation domains and sgRNAs targeting the *Lama1* promoter. We first demonstrate robust upregulation of *Lama1* in myoblasts, and following AAV9-mediated intramuscular delivery, in skeletal muscles of *dy^{2j}/dy^{2j}* mouse model of MDC1A.

We therefore assessed whether systemic upregulation of *Lama1* would yield therapeutic benefits in *dy^{2j}/dy^{2j}* mice. When the intervention was started early in pre-symptomatic *dy^{2j}/dy^{2j}* mice, *Lama1* upregulation prevented muscle fibrosis and hindlimb paralysis. An important question for future therapeutic approaches for a variety of disorders relates to the therapeutic window and phenotypic reversibility. This is particularly true for muscular dystrophies as it has long been hypothesized that fibrotic changes in skeletal muscle represent an irreversible disease state that would impair any therapeutic intervention at advanced stages of the disease. Here, we demonstrate that dystrophic features and disease progression were significantly improved and reversed when the treatment was initiated in symptomatic 3-week old *dy^{2j}/dy^{2j}* mice with already-apparent hind limb paralysis and significant muscle fibrosis.

Collectively, our data demonstrate the feasibility and therapeutic benefit of CRISPR/dCas9-mediated modulation of a disease modifier gene, which opens up an entirely new and mutation-independent treatment approach for all MDC1A patients. Moreover, this treatment strategy provides evidence that muscle fibrosis can be reversible to some degree, thus extending the therapeutic window for this disorder. Our data provide a proof-of-concept strategy that can be applied to a variety of disease modifier genes and a powerful therapeutic approach for various inherited and acquired diseases.

67 Congenital muscular dystrophy 1A (MDC1A) is an autosomal recessive neuromuscular
68 disease associated with a high degree of morbidity and mortality in early childhood ¹. It is
69 caused by mutations in the *LAMA2* gene encoding Laminin α 2 chain (LAMA2), which
70 interacts with the β 1 and γ 1 chains to form the heterotrimer Laminin-211, an extracellular
71 matrix protein (reviewed in ³). Laminin-211 interacts with α -dystroglycan and α 7 β 1
72 integrin in skeletal muscle and Schwann cells, providing the necessary roles such as
73 survival and stability of myotubes, as well as proper neurite growth, axon myelination
74 and migration of Schwann cells. Lack of Laminin-211 in MDC1A causes degeneration of
75 skeletal muscle and impaired Schwann cell differentiation, resulting in a cascade of
76 secondary events including apoptosis/necrosis of muscle fibers, inflammation, and
77 fibrosis, which ultimately precipitate the disease. Despite significant advances in our
78 understanding of MDC1A pathophysiology, currently, there is no cure.
79 Due to the genetic nature of the disease, gene therapy is a promising treatment option
80 for MDC1A. The large size of *LAMA2* transcript, however, presents a challenge with
81 respect to gene delivery. We have recently overcome this challenge by using
82 CRISPR/Cas9 technology to correct a mutation in *Lama2* gene *in vivo* ⁴. We focused on
83 *dy^{2j}/dy^{2j}* mouse model, which harbors a splice site mutation in *Lama2* causing
84 spontaneous exon skipping and truncation of N-terminal domain of the protein ⁵. We
85 developed an exon inclusion strategy to correct the splice mutation, leading to
86 restoration of full-length *Lama2*. This study established the first direct correction of the
87 primary genetic defect underlying MDC1A in an *in vivo* model.
88 To date, there are over 350 reported pathogenic nonsense-, missense-, splice site- and
89 deletion mutations in the *LAMA2* gene. Given the variety of MDC1A-causing genomic
90 alterations, CRISPR/Cas9-mediated gene editing strategies would require design and
91 thorough analysis of multiple sgRNAs specific for each individual mutation. Moreover,
92 safety concerns regarding CRISPR/Cas9's potential mutagenic nature and the presence
93 of off-target effects after gene editing remain, which together may prove to be
94 challenging from a safety and regulatory point-of-view. In contrast, the attenuation of
95 disease pathogenicity by targeted modulation of disease modifier gene expression would
96 be a potentially safer alternative and beneficial to all individuals with MDC1A.
97 One of the strongest reported disease modifiers for MDC1A is Laminin- α 1 protein, which
98 is structurally similar to Laminin- α 2 (**Fig. 1a**). However, Laminin- α 1 is not expressed in
99 skeletal muscles or Schwann cells. A series of studies previously demonstrated that
100 transgenic expression of *Lama1*, encoding Laminin- α 1, rescued both myopathy and

peripheral neuropathy in dy^{2j}/dy^{2j} ⁶ and dy^{3K}/dy^{3K} ^{2,7-10}; the latter also showed increased lifespan. Although these studies firmly established compensatory function of Laminin- α 1 in MDC1A, utilization of this modifier as a postnatal gene therapy is hampered by the size of *Lama1* cDNA, which exceeds the 4.7 kb packaging capacity of AAV vectors. Advances in CRISPR/Cas9 technologies have provided opportunities for regulating gene expression and creating epigenetic alterations without introducing DNA double-strand breaks (DSBs); commonly termed CRISPR transcriptional activation/inhibition system. The strategy utilizes nuclease-deficient Cas9 (dCas9), which is unable to cleave DNA due to mutations within the nuclease domains, but still retains the ability to specifically bind to DNA when guided by a single guide RNA (sgRNA) ^{11,12}. Using the originally described *S. pyogenes* (*Sp*) dCas9 fused to multiple copies of the VP16 transcriptional activator, our group and others have demonstrated utilization of the CRISPR/dCas9 system to upregulate expression of modifier genes *in vitro* ¹¹⁻¹⁴. A major challenge for *in vivo* applications lies in the large size of *SpdCas9* and its transcriptional activator fusion derivatives that exceed AAV genome packaging capacity. To accommodate this limitation, we sought to adapt the transcriptional upregulation system and utilize a significantly smaller Cas9 protein derived from *S. aureus* (*Sa*)¹⁵ to upregulate MDC1A modifier *Lama1*. We hypothesized that CRISPR/dCas9-mediated transcriptional upregulation of *Lama1* would be sufficient to compensate for the lack of *Lama2* and ameliorate disease phenotypes in dy^{2j}/dy^{2j} mice.

First, we mutagenized *SaCas9* endonuclease catalytic residues (D10A, N580A) to create *SadCas9* and then fused it to transcriptional activators VP64 (four copies of VP16) on both N- and C-termini (**Fig. S1**). We tested the ability of the system, denoted as *SadCas9*-2xVP64, to upregulate the expression of minCMV-driven *tdTomato* gene in 293T cells ¹⁶. In the absence of the *SadCas9*-2xVP64, the expression of *tdTomato* was undetectable due to the low baseline activity of minCMV promoter (**Fig. S1a, b**). In the presence of *SadCas9*-2xVP64 in combination with an sgRNA targeting the minCMV locus, we observed high *tdTomato* fluorescence signal (**Fig. S1c, d**), indicating the general applicability of this system to modulate expression of a gene of interest.

Subsequently, we tailored the system to upregulate *Lama1* expression and designed five sgRNAs, denoted as g1 to g5, within the 500 nucleotides region immediately upstream of the *Lama1* transcription start site (**Figs. 1b, c**). When co-expressed with *SadCas9*-VP64 in C2C12 murine myoblasts, 3 out of 5 sgRNAs, namely g1, g2, and g5 consistently induced significant increase of *Lama1* transcript expression (**Figs. 1d, e**).

Although all sgRNAs were designed to target a chromatin-accessible region derived from DNase1 hypersensitivity- and assay for transposase-accessible chromatin (ATAC-Seq) sites (**Fig. 1b**), g3 and g4 failed to increase expression of *Lama1* above the basal level. This corroborates previously reported findings that chromatin accessibility is not a reliable predictor of successful gene activation^{12,17,18}. Nonetheless, the sgRNA closest to the transcription start site, g1, and the combination of the three most optimum sgRNAs (g1, g2, g5) resulted in a robust increase in Lama1 protein expression in *dy^{2j}/dy^{2j}*-derived myoblasts (**Figs. 1f, 1g**), warranting further investigation *in vivo*.

We then treated 3-week-old *dy^{2j}/dy^{2j}* mice with an AAV9 carrying FLAG-tagged SadCas9-2xVP64 in the absence of sgRNA (no guide) or with g1 (single guide) or a combination of g1, g2 and g5 (three guides) (**Fig. 2a**). Due to the packaging capacity of the AAV, the dCas9-2xVP64 and the three guides were split into two vectors (**Fig. 2a**). Each mouse received a single intramuscular injection of 7.5×10^{11} viral genomes of AAV9, which was doubled to 1.5×10^{12} viral genomes total for the three guide cohort, in the right *tibialis anterior* (TA) and was sacrificed 4 weeks post injection. FLAG expression was detected by western blot in all SadCas9-2xVP64-injected right TA muscles, however, only those injected with guide-containing constructs showed Lama1 upregulation (**Fig. 2b**). Similarly, immunofluorescence staining revealed sarcolemmal expression of Lama1 (**Fig. 2c**, upper), indicating proper protein localization. Furthermore, H&E staining exhibited considerably improved muscle architecture (**Fig. 2c**, lower) in the single- and three guide treatment groups, as compared to the no guide controls.

Next, we sought to investigate whether upregulation of *Lama1* could be achieved systemically *in vivo* and, if administered into pre-symptomatic neonatal *dy^{2j}/dy^{2j}* mice, would prevent the manifestation of dystrophic pathophysiology and paralysis. AAV9 particles carrying either no guide or a combination of three guides were injected into the temporal vein of 2-day-old (P2) *dy^{2j}/dy^{2j}* mice (**Fig. 3a**). Seven weeks post injection, the animals that received the three guides treatment showed an absence of classical hindlimb contracture, unlike the control group (**Fig. 3b; Supplementary videos 1 and 2**). Western blot, immunofluorescence and H&E staining of TA and gastrocnemius muscles demonstrated robust expression of Lama1 on the sarcolemmal membrane (**Fig. 3c, 3d**), leading to ~50% reduction of fibrosis (**Figs. 3e, 3f, S2**).

We subsequently examined the ability of *Lama1* upregulation to reverse established muscular and peripheral nerve dysfunctions by initiating the treatment at 3 weeks of age,

when paralysis was already apparent^{19,20} (**Fig. 4a**). We first tested three different doses, ranging from 7.5×10^{10} to 3×10^{11} viral genome/gram of mouse per AAV9, which was doubled for the three guide cohorts due to the utilization of two vectors, and found that the highest dose resulted in homogeneous Lama1-positive fibers (**Fig. S3a**), significant improvement of muscle function and mobility (**Figs. S3b, S3c**). Longitudinal measurement of animal mobility and stand-up activity in a non-invasive open field activity assay showed significant difference between no guide and three guides-treated mice starting at 5 and 6 weeks old, respectively, which was sustained over time (**Figs. 4b, 4c**). Furthermore, specific tetanic force, which measures the aggregate torque produced by the dorsi flexor muscles, was also improved in the three guides-treated mice (**Fig. 4d**). In line with this finding, we observed a significant increase in nerve conduction velocity, which indicates restoration of the myelination defect and contributes to neuromuscular functionality (**Fig. 4e**). The absence of paralysis in the hind limbs and markedly improved movement of the mice were evident at the end of the treatment regimen (**Supplementary videos 3, 4**).

Molecular analysis of the treated mice revealed strong Lama1 expression by immunostaining and western blot (**Figs. 4f, 4g, S4**), which was accompanied by normalization of $\alpha 4$ chain of the laminin subunit (**Figs. S5, S6**)⁶, significant improvement in muscle histopathology (**Fig. 4h**) and approximately 50% reduction in the fibrotic area (**Fig. 4i**) compared to the no guide control group. There was a trend towards larger fibers in the treated mice, although it did not reach statistical significance due to the large variation between animals (**Fig. 4j**). In addition, upregulation of Lama1 was also observed in the endoneurium of sciatic nerves and resulted in restoration of myelination defect (**Figs. 4k, S7**), supporting the improvement of nerve conduction velocity and lack of paralysis in the mice (**Fig. 4e, Supplementary videos 3, 4**). Quantification of the AAV genome copy number revealed accumulation of most of the viral genome in the liver, which is expected from intravenous delivery. Nevertheless, approximately 1.11 ± 0.4 and 34.1 ± 4.3 copies/diploid genome of the viral genome were detected in the sciatic nerves and skeletal muscles, respectively (**Fig. S8**). Remarkably, even relatively low transduction efficiency in sciatic nerves was sufficient for functionally significant upregulation of Lama1 expression.

Finally, we investigated the genome wide effects of CRISPR/dCas9-mediated Lama1 upregulation by performing RNA-sequencing on quadriceps muscles isolated from mice treated with AAV9 carrying no guide and three guides (**Figs. S9, S10, Supplemental**

Tables 1-3). Age-matched wildtype and dy^{2j}/dy^{2j} mice served as controls. We observed a 3.6-log₂fold upregulation of *Lama1* (defined by a false discovery rate, FDR <0.05) when comparing the mice treated with AAV9 carrying no guide and three guides (**Fig. S9a**). The transcriptional change was even higher at 9-log₂fold when comparing untreated dy^{2j}/dy^{2j} with three guide-treated dy^{2j}/dy^{2j} (**Fig. S9b, S9c**). Hierarchical clustering between groups revealed clustering between wildtype and three guides, whereas the untreated cohort was clustered together with no guide-treated mice (**Figs. S9d, S10**). We also computationally predicted 704 potential off-target binding sites for the three sgRNAs targeting *Lama1* promoter in the mouse genome, selected based on the presence of a 6bp PAM-proximal seed sequence and fewer than ten total mismatches to the cognate target sequence (**Supplemental Tables 4-5**). None of the top 100 genes contained a predicted off-target site within 60 kilobases of the gene body, with the average distance from off-target site to the gene body being 2.2 megabases. Taken together, our results provide strong evidence of the robustness and durability of CRISPR/dCas9-mediated *Lama1* upregulation for the treatment of MDC1A. Additionally, we show the therapeutic promise of the strategy for reversal of dystrophic feature in skeletal muscle and peripheral neuropathy in dy^{2j}/dy^{2j} mouse model of MDC1A, which ultimately halts progression of the disease, without any confounding off-target effects.

One challenge in developing a therapy for MDC1A is that the heterogeneity of mutations often leads to variable disease severity and progression. Therefore, there is an urgent need to develop a universal, mutation-independent strategy that provides a treatment approach for all patients with MDC1A. Our study establishes a framework in which CRISPR/dCas9 transcriptional upregulation of a disease modifier gene, such as *Lama1*, ameliorates disease symptoms *in vivo* and has the potential to be applied to all MDC1A patients, irrespective of their mutations.

Advances have been made towards elucidating MDC1A pathogenesis due to the availability of several mouse models with absent or reduced *Lama2* expression. Yet, an important question in the development of therapeutics and clinical trials in MDC1A is the reversibility of symptoms caused by muscle fibrosis and nerve abnormalities. This issue has been challenging to address in patients²¹, however, the ability to modify gene expression in postnatal animals allowed us to address this question and begin to investigate the therapeutic window of intervention.

We have previously demonstrated that an early intervention using CRISPR/Cas9-mediated correction of a splicing defect resulted in robust *Lama2* restoration and prevention of disease manifestation⁴. Here we showed that upregulation of *Lama1*, when initiated at pre-disease-onset, leads to similar prevention. Importantly, when the therapeutic intervention was initiated at older age, significant rescue of the phenotypes was attainable, indicating that post-symptomatic treatment provides a significant benefit in the *dy^{2j}/dy^{2j}* mouse model.

In addition to *Lama1* upregulation described in this study, a number of disease modifying strategies are currently being explored in MDC1A animal models, including treatment with miniaturized agrin²²⁻²⁴ and laminin- α 1 LN-domain nidogen-1 (α LNNd)²⁵⁻²⁷. While the efficacy of α LNNd has only been explored in transgenic mice, AAV-mediated delivery of mini agrin has been shown to normalize most histopathological parameters in skeletal muscle, and improve myelination and regeneration of Schwann cells of the peripheral nerves^{22,24}. Despite the observed phenotypic improvement, the mini agrin-treated mice still have a lower survival rate compared to wild-type animals expressing full-length agrin, suggesting the potential shortcoming of the shortened version of agrin, which may be overcome by its full-length form in native glycosylation state²⁵. It is important to note that many preclinical studies in MDC1A were carried out in the more severe *dy^w/dy^w* mouse model. Therefore, as a follow up on our study, it will be important to evaluate our approach in the *dy^w/dy^w* mice to assess critical parameter such as survival, which is not

possible in the dy^{2j}/dy^{2j} mouse model due to its near-normal life span. Overall, our approach in combining the CRISPR/dCas9-mediated transcriptional upregulation and AAV9 as a delivery vehicle can be translated to many other disease modifiers, such as agrin, as well as conditions where modulation of disease modifiers is required within both skeletal muscles and peripheral nerves.

In fact, neuromuscular disorders have provided excellent examples to demonstrate the role of disease modifiers (recently reviewed in ²⁸). Beyond MDC1A, several studies have demonstrated that upregulation of *Lama1* stabilizes the sarcolemmal membrane in dystrophin-deficient mouse models ²⁹. The most advanced approach is via delivery of Laminin-111 protein, although the efficacy remains low and the need to produce a large amount of bioactive protein is challenging ²⁹. In addition, the utilization of CRISPR/dCas9 system to upregulate *Lama1* in *mdx* mice has been achieved locally via electroporation, which is not easily translatable into clinical settings ³⁰. Our strategy of employing AAV-mediated *S. aureus* dCas9 to upregulate *Lama1* *in vivo* may be tested further as a potential therapy in the context of Duchenne muscular dystrophy.

In addition, since the CRISPR/dCas9-mediated transcriptional modulation acts directly on the endogenous locus, it may be used in conjunction with a mutation-correction approach where the level of restoration of the defective gene is suboptimal, therefore necessitating further amplification to reach therapeutic efficacy ^{4,31-36}.

A very recent study by Liao *et al* described utilization of the CRISPR/Cas9 system to recruit MCP:P65:HSF1 transcriptional activation complex to induce expression of target genes in skeletal muscle, kidney and liver tissues ¹⁸. This resulted in phenotypic augmentation such as enhanced muscle mass and substantial improvement in disease pathophysiology, thereby highlighting the feasibility of using CRISPR/dCas9-mediated transcriptional activation as a possible therapeutic modality. However, their study relied almost exclusively on a Cas9-expressing transgenic mouse model or local intramuscular treatments, and therefore it is difficult to extrapolate the efficacy of this strategy to disease-relevant models. In contrast, we successfully demonstrated robust upregulation of *Lama1* after systemic delivery of therapeutic components in a relevant mouse model of disease that does not constitutively express Cas9.

Finally, the modular nature of the CRISPR/dCas9 system can be utilized to not only to upregulate, but also to downregulate target gene expression. The latter can be achieved by coupling dCas9 with transcriptional repressor such as Kruppel-associated box (KRAB) ^{17,37}. A very recent study described that following sarcolemmal injury, the muscle

290 membrane resealing process is greatly improved upon the deletion of Osteopontin,
 291 which acts in a concerted fashion with protective modifiers such as Latent TGF- β binding
 292 protein (LTBP4) and Annexins 1 and 6^{38,39}. The combinatorial effects of such modifiers,
 293 whether they are additive, synergistic or even opposing in action, represent a new
 294 paradigm for lessening disease phenotypes. A foreseeable application of
 295 CRISPR/dCas9-mediated modulation is in the upregulation of protective disease
 296 modifier genes, such as Lama1 or LTBP4, with concurrent downregulation of detrimental
 297 genes, such as Osteopontin, providing a combinatorial therapeutic approach.
 298 In summary, our study establishes a framework to utilize CRISPR/dCas9 to modulate
 299 gene expression of disease modifiers that should be considered as a mutation-
 300 independent therapeutic strategy not only to MDC1A, but also to various other inherited
 301 and acquired diseases.

Methods

Engineering of activation constructs

A fragment containing a catalytically inactive *SaCas9* coupled to two flanking VP64 transactivator domains was synthesized by BioBasic Canada and cloned into pX601 (Addgene 61591) using *AgeI* and *EcoRI* directional cloning to generate 3XFLAG-VP64-SadCas9 (D10A/N580A)-NLS-VP64 plasmid (**Figs. 2a, S11, Supplemental Table S6**). Each sgRNA (**Supplemental Table S6**) was subsequently introduced using *BsaI* directional cloning. To generate the three guides only construct (**Fig. 2a**), a fragment containing three repetitive regions of U6 promoter and *S. aureus* guide scaffold was assembled, with short linkers in between each region (BioBasic Canada). The fragment was cloned into *KpnI* and *NotI* sites of a pX601-derivative plasmid.

Cell Culture

Primary myoblasts were isolated from the Extensor Digitorum Longus (EDL) Muscle of *dy^{2J}/dy^{2J}* mice, as previously described⁴⁰ and maintained in DMEM supplemented with 1% chicken embryo extract (GeminiBioscience), 10% horse serum, 1% penicillin/streptomycin and 1% L-glutamine (all from Gibco, unless indicated otherwise). HEK293 and C2C12 cells were maintained in DMEM supplemented with 10% FBS, 1% penicillin/streptomycin and 1% L-glutamine (all from Gibco). All cells were maintained at 37°C with 5% CO₂.

Transfection of HEK293T cells was performed as previously described². C2C12 and *dy^{2J}/dy^{2J}* cells were transfected in 12-well plates using the Neon Transfection System (Invitrogen). 400,000 cells were electroporated with 1.5 µg of DNA utilizing optimization program 16 (pulse voltage: 1400V, pulse width: 20ms, pulse number: 2). Cells were grown for 72 hours, after which RNA or protein was subsequently collected for protein analysis and guide screening.

Animals, virus production and injections

dy^{2J}/dy^{2J} mice were purchased from the Jackson Laboratory and maintained in the Toronto Center for Phenogenomics. Both male and female were used in the analyses. All animal experiments were performed according to Animal Use Protocol number 20-0305. SadCas9-2xVP64, single guide and three guides plasmids (**Fig. 2a, Supplemental Table S6**) were packaged into AAV9 vectors by Vigene Biosciences as

previously described³. For intramuscular and temporal vein injection into neonatal pups, the dose of 7.5×10^{11} viral genomes each was used. Due to the limitation in packaging capacity, two AAVs were needed for the three guides cohort (**Fig. 2a**), therefore the total virus injected was 1.5×10^{12} viral genomes per animal. Injection volume was brought to 50 μ l with 1XPBS (Gibco).

For the tail vein injection in young, 3 week old mice, three different doses were initially tested: 7.5×10^{10} , 1.5×10^{11} and 3×10^{11} viral genomes per gram of mouse (**Fig. S3**).

Similar to the intramuscular and temporal vein injections, two AAVs were needed for the three guides cohort, therefore the total dose used in experiments described in **Figs. 4, S4-S10** was 6×10^{11} viral genomes per gram of mouse. Injection volume was brought to 100 μ l with 1XPBS (Gibco).

RNA isolation, guide screening and RT-PCR

RNA was isolated from cultured cells and mouse tissue sections, and cDNA synthesis was performed as previously described³. PCR amplification was utilized to assess the efficiency of each guide in upregulating *Lama1* expression using a primer in *Lama1* exon 55 (RDC 1919) and a second primer spanning the junction of exons 55 and 56 (RDC 1920). Sequences are listed in **Supplemental Table S6**.

qPCR utilizing Fast SYBR green Master Mix (Qiagen) on a Step One Plus Real Time PCR (Applied Biosystems) was performed. *Lama1* expression was analyzed using a primer in *Lama1* exon 55 (RDC 1919) and one spanning the junction of exons 55 and 56 (RDC 1920). Primers against endogenous Gapdh (RDC 345 and 346) were used as an internal control. $\Delta\Delta C_t$ was analyzed to assess fold changes between treated and untreated samples.

Protein Isolation and western blot

Protein was isolated from *dy^{2j}/dy^{2j}* myoblasts and C2C12 cells by adding 150 μ l of a 1:1 part solution of RIPA homogenizing buffer (50-mM Tris HCl pH 7.4, 150-nM NaCl, 1-mM EDTA) and RIPA double-detergent buffer (2% deoxycholate, 2% NP40, 2% Triton X-100 in RIPA homogenizing buffer) supplemented with protease-inhibitor cocktail (Roche). Cells were subsequently scraped from the bottom of each well, collected and incubated on ice for 30 min. Cells then centrifuged at 12000xg for 15 min at 4°C and the supernatant was collected and stored at -80°C. Protein from mouse tissue sections was collected as previously described³. Whole protein concentration was measured using

Pierce BCA protein assay kit according to the manufacturer's protocol (Thermo Fisher Scientific). Western blot was performed as previously described³. Primary antibodies used were rabbit Anti-LN α 1 E3 (a gift from Dr. Peter Yurchenco, 0.6 μ g/ml), mouse monoclonal M2 anti-Flag (Sigma Aldrich F1804, 1:1000) and rabbit polyclonal anti-GAPDH (Santa Cruz sc-25778, 1:5000).

Immunofluorescence and H&E staining

Muscles and nerves were sectioned at 8 μ m thickness and processed for immunofluorescence analyses according to standard procedures. Antibodies used for immunofluorescence staining were rat monoclonal against Laminin α 1 (mAb200, Durbeej Lab, 1:20), α 2 (4H8-2, Sigma, 1:500), γ 1 chain (clone A5, Thermo Fisher Scientific), rabbit polyclonal against Laminin α 4 chain (kindly provided by Dr. Sasaki), mouse monoclonal against NF-H (Biolegend SMI 31, 1:1000), goat polyclonal anti-rat Alexa Fluor 555 (Thermo Fisher Scientific, 1:250) and goat polyclonal anti-mouse Alexa Fluor 488 (Thermo Fisher Scientific, 1:250). H&E staining was performed as previously described⁴. Both immunofluorescence and H&E slides were scanned with the 3Dhistech Panoramic 250 Flash II digital scanner and analyzed with CaseViewer software, with the exception of **Figs. S5, S6**, which were analyzed with a Zeiss Axioplan fluorescence microscope (Carl Zeiss) and images were captured using an ORCA 1394 ER digital camera (Hamamatsu Photonics) and Open Lab software version 4 (Improvision).

Toluidine blue staining and electron microscopy

Freshly isolated mouse sciatic nerves were halved and fixed in a solution of 2% paraformaldehyde and 2.5% glutaraldehyde in 0.1M sodium cacodylate buffer until further use. For embedding, the specimens were rinsed with the 0.1M sodium cacodylate buffer, post-fixed in 1% osmium tetroxide in the washing buffer, dehydrated in a graded ethanol series followed by propylene oxide, and embedded in Quetol-Spurr resin overnight at 65°C. Sections of 90nm thickness were cut on a Leica EM UC7 ultramicrotome, stained with uranyl acetate and lead citrate, and imaged on a FEI Tecnai 20 electron microscope at 4400-, 10,000-, and 44,000X magnifications. The same 90nm sections were stained with toluidine blue and imaged on a Leica DM-2000. All reagents were purchased from Electron Microscopy Sciences. Quantification of myelin thickness was measured using ImageJ from at least 14 random axons per field (at least 5 fields per animal)⁴¹.

Open field and *in vivo* muscle force assays

Open field activity test and assessment of *in vivo* muscle force were performed on tail vein injected cohorts at the Lunenfeld-Tanenbaum Research Institute's Centre for Modeling Human Disease Mouse Phenotyping Facility. For the open field test, mice were placed in the frontal center of a transparent Plexiglas open field (41.25 cm × 41.25 cm × 31.25 cm) illuminated by 200 lx. A trained operator, who is unaware of the nature of the projects and treatments, performed experiments. The VersaMax Animal Activity Monitoring System recorded vertical activities and total distance travelled for 20 minutes per animal.

In vivo muscle contraction test was performed using 1300A: 3-in-1 Whole Animal System and analyzed using Dynamic muscle control/analysis (DMC/DMA) High throughput software suite (Aurora Scientific). The mice were anaesthetised with intraperitoneal injection of ketamine/xylazine cocktail at 100 mg/kg and 10 mg/kg of body weight, respectively. Contractile output was measured via percutaneous electrodes that stimulate specific nerves innervating the plantar flexors. Specific tetanic force (200 ms of 0.5-ms pulses at 125 Hz) was recorded and corrected to body weight.

Nerve conduction velocity

Mice were anesthetized with urethane (1.2 mg/g i.p.) and underwent a tracheotomy to maintain their airway but were not artificially ventilated. Core body temperature was maintained at 34-36.5°C using a feedback-controlled heating pad (TR-200; Fine Science Tools). The sciatic nerve was exposed at two locations via incisions (~15 mm) above the left knee (site #1) and along the sacral region of the vertebral column (site #2). After separating the nerve from adjacent tissue, a bipolar hook electrode was applied to the nerve at each site. Each electrode comprised two chlorided silver wires (0.01" diameter, A-M systems) placed ~1 mm apart and bent at the tip to form hooks, and attached to a stimulator (Model DS3, Digitimer Ltd). Stimulating electrodes were insulated from underlying tissue using a small piece of plastic paraffin film. Stimulus duration was fixed at 20 µs and current intensity was varied. Stimulus timing was controlled by computer using a Power1401 computer interface and Signal v5 software (Cambridge Electronic Design, CED). The compound muscle action potential (CMAP) was recorded using needle electrodes (~30 G, BD *PrecisionGlide*™), one inserted into the gastrocnemius muscle and the other (reference) electrode inserted into the Achilles tendon. The CMAP signal was amplified, low-pass filtered at 10 Hz and high-pass filtered at 1 kHz (DAM 80,

World Precision Instruments), and digitized at 40 kHz using the Power1401 and Signal v5 software (CED). To calculate conduction velocity (CV) from CMAP responses, the sciatic nerve was stimulated at just-maximal intensity (beyond which there is no change in CMAP amplitude or latency) three times at site #1 and again three times at site #2. Nerve conduction velocity was calculated as the difference between average CMAP latencies for each stimulation site divided by the length of nerve (7.5-10 mm) separating the sites.

Vector genome quantification

Evaluation of AAV genome distribution was performed as previously published⁴² with a few modifications. Genomic DNA was extracted from tibialis anterior, sciatic nerve, and liver of treated mice using Qiagen Blood and Tissue Kit. 90 ng of DNA was amplified using primers located in between the two inverted tandem repeats (ITRs) (RDC 1687, RDC 1679, **Supplemental Table S7**) using Fast SYBR green master mix (Qiagen) on a Step One Plus Real Time PCR (Applied Biosystems). The Ct value of each reaction was converted to viral genome copy number by interpolating against the copy number of standard curve of a known plasmid containing the sgRNA cassette (RDC 362). The amount of DNA input (90 ng per tissue) was used as a conversion factor to diploid genome (7 picogram DNA = 1 diploid genome).

RNA-Sequencing

Total RNA was isolated from quadriceps muscles using RNeasy kit (Qiagen) and quantified using Qubit RNA HS assay (Thermo Fisher Scientific). RNA-Sequencing was performed by the Centre for Applied Genetics in Toronto using the Illumina HiSeq 2500 system, producing 120bp paired-end reads. Raw transcript reads were aligned to the GRCm38 mouse genome (mm10) using HISAT2⁴³. HTSeq was used to determine the absolute number of read counts for each gene⁴⁴. Only genes with at least 1 read per million in at least three replicates were kept for downstream analysis. Normalization and differential expression analysis was completed using the R packages limma, v3.36.3 and edgeR, v.3.22.3⁴⁵. Differentially expressed genes were defined as genes with a more than twofold change and adjusted $P < 0.05$. Off-target analysis was conducted using a list of 704 computationally predicted using Cas-OFFinder⁴⁶. Using the Bedtools suite “closest” function, each of the top 100 differentially expressed genes was matched to the

nearest off-target loci to determine the shortest distance from off-target loci to differentially expressed gene body.

Statistical Analysis

GraphPad Prism (GraphPad software) was utilized to perform all statistical analyses. Two-tailed Student's t-tests to evaluate statistical significance between two groups was performed. Significance was considered to be $P < 0.05$.

Data availability

The authors declare that the main data supporting the findings of this study are available within the article and its Supplementary Information files. Extra data are available from the corresponding authors upon request.

Acknowledgements

The Cohn lab members are gratefully acknowledged for the technical support and critical input in this study. We thank P. Yurchenco (Rutgers University) for providing critical reagents in this study, C. Rand (Aurora Scientific), I. Vukobradovic (Clinical Phenotyping Core, Centre for Modeling Human Disease), R. Smith (Prescott Lab) for assistance with functional and behavioral studies, D. Holmyard (Nanoscale Biomedical Imaging Facility) for electron microscopy analysis and S. Pereira (The Centre for Applied Genomics) for genomic data acquisition.

This work was supported by AFM-Telethon, Cure CMD, Muscular Dystrophy Association (to D.U.K.), Rare Disease Foundation Microgrant (to P.S.B. and S.E.), SickKids Restracom (to S.E.), CIHR Summer Studentship (to R.M.), Natural Sciences and Engineering Research Council of Canada, Canadian Institute for Health Research, SickKids Foundation and R.S. McLaughlin Foundation Chair in Pediatrics (to R.D.C).

Author contributions

D.U.K., E.A.I., R.D.C. conceived the study, D.U.K., P.S.B., S.E., D.A.B., K.I.G., K.L., E.H., R.K., K.M.P., R.M., performed the experiments, D.A.B., K.I.G., M.D., S.A.P. provided critical reagents, D.U.K., P.S.B., S.E., D.A.B., K.I.G., S.A.P., E.A.I., R.D.C. analyzed data, D.U.K., E.A.I., R.D.C. supervised the study, D.U.K. wrote the manuscript with inputs from the other authors. All authors provided feedback and agreed on the final manuscript.

Figure legends

Figure 1

Fig. 1. SadCas9-2xVP64-mediated upregulation of *Lama1* in vitro. (a) Lama1 and Lama2 protein alignment. Total amino acid (aa) and percentage similarity between each domain are indicated. (b) Analyses of Lama1 proximal promoter. Five sgRNAs (g1-g5) were designed in the proximal promoter region of Lama1 immediately upstream of the transcription start site (TSS), as indicated by CAGE tags (red). Chromatin accessibility in skeletal muscle tissue and cells (retrieved using Digital DNase footprinting and ATAC-Seq) are shown in blue. Data were plotted according to positions from the UCSC Genome Browser. (c) Positions of the five sgRNAs relative to *Lama1* TSS. Arrowheads indicate the direction of each sgRNA. Sequences of each sgRNA (red) are immediately downstream (5') of *Sa* PAM sequences (NNGRRT) (in blue). ATG indicates translation start site. (d-e) C2C12 myoblasts were transfected with a plasmid containing SadCas9-2xVP64 and the corresponding sgRNA(s) targeting *Lama1*, and 72 hours post-transfection, analyzed by (d) RT-PCR and (e) qRT-PCR. Single and combination of optimal sgRNAs were transfected into *dy^{2j}/dy^{2j}* myoblasts and Lama1 protein expression was assessed by western blot (f). FLAG expression serves as transfection control and used to (g) normalize Lama1 upregulation by densitometry analysis. Data are presented

as mean $\pm$ standard deviation from at least three independent experiments. Statistical analysis was performed using Student's *t*-test. $**P<0.01$.

Figure 2

Fig. 2. Upregulation of Lama1 in *tibialis anterior* (TA) muscles of *dy^{2j}/dy^{2j}* mice following intramuscular administration. (a) Right TA muscles of 3-week old *dy^{2j}/dy^{2j}* mice were injected with AAV9-carrying no guide (n=4; 7.5x10¹¹ viral genomes), single guide (n=4; 7.5x10¹¹ viral genomes) or three guides (n=4; split into two vectors, thus total dose was 2x7.5x10¹¹ viral genomes). ITR: Inverted Terminal Repeats. CMV and U6 promoters are depicted in arrows. Left TA muscles serve as control. (b) Western blot analysis on Lama1, FLAG-tagged SadCas9, and Gapdh expression. (c) Immunofluorescence (upper) and H&E (lower) stainings on cross-sections of right TA muscles from each treatment group. Scale bar: 50 μ m. Representative images from one animal per treatment group are shown.

Figure 3

Fig. 3. Early intervention to upregulate Lama1 prevents disease progression in *dy^{2j}/dy^{2j}* mice. (a) Two-day-old neonatal *dy^{2j}/dy^{2j}* mice were injected with AAV9 carrying no guide (n=3; 7.5x10¹¹ viral genomes) or three guides (n=4; split into two vectors, thus total dose was 2x7.5x10¹¹ viral genomes) via temporal vein and sacrificed 7 weeks later. (b) Photographs of *dy^{2j}/dy^{2j}* from both treatment groups prior to sacrificing. Note the

difference in hindlimb contractures. Lama1 expression was analyzed by (c) western blot and (d) immunofluorescence staining, and general histopathology was evaluated by (e) H&E staining. The muscle groups analyzed are indicated on each panel, except on (c), which shows tibialis anterior (TA) muscles. Scale bars: 100 μ m (d), 200 μ m (e). (f) The percentage of fibrosis from gastrocnemius (upper) and TA (lower) muscles are calculated and presented as mean $\pm$ standard deviation. Statistical analysis was performed using Student's *t*-test. **P*<0.05.

Figure 4

Fig. 4. Upregulation of Lama1 in older dy^{2j}/dy^{2j} mice halts disease progression.

(a) Three-week old dy^{2j}/dy^{2j} mice were injected with AAV9 carrying no guide (denoted as -; n=9; 3×10^{11} viral genomes/gram) or three guides (denoted as +; n=6; split into two vectors, thus total dose was $2 \times 3 \times 10^{11}$ viral genomes/gram) via tail vein. Grey box represents period before treatment. (b, c) Open field activity assay was performed weekly, before (grey-shaded) and after treatment, and the mice were tested for (d) muscle contractile and (e) nerve conduction velocity at the end of the treatment regimen, prior to molecular analyses of the tissues. (f) Immunofluorescence staining and (g) western blot to analyze Lama1 expression and (h) H&E staining to quantify (i) fibrosis and (j) fiber size were performed on gastrocnemius muscles. (k) Sciatic nerves were stained for Lama1 (red) and Neurofilament H (green) (left). Nuclei were counterstained by DAPI (blue). Higher magnification images from the dotted area are shown. Representative toluidine blue staining (middle) and electron micrograph (right) are shown. Asterisks indicate region of amyelinated axon fibers. Scale bars: 100 μ m (f), 50 μ m (h), and 200 μ m (k). Data are presented as mean $\pm$ standard deviation. Statistical analysis was performed using Student's *t*-test. **P*<0.05. ***P*<0.01.

References

1. Dowling, J. J., H, D. G., Cohn, R. D., Campbell, C. Treating pediatric neuromuscular disorders: The future is now. *American journal of medical genetics. Part A* 2017.
2. Gawlik, K., Miyagoe-Suzuki, Y., Ekblom, P., Takeda, S., Durbeej, M. Laminin alpha1 chain reduces muscular dystrophy in laminin alpha2 chain deficient mice. *Human molecular genetics* 13:16. 2004.
3. Yurchenco, P. D., McKee, K. K., Reinhard, J. R., Ruegg, M. A. Laminin-deficient muscular dystrophy: Molecular pathogenesis and structural repair strategies. *Matrix biology : journal of the International Society for Matrix Biology* 2017.
4. Kemaladewi, D. U., Maino, E., Hyatt, E., Hou, H., Ding, M., Place, K. M., Zhu, X., Bassi, P., Baghestani, Z., Deshwar, A. G., Merico, D., Xiong, H. Y., Frey, B. J., Wilson, M. D., Ivakine, E. A., Cohn, R. D. Correction of a splicing defect in a mouse model of congenital muscular dystrophy type 1A using a homology-directed-repair-independent mechanism. *Nat Medicine*. 23:8. 2017.
5. Sunada, Y., Bernier, S. M., Utani, A., Yamada, Y., Campbell, K. P. Identification of a novel mutant transcript of laminin alpha 2 chain gene responsible for muscular dystrophy and dysmyelination in dy2J mice. *Human molecular genetics* 4:6. 1995.
6. Gawlik, K. I., Harandi, V. M., Cheong, R. Y., Petersen, A., Durbeej, M. Laminin alpha1 reduces muscular dystrophy in dy(2J) mice. *Matrix biology : journal of the International Society for Matrix Biology* 2018.
7. Gawlik, K. I., Li, J. Y., Petersen, A., Durbeej, M. Laminin alpha1 chain improves laminin alpha2 chain deficient peripheral neuropathy. *Human molecular genetics* 15:18. 2006.

8. Gawlik, K. I., Mayer, U., Blomberg, K., Sonnenberg, A., Ekblom, P., Durbeej, M. Laminin alpha1 chain mediated reduction of laminin alpha2 chain deficient muscular dystrophy involves integrin alpha7beta1 and dystroglycan. *FEBS letters* 580:7. 2006.
9. Gawlik, K. I., Akerlund, M., Carmignac, V., Elamaa, H., Durbeej, M. Distinct roles for laminin globular domains in laminin alpha1 chain mediated rescue of murine laminin alpha2 chain deficiency. *PloS one* 5:7. 2010.
10. Gawlik, K. I., Durbeej, M. Transgenic overexpression of laminin alpha1 chain in laminin alpha2 chain-deficient mice rescues the disease throughout the lifespan. *Muscle & nerve* 42:1. 2010.
11. Maeder, M. L., Linder, S. J., Cascio, V. M., Fu, Y., Ho, Q. H., Joung, J. K. CRISPR RNA-guided activation of endogenous human genes. *Nature methods* 10:10. 2013.
12. Perez-Pinera, P., Kocak, D. D., Vockley, C. M., Adler, A. F., Kabadi, A. M., Polstein, L. R., Thakore, P. I., Glass, K. A., Ousterout, D. G., Leong, K. W., Guilak, F., Crawford, G. E., Reddy, T. E., Gersbach, C. A. RNA-guided gene activation by CRISPR-Cas9-based transcription factors. *Nature methods* 10:10. 2013.
13. Gonzalez, S., Fernando, R. N., Perrin-Tricaud, C., Tricaud, N. In vivo introduction of transgenes into mouse sciatic nerve cells in situ using viral vectors. *Nature protocols* 9:5. 2014.
14. Wojtal, D., Kemaladewi, D. U., Malam, Z., Abdullah, S., Wong, T. W., Hyatt, E., Baghestani, Z., Pereira, S., Stavropoulos, J., Mouly, V., Mamchaoui, K., Muntoni, F., Voit, T., Gonorazky, H. D., Dowling, J. J., Wilson, M. D., Mendoza-Londono, R., Ivakine, E. A., Cohn, R. D. Spell Checking Nature: Versatility of CRISPR/Cas9 for Developing Treatments for Inherited Disorders. *Am J Hum Genet* 98:1. 2016.
15. Ran, F. A., Cong, L., Yan, W. X., Scott, D. A., Gootenberg, J. S., Kriz, A. J., Zetsche, B., Shalem, O., Wu, X., Makarova, K. S., Koonin, E. V., Sharp, P. A., Zhang, F. In vivo genome editing using Staphylococcus aureus Cas9. *Nature* 520:7546. 2015.
16. Esvelt, K. M., Mali, P., Braff, J. L., Moosburner, M., Yaung, S. J., Church, G. M. Orthogonal Cas9 proteins for RNA-guided gene regulation and editing. *Nature methods* 10:11. 2013.
17. Qi, L. S., Larson, M. H., Gilbert, L. A., Doudna, J. A., Weissman, J. S., Arkin, A. P., Lim, W. A. Repurposing CRISPR as an RNA-guided platform for sequence-specific control of gene expression. *Cell* 152:5. 2013.
18. Liao, H. K., Hatanaka, F., Araoka, T., Reddy, P., Wu, M. Z., Sui, Y., Yamauchi, T., Sakurai, M., O'Keefe, D. D., Nunez-Delgado, E., Guillen, P., Campistol, J. M., Wu, C. J., Lu, L. F., Esteban, C. R., Izpisua Belmonte, J. C. In Vivo Target Gene Activation via CRISPR/Cas9-Mediated Trans-epigenetic Modulation. *Cell* 171:7. 2017.
19. Pasteuning-Vuhman, S., Putker, K., Tanganyika-de Winter, C. L., Boertje-van der Meulen, J. W., van Vliet, L., Overzier, M., Plomp, J. J., Aartsma-Rus, A., van Putten, M. Natural disease history of the dy2J mouse model of laminin alpha2 (merosin)-deficient congenital muscular dystrophy. *PloS one* 13:5. 2018.
20. Kemaladewi, D. U., Benjamin, J., Hyatt, E., Ivakine, E. A., Cohn, R. D. Increased polyamine as protective disease modifier in Congenital Muscular Dystrophy. *Human molecular genetics* 2018.
21. Bonnemann, C. G., Wang, C. H., Quijano-Roy, S., Deconinck, N., Bertini, E., Ferreira, A., Muntoni, F., Sewry, C., Beroud, C., Mathews, K. D., Moore, S. A., Bellini, J., Rutkowski, A., North, K. N., Members of International Standard of Care Committee for Congenital Muscular, D. Diagnostic approach to the congenital muscular dystrophies. *Neuromuscular disorders : NMD* 24:4. 2014.
22. Qiao, C., Li, J., Zhu, T., Draviam, R., Watkins, S., Ye, X., Chen, C., Li, J., Xiao, X. Amelioration of laminin-alpha2-deficient congenital muscular dystrophy by somatic

- gene transfer of miniagrin. *Proceedings of the National Academy of Sciences of the United States of America* 102:34. 2005.
23. Bentzinger, C. F., Barzaghi, P., Lin, S., Ruegg, M. A. Overexpression of mini-agrin in skeletal muscle increases muscle integrity and regenerative capacity in laminin-alpha2-deficient mice. *FASEB journal : official publication of the Federation of American Societies for Experimental Biology* 19:8. 2005.
24. Qiao, C., Dai, Y., Nikolova, V. D., Jin, Q., Li, J., Xiao, B., Li, J., Moy, S. S., Xiao, X. Amelioration of Muscle and Nerve Pathology in LAMA2 Muscular Dystrophy by AAV9-Mini-Agrin. *Molecular Therapy* 9:2018.
25. Meinen, S., Barzaghi, P., Lin, S., Lochmuller, H., Ruegg, M. A. Linker molecules between laminins and dystroglycan ameliorate laminin-alpha2-deficient muscular dystrophy at all disease stages. *The Journal of cell biology* 176:7. 2007.
26. Reinhard, J. R., Lin, S., McKee, K. K., Meinen, S., Crosson, S. C., Sury, M., Hobbs, S., Maier, G., Yurchenco, P. D., Ruegg, M. A. Linker proteins restore basement membrane and correct LAMA2-related muscular dystrophy in mice. *Science translational medicine* 9:396. 2017.
27. McKee, K. K., Crosson, S. C., Meinen, S., Reinhard, J. R., Ruegg, M. A., Yurchenco, P. D. Chimeric protein repair of laminin polymerization ameliorates muscular dystrophy phenotype. *The Journal of clinical investigation* 127:3. 2017.
28. Hightower, R. M., Alexander, M. S. Genetic modifiers of Duchenne and facioscapulohumeral muscular dystrophies. *Muscle & nerve* 57:1. 2018.
29. Rooney, J. E., Guppur, P. B., Burkin, D. J. Laminin-111 protein therapy prevents muscle disease in the mdx mouse model for Duchenne muscular dystrophy. *Proceedings of the National Academy of Sciences of the United States of America* 106:19. 2009.
30. Perrin, A., Rousseau, J., Tremblay, J. P. Increased Expression of Laminin Subunit Alpha 1 Chain by dCas9-VP160. *Molecular therapy. Nucleic acids* 6:2017.
31. Yuan, J., Ma, Y., Huang, T., Chen, Y., Peng, Y., Li, B., Li, J., Zhang, Y., Song, B., Sun, X., Ding, Q., Song, Y., Chang, X. Genetic Modulation of RNA Splicing with a CRISPR-Guided Cytidine Deaminase. *Molecular cell* 72:2. 2018.
32. Villiger, L., Grisch-Char, H. M., Lindsay, H., Ringnalda, F., Pogliano, C. B., Allegri, G., Fingerhut, R., Haberle, J., Matos, J., Robinson, M. D., Thony, B., Schwank, G. Treatment of a metabolic liver disease by in vivo genome base editing in adult mice. *Nature medicine* 24:10. 2018.
33. Bengtsson, N. E., Hall, J. K., Odom, G. L., Phelps, M. P., Andrus, C. R., Hawkins, R. D., Hauschka, S. D., Chamberlain, J. R., Chamberlain, J. S. Muscle-specific CRISPR/Cas9 dystrophin gene editing ameliorates pathophysiology in a mouse model for Duchenne muscular dystrophy. *Nature communications* 8:2017.
34. Nelson, C. E., Hakim, C. H., Ousterout, D. G., Thakore, P. I., Moreb, E. A., Rivera, R. M., Madhavan, S., Pan, X., Ran, F. A., Yan, W. X., Asokan, A., Zhang, F., Duan, D., Gersbach, C. A. In vivo genome editing improves muscle function in a mouse model of Duchenne muscular dystrophy. *Science* 2015.
35. Tabebordbar, M., Zhu, K., Cheng, J. K., Chew, W. L., Widrick, J. J., Yan, W. X., Maesner, C., Wu, E. Y., Xiao, R., Ran, F. A., Cong, L., Zhang, F., Vandenberghe, L. H., Church, G. M., Wagers, A. J. In vivo gene editing in dystrophic mouse muscle and muscle stem cells. *Science* 2015.
36. Long, C., Amoasii, L., Mireault, A. A., McAnally, J. R., Li, H., Sanchez-Ortiz, E., Bhattacharyya, S., Shelton, J. M., Bassel-Duby, R., Olson, E. N. Postnatal genome editing partially restores dystrophin expression in a mouse model of muscular dystrophy. *Science* 2015.

37. Thakore, P. I., D'Ippolito, A. M., Song, L., Safi, A., Shivakumar, N. K., Kabadi, A. M., Reddy, T. E., Crawford, G. E., Gersbach, C. A. Highly specific epigenome editing by CRISPR-Cas9 repressors for silencing of distal regulatory elements. *Nature methods* 12:12. 2015.
38. Lamar, K. M., Bogdanovich, S., Gardner, B. B., Gao, Q. Q., Miller, T., Earley, J. U., Hadhazy, M., Vo, A. H., Wren, L., Molkentin, J. D., McNally, E. M. Overexpression of Latent TGFbeta Binding Protein 4 in Muscle Ameliorates Muscular Dystrophy through Myostatin and TGFbeta. *PLoS genetics* 12:5. 2016.
39. Quattrocelli, M., Capote, J., Ohiri, J. C., Warner, J. L., Vo, A. H., Earley, J. U., Hadhazy, M., Demonbreun, A. R., Spencer, M. J., McNally, E. M. Genetic modifiers of muscular dystrophy act on sarcolemmal resealing and recovery from injury. *PLoS genetics* 13:10. 2017.
40. Kemaladewi, D. U., de Gorter, D. J., Aartsma-Rus, A., van Ommen, G. J., ten Dijke, P., t Hoen, P. A., Hoogaars, W. M. Cell-type specific regulation of myostatin signaling. *FASEB journal : official publication of the Federation of American Societies for Experimental Biology* 26:4. 2012.
41. Zhang, Y., Argaw, A. T., Gurfein, B. T., Zameer, A., Snyder, B. J., Ge, C., Lu, Q. R., Rowitch, D. H., Raine, C. S., Brosnan, C. F., John, G. R. Notch1 signaling plays a role in regulating precursor differentiation during CNS remyelination. *Proceedings of the National Academy of Sciences of the United States of America* 106:45. 2009.
42. Hakim, C. H., Wasala, N. B., Pan, X., Kodippili, K., Yue, Y., Zhang, K., Yao, G., Haffner, B., Duan, S. X., Ramos, J., Schneider, J. S., Yang, N. N., Chamberlain, J. S., Duan, D. A Five-Repeat Micro-Dystrophin Gene Ameliorated Dystrophic Phenotype in the Severe DBA/2J-mdx Model of Duchenne Muscular Dystrophy. *Molecular therapy. Methods & clinical development* 6:2017.
43. Kim, D., Langmead, B., Salzberg, S. L. HISAT: a fast spliced aligner with low memory requirements. *Nature methods* 12:4. 2015.
44. Anders, S., Pyl, P. T., Huber, W. HTSeq--a Python framework to work with high-throughput sequencing data. *Bioinformatics* 31:2. 2015.
45. Ritchie, M. E., Phipson, B., Wu, D., Hu, Y., Law, C. W., Shi, W., Smyth, G. K. limma powers differential expression analyses for RNA-sequencing and microarray studies. *Nucleic acids research* 43:7. 2015.
46. Bae, S., Park, J., Kim, J. S. Cas-OFFinder: a fast and versatile algorithm that searches for potential off-target sites of Cas9 RNA-guided endonucleases. *Bioinformatics* 30:10. 2014.

Extended data/supplementary figures

Figure S1

Fig. S1. SadCas9-2xVP64 enhances expression of minCMV-driven *tdTomato* in *vitro*. HEK293T was transfected with (a,b) a plasmid containing minCMV-driven *tdTomato* gene only, or (c,d) in combination with a plasmid containing SadCas9-2xVP64 and a sgRNA targeting the minCMV promoter. (b,d) Cells were imaged for *tdTomato* expression by fluorescent microscopy. Scale bar: 50 μ m.

Figure S2

Fig. S2. Expression of Lama1 and muscle architecture following early intervention in neonatal *dy^{2j}/dy^{2j}* mice. Lower magnification of immunofluorescence and H&E staining images of Fig. 3 in the main manuscript.

Figure S3

Fig. S3. Upregulation of Lama1 corresponds to improvement of muscle functions.

Three-weeks old dy^{2j}/dy^{2j} mice were injected systemically via tail vein with different doses of AAV9: 7.5×10^{10} , 1.5×10^{11} , or 3×10^{11} viral genome (vg)/gram of mouse. Two AAVs were necessary for the three guide cohort, therefore the total doses of virus injected was 1.5×10^{11} , 3×10^{11} , and 6×10^{11} viral genomes/gram of mouse. TA muscles isolated four weeks later were stained for Lama1 expression (**a**). Asterisks indicate Lama1-positive fibers in the low dose cohort. Scale bar: 100 μ m. *In vivo* contractile force assay (**b**) and open field test (**c**) performed in the end of the treatment regimen. Data are presented as average $\pm$ standard deviation. Statistical analysis was performed using one-way ANOVA. NS: not significant. ** $P < 0.01$.

Figure S4

Fig. S4. Representative images of Lama1-positive muscle sections

Mice were injected systemically with three guides at the age of three weeks old via tail vein, and sacrificed at the age of 11-12 weeks old. Muscles were stained for Lama1 expression (red). Scale bar: 500 μm.

Figure S5

Fig. S5. Expression of Laminin (LM) subunits in tibialis anterior muscles.

Expression of LM subunits in tibialis anterior from wild-type, dy^{2j}/dy^{2j} (intravenously injected with AAV carrying no guide or three guides) and $dy^{2j}/LM\alpha1$ (with transgenic overexpression of LM $\alpha1$) mice (n=3 for each group). Expression of LM $\alpha1$ chain in tibialis anterior is comparable between dy^{2j}/dy^{2j} AAV-three guide-treated mice and transgenic mice. No major differences in expression of LM $\alpha2$ and LM $\gamma1$ were detected between the groups. LM $\alpha4$ chain is upregulated in muscle from dy^{2j}/dy^{2j} AAV-no guide-treated mice and its expression is partially normalized in muscle from dy^{2j}/dy^{2j} AAV-three guide treated mice. Scale bar: 50 μ m.

Figure S6

Fig. S6. Expression of Laminin (LM) subunits in gastrocnemius muscles.

Expression of LM subunits in tibialis anterior from wild-type, dy^{2j}/dy^{2j} (intravenously injected with AAV carrying no guide or three guides) and $dy^{2j}/LM\alpha1$ (with transgenic overexpression of LM $\alpha1$) mice (n=3 for each group). Expression of LM $\alpha1$ chain in tibialis anterior is comparable between dy^{2j}/dy^{2j} AAV-three guide-treated mice and transgenic mice. No major differences in expression of LM $\alpha2$ and LM $\gamma1$ were detected between the groups. LM $\alpha4$ chain is upregulated in muscle from dy^{2j}/dy^{2j} AAV-no guide-treated mice and its expression is partially normalized in muscle from dy^{2j}/dy^{2j} AAV-three guide treated mice. Scale bar: 50 μ m.

Fig. S7. Myelination of sciatic nerves.

Electron microscopy images of sciatic nerves isolated from dy^{2j}/dy^{2j} mice injected with AAV carrying no guide or three guides (n=3 for each group). Myelin thickness was quantified using ImageJ and presented as average $\pm$ standard deviation. Statistical analysis was performed using Student's t -test. $*P < 0.05$. Scale bar: 5 μ m.

Figure S8

Fig. S8. Quantitative evaluation of AAV genome distribution.

Genomic DNA isolated from tibialis anterior muscle, sciatic nerve and liver of dy2j mice injected with AAV carrying three guides (n=8) was amplified for the presence of viral genome by qPCR. Data are presented as average $\pm$ SEM.

Figure S9

Fig. S9. Genome-wide analysis of gene expression of SadCas9-VP64-treated dy^{2j}/dy^{2j} mice.

Differential expression analysis derived from RNA-Sequencing results from the quadriceps of treated and untreated mice (**a-c**), comparing SadCas9-2xVP64 alone and SadCas9-2xVP64 with three guides targeting *Lama1* (**a**), untreated to SadCas9-2xVP64 alone (**b**), and untreated to SadCas9-2xVP64 with three guides targeting *Lama1* (**c**). Gene significantly differentially expressed, (FDR < 0.05), are coloured. Red data points indicate a log-fold change greater than one, while blue data points indicate a log-fold change less than one. Hierarchical clustering was performed on the normalized counts-per-million expression data (**d**).

Figure S10

Fig. S10. Top 100 differentially expressed genes.

Heatmap illustrating normalized log-CPM values for the top 100 genes differentially expressed in SadCas9-2xVP64 with three guide treated quadriceps versus untreated quadriceps. Red indicates higher expression while blue indicates lower expression.

Start 3X FLAG
M - **DYKDHDGDYKDHDIDYKDDDDK** - HGPR -
 VP64
DALDDFDLMLGSDALDDFDLMLGSDALDDFDLMLGSDALDDFDLDM -
 NLS
 VN - **PKKKRKV** - GIHGVPA -
 D10A
KRNYILGLA*IGITSVGYGIIDYETRDVIDAGVRLFKEANVENNEGRRSKRGARRL
KRRRRHRIQRVKLLFDYNLLTDHSELGINPYEARVKGLSQKLSEEFSAALLHLAK
RRGVHNVNEVEEDTGNELSTKEQISRNSKALEEKYVAELQLERLKKDGEVRGSINRF
KTSDYVKEAKQLLKVKAYHQLDQSFIDTYIDLLETRRTYYEGPGEKSPFGWKDIKE
WYEMLMGHCTYFPEELRSVKYAYNADLYNALNDLNNLVITRDENEKLEYEYEFQIEN
VFKQKKKPTLKQIAKEILVNEEDIKGYRVTSTGKPEFTNLKVYHDIKDITARKEIENAEL
LDQIAKILTIYQSSEDIQEELTNLNSLTQEEIEQISNLKGYTGTHNLSLKAINLILDELWH
TNDNQIAIFNRLKLVPKKVDLSQQKEIPTTLVDDFILSPVVKRSFIQSIKVINAIKKYGLP
NDIIELAREKNSKDAQKMINEMQKRNRQTNERIEEIIRTTGKENAKYLIEKIKLHDMQE
 N580A
GKCLYSLEAIPLEDLLNNPFNYEVDHIIIPRSVSFDNSFNKVLVKQEEA*SKKGNRTPF
QYLSSSDSKISYETFKKHILNLAAGKGRISKTKKEYLLEERDINRFSVQKDFINRNLVDT
RYATRGLMNLRSYFRVNNLDVKVKSINGGFTSFLRRKWKFKKERNKGYKHHAEADA
LIANADFIFKEWKLDKAKKVMENQMFEKQAESMPEIETEQEYKEIFITPHQIKHIKD
FKDYKYSHRVDKKPNRELINDTLYSTRKDDKGNTLIVNNLNGLYDKDNDKLLKLINKS
PEKLLMYHHPQTYQKLKLIMEQYGDEKNPLYKYYEETGNYLTKYSKKDNGPVIKKIK
YYGNKLNAHLDDTDDYPNSRNKVVKLSLKPYPFDVYLDNGVYKFVTVKNLDVIKKENY
YEVNSKCYEEAKLKKISNQAEFASFYNNDLIKINGELYRVIGVNNDLLNRIEVNMDIT
YREYLENMNDKRPPRIIKTIASKTQSIKKYSTDILGNLYEVKSKKHPQIIKKG -
 NLS
KRPAATKKAGQAKKKK - GS -
 VP64
DALDDFDLMLGSDALDDFDLMLGSDALDDFDLMLGSDALDDFDLML

Fig. S11. Protein encoded in the SadCas9-VP64 transcriptional activator plasmid.

Different domains are annotated accordingly. Amino acids encoding SadCas9 are in bold and italic, and the mutated residues D10A and N580A are indicated in red asterisk (*). NLS: Nuclear localization signal.

Supplemental video legends

Video S1. Phenotype of control dy^{2j}/dy^{2j} mouse following early intervention.

The dy^{2j}/dy^{2j} mouse was injected with AAV9 containing *SadCas9*-2xVP64 (no guide) at P2 (pre-symptomatic stage) via temporal vein and video was taken at the age of 7-week old. Hind limb paralysis, contracture and kyphosis resulting from lack of functional *Lama2* and compensatory *Lama1* are apparent.

Video S2. Phenotype of treated dy^{2j}/dy^{2j} mouse following early intervention.

The dy^{2j}/dy^{2j} mouse was injected with AAV9 containing *SadCas9*-2xVP64 and sgRNAs targeting *Lama1* proximal promoter (three guides) at P2 (pre-symptomatic stage) via temporal vein and video was taken at the age of 7-week old. Upregulation of compensatory *Lama1* expression ameliorates the hind limb paralysis, contracture and kyphosis.

Video S3. Phenotype of control dy^{2j}/dy^{2j} mouse following intervention at symptomatic stage.

The dy^{2j}/dy^{2j} mouse was injected with AAV9 containing *SadCas9*-2xVP64 (no guide) at 3-week old (pre-symptomatic stage) via tail vein. Video was taken at the age of 7-week old. Hind limb paralysis, contracture and kyphosis resulting from lack of functional *Lama2* and compensatory *Lama1* are apparent.

Video S4. Phenotype of treated dy^{2j}/dy^{2j} mouse following intervention at symptomatic stage.

The dy^{2j}/dy^{2j} mouse was injected with AAV9 containing *SadCas9*-2xVP64 and sgRNAs targeting *Lama1* proximal promoter (three guides) at 3-week old (pre-symptomatic stage) via tail vein. Video was taken at the age of 7-week old. Dystrophic features and disease progression were significantly improved and partially reversed following upregulation of *Lama1*.

Supplemental tables

Supplemental table 1. Top 100 genes differentially expressed in “SadCas9-2xVP64 + three guides” compared to “untreated”

Supplemental table 2. Top 100 genes differentially expressed in “SadCas9-2xVP64 + three guides” compared to “SadCas9-2xVP64 + no guide”

Supplemental table 3. Top 100 genes differentially expressed in “SadCas9-2xVP64 + no guide” compared to “untreated”

Supplemental table 4. Analysis showing distance from off-target site to differentially expressed gene body

Supplemental table 5. List of computationally predicted off-target sites

Supplemental table 6. Primers, plasmids and sgRNAs used in this study

Supplemental Table 1. Top 100 genes differentially expressed in "SadCas9-2xVP64 + three guides" compared to "untreated"

SYMBOL	TXCHROM	logFC	AveExpr	t	P.Value	adj.P.Val
Ces1c	chr8	8.06720501	-4.0858727	25.0852651	7.60E-11	1.10E-06
1810053823	chr16	10.5369617	-2.0993569	17.6475033	2.81E-09	6.76E-06
Fabp1	chr6	7.92790193	-3.3988185	18.7401814	1.44E-09	6.76E-06
1700003G13	chr9	7.85466761	-3.8132313	17.9773962	2.19E-09	6.76E-06
Cyp2c69	chr19	6.79884139	-4.4022125	17.5155103	2.77E-09	6.76E-06
Slc7a4	chr16	5.42020681	0.54904376	17.737618	2.38E-09	6.76E-06
Ttr	chr18	8.97781797	-3.6645933	17.0207698	3.88E-09	7.51E-06
Apoa2	chr1	5.40091384	-0.6005275	16.7716387	4.16E-09	7.51E-06
Lama1	chr17	8.98321051	0.34708087	15.7290019	8.52E-09	1.37E-05
ApoB	chr12	7.80234986	-3.4299405	15.0813327	1.26E-08	1.82E-05
Tat	chr8	5.3144494	-3.9814489	14.66332	1.58E-08	2.07E-05
Serpina1c	chr12	8.1709174	-3.2361681	14.3754788	2.04E-08	2.27E-05
Fam25c	chr14	7.39071119	-0.5378939	14.4492017	1.90E-08	2.27E-05
Serpina3k	chr12	9.29387622	-2.7989697	13.9984407	2.72E-08	2.81E-05
Igf1bp1	chr11	6.69308056	-4.2970164	13.6072358	3.38E-08	3.26E-05
Mup3	chr4	7.59035385	-2.0248159	13.5095555	3.70E-08	3.35E-05
Rdh7	chr10	6.12757345	-4.306269	12.9783192	5.31E-08	4.07E-05
Gys2	chr6	4.5777154	-1.5679355	13.0234776	5.00E-08	4.07E-05
Wnt4	chr4	4.20831956	5.03311767	12.9293649	5.35E-08	4.07E-05
Gc	chr5	8.24180382	-3.5843802	12.9381073	5.75E-08	4.07E-05
Klrg2	chr6	4.59908759	-1.1223193	12.8010591	5.92E-08	4.07E-05
Srrm4os	chr5	4.04588484	3.61138985	12.4826437	7.52E-08	4.94E-05
Mat1a	chr14	7.36113899	-3.7343781	12.3707009	8.71E-08	5.47E-05
Alb	chr5	9.586285	0.41592462	12.2174291	1.04E-07	6.01E-05
Serpina1a	chr12	6.95305368	-3.6425688	12.1347225	1.04E-07	6.01E-05
Azgp1	chr5	6.86136461	-4.0619138	11.9945852	1.16E-07	6.46E-05
Fgb	chr3	7.48821994	-3.1735567	11.6312338	1.59E-07	8.50E-05
Cidea	chr18	5.07510399	2.21493926	11.4830743	1.71E-07	8.82E-05
9030619P08	chr15	3.92341807	-0.2600478	11.3463138	1.89E-07	9.42E-05
Fgg	chr3	7.33395248	-3.4152783	11.1329778	2.41E-07	0.00011613
Mmd2	chr5	6.67105341	-3.3145461	11.0364562	2.58E-07	0.00012028
Cyp3a11	chr5	8.17360921	-3.6630995	10.8836274	3.06E-07	0.00013811
Slc27a2	chr2	6.6827	-3.1819203	10.7687412	3.26E-07	0.00014295
Ambp	chr4	6.13758126	-3.4506026	10.6633966	3.54E-07	0.00015057
Fga	chr3	7.62720297	-3.9076478	10.5588289	4.03E-07	0.00016636
Samd11	chr4	4.11178696	-0.4979967	10.4245456	4.25E-07	0.00017075
Fabp4	chr3	2.15462805	9.01447477	10.3115148	4.68E-07	0.00018279
Ighg1	chr12	8.46471745	-3.4389537	10.3557091	4.95E-07	0.00018754
Lctf	chr9	4.2237416	-0.3808795	10.2088981	5.19E-07	0.00018754
Pvalb	chr15	2.52928689	12.8626777	10.2161746	5.11E-07	0.00018754
Apoa5	chr9	6.60004476	-4.1263676	10.219239	5.36E-07	0.00018765
Vps13c	chr9	-2.1668504	3.212495	-10.146085	5.45E-07	0.00018765
Gm4841	chr18	3.86778458	5.35415478	9.78492273	7.72E-07	0.00025932
Prkar2b	chr12	3.97928004	3.13121334	9.7483776	8.00E-07	0.00026274
Pdpr	chr8	-2.203443	3.2904627	-9.4808172	1.03E-06	0.00033107
Hhip1	chr12	-2.7573871	2.31203806	-9.4137394	1.10E-06	0.00034623
Epn3	chr11	4.58955952	-1.5891827	9.3486758	1.19E-06	0.00036635
Aldh1a7	chr19	3.50528717	1.27190747	9.26652168	1.28E-06	0.00038529
Apoa1	chr9	8.39321713	-2.4434213	9.24069066	1.44E-06	0.00042593
Aspg	chr12	4.61360624	-0.6564861	9.09955804	1.53E-06	0.00044273
Adgrd1	chr5	-2.427791	3.44722048	-8.9858424	1.70E-06	0.00048062
Kng1	chr16	5.95942358	-3.7065193	8.99480646	1.75E-06	0.00048561
Ucp1	chr8	11.3963873	-1.5055582	9.07259957	1.85E-06	0.000504
Cps1	chr1	5.87067733	-3.3848414	8.82056311	2.09E-06	0.00055915
Sfrp4	chr13	-3.0424407	4.08490344	-8.7250934	2.23E-06	0.00058511
Jmjd1c	chr10	-3.2313535	1.84643433	-8.5191608	2.77E-06	0.00071562
Orm2	chr4	6.27368915	-2.5305119	8.54126641	2.83E-06	0.00071836
Atp6v1g1	chr4	1.90407218	8.11437097	8.46629447	2.93E-06	0.00072976
NA	NA	3.54327015	-0.8576183	8.37633903	3.24E-06	0.00079375
Elovl6	chr3	3.24419618	3.37792107	8.22580012	3.81E-06	0.0009186
Plg	chr17	4.98742747	-2.84041	8.16368792	4.16E-06	0.00098659
Pzp	chr6	6.39214719	-3.2228911	8.1148596	4.53E-06	0.00105615
Thrsp	chr7	3.26554164	6.03631914	7.85593444	5.77E-06	0.00131835
Gfpt2	chr11	-2.0366124	3.28844052	-7.8437333	5.84E-06	0.00131835
Bhmt	chr13	6.2546489	-3.2262881	7.78687551	6.55E-06	0.00145525
Apoc3	chr9	6.95422106	-2.1511746	7.78192704	6.70E-06	0.00146687
Proca1	chr11	2.63255929	1.56171029	7.68968505	6.97E-06	0.00150381
Hpd	chr5	6.45914605	-3.386459	7.69200292	7.34E-06	0.00156002
Pcsk6	chr7	-1.8922339	4.14311915	-7.6077764	7.67E-06	0.00160525
Dnajb13	chr7	5.86226504	-1.8392095	7.50871687	8.97E-06	0.0018516
NA	NA	-2.8570014	1.63865163	-7.4503139	9.23E-06	0.00187895
Myo5b	chr18	2.30425852	1.18853647	7.43398437	9.40E-06	0.00188746
Fam213b	chr4	2.52777314	7.1852214	7.33239481	1.06E-05	0.00210163
Aldob	chr4	6.06208644	-2.8991635	7.31447554	1.14E-05	0.0021888
Uox	chr3	5.46798424	-3.8910942	7.31419174	1.12E-05	0.0021888
NA	NA	-3.0351581	12.8448432	-7.2578878	1.16E-05	0.00221053
Mfrn	chr10	1.89226589	5.31305923	7.19570826	1.25E-05	0.00234855
Me1	chr9	1.9768026	6.91692293	7.13418269	1.35E-05	0.00243628
Agbl1	chr7	-2.0885962	1.3608529	-7.1465611	1.33E-05	0.00243628
NA	NA	-2.9421529	5.7414647	-7.1386369	1.34E-05	0.00243628
NA	NA	-2.8056988	10.2088761	-7.0774684	1.45E-05	0.00258145
Trpm1	chr7	2.96315703	0.3806044	7.01294075	1.57E-05	0.00276221
Cst6	chr19	2.23495652	4.66958372	7.00007046	1.59E-05	0.00276912
Igtp	chr11	1.86743947	4.54464325	6.89217296	1.82E-05	0.00312869
NA	NA	4.69385795	0.68222882	6.84250559	1.97E-05	0.00331558
Med12	chrX	-2.8614945	3.45721138	-6.8346408	1.96E-05	0.00331558
Serpina1d	chr12	6.32897767	-3.6602287	6.8549536	2.01E-05	0.00334534
NA	NA	4.3761592	4.3735629	6.78718488	2.11E-05	0.00342487
Kcnk3	chr5	3.81800932	1.86995833	6.77998928	2.11E-05	0.00342487
Car5b	chrX	2.17094632	2.52751196	6.69419221	2.34E-05	0.00374934
Aldh1a2	chr9	-3.9155062	1.81350194	-6.6857214	2.38E-05	0.0037799
Grem2	chr1	2.40181819	2.79352205	6.59957294	2.64E-05	0.00414096
Ces1d	chr8	4.10023518	3.35125626	6.59264986	2.69E-05	0.00417708
Ston1	chr17	-2.2562933	0.96729497	-6.573946	2.72E-05	0.00418839
Hpx	chr7	4.63301432	-1.3730015	6.56670755	2.80E-05	0.0042629
ApoH	chr11	2.91855783	-0.5038737	6.52074789	2.92E-05	0.00432487
Ces5a	chr8	2.60133348	0.55171618	6.51934336	2.93E-05	0.00432487
Dpp6	chr5	-2.9196681	0.2619482	-6.5178887	2.93E-05	0.00432487
NA	NA	-3.2449209	-0.3411066	-6.4875991	3.06E-05	0.00445983
Hist1h4i	chr13	1.82127787	3.91078129	6.41227049	3.36E-05	0.00485751

Supplemental Table 2. Top 100 genes differentially expressed in
"SadCas9-2xVP64 + three guides" compared to "SadCas9-2xVP64 no guide"

SYMBOL	TXCHROM	logFC	AveExpr	t	P.Value	adj.P.Val
Ces1c	chr8	7.68192528	-4.0858727	23.7177026	1.33E-10	1.92E-06
Tat	chr8	6.51413216	-3.9814489	18.7406265	1.40E-09	1.01E-05
Slc7a4	chr16	5.42395906	0.54904376	16.666776	4.43E-09	1.30E-05
Cyp2c69	chr19	6.41356165	-4.4022125	16.3517656	5.46E-09	1.30E-05
Fabp1	chr6	7.01430136	-3.3988185	16.2688646	5.82E-09	1.30E-05
1810053823	chr16	8.36567269	-2.0993569	16.2583139	6.04E-09	1.30E-05
Ttr	chr18	8.59253823	-3.6645933	16.1987709	6.30E-09	1.30E-05
Apoa2	chr1	4.31309488	-0.6005275	13.99838	2.46E-08	4.44E-05
1700003G13	chr9	6.16709101	-3.8132313	13.5514729	3.48E-08	5.59E-05
Rdh7	chr10	6.27061455	-4.306269	13.3403682	4.07E-08	5.88E-05
Apob	chr12	6.36042846	-3.4299405	11.8844821	1.26E-07	0.00015264
Alb	chr5	6.60124813	0.41592462	11.8383922	1.31E-07	0.00015264
Gc	chr5	7.61086889	-3.5843802	11.8108876	1.37E-07	0.00015264
Mup3	chr4	5.93703811	-2.0248159	11.5983687	1.58E-07	0.00016265
Igf1bp1	chr11	5.77947999	-4.2970164	11.4236063	1.82E-07	0.00017145
Serpina1a	chr12	6.56777394	-3.6425688	11.3493672	1.97E-07	0.00017145
Fam25c	chr14	4.42384018	-0.5378939	11.2759968	2.02E-07	0.00017145
Klrg2	chr6	4.01555522	-1.1223193	11.016482	2.51E-07	0.000195
Serpina1c	chr12	6.4833408	-3.2361681	10.992408	2.67E-07	0.000195
Fgg	chr3	7.23133838	-3.4152783	10.9526164	2.81E-07	0.000195
Fgb	chr3	7.10294021	-3.1735567	10.9405546	2.83E-07	0.000195
Klf1a	chr1	-3.2035885	1.72216362	-10.578571	3.68E-07	0.0002416
Lama1	chr17	3.60580481	0.34708087	10.3369068	4.59E-07	0.00028082
Serpina3k	chr12	6.46825197	-2.7989697	10.3674744	4.66E-07	0.00028082
Knq1	chr16	6.59377507	-3.7065193	10.145317	5.74E-07	0.00033198
Fga	chr3	7.24192323	-3.9076478	9.94497979	7.04E-07	0.00039142
Cidea	chr18	4.23425211	2.21493926	9.56079601	9.62E-07	0.00050767
Mat1a	chr14	5.91921759	-3.7343781	9.56655238	9.84E-07	0.00050767
Cyp3a11	chr5	7.26000864	-3.6630995	9.49753456	1.09E-06	0.00054116
Cadm4	chr7	-3.0854364	3.43404374	-9.3112336	1.22E-06	0.00058834
Slc27a2	chr2	5.77735647	-3.1819203	9.16142955	1.47E-06	0.00068446
Ucp1	chr8	11.5394284	-1.5055582	9.19742703	1.63E-06	0.00073752
Myo5b	chr18	2.70815964	1.18853647	8.91782918	1.82E-06	0.00079698
Mbp	chr18	-3.1241971	6.32564126	-8.796965	2.07E-06	0.00087745
Uox	chr3	6.38500137	-3.8910942	8.81536963	2.13E-06	0.00087745
NA	NA	7.30546168	-3.4389537	8.74748808	2.33E-06	0.00093426
Prx	chr7	-3.432919	5.10990804	-8.6589476	2.39E-06	0.00093426
Mpz	chr1	-3.1391767	7.72442679	-8.5709125	2.62E-06	0.00095901
Samd11	chr4	3.4319878	-0.4979967	8.53270285	2.74E-06	0.00095901
Apoa5	chr9	5.68644419	-4.1263676	8.55205544	2.76E-06	0.00095901
Azgp1	chr5	5.17378801	-4.0619138	8.54116051	2.77E-06	0.00095901
Ambp	chr4	5.00265745	-3.4506026	8.53232871	2.79E-06	0.00095901
Aspg	chr12	4.49864426	-0.6564861	8.45164468	3.02E-06	0.00101368
9030619P08	chr15	2.99612235	-0.2600478	8.36658783	3.27E-06	0.00105887
Sbsn	chr7	-2.6633714	0.90905622	-8.3572564	3.30E-06	0.00105887
Plg	chr17	5.52507799	-2.84041	8.36109263	3.39E-06	0.00106392
Cldn19	chr4	-3.2653338	2.7388015	-8.2964238	3.53E-06	0.00108534
Gys2	chr6	2.88423499	-1.5679355	8.17357339	4.04E-06	0.00121478
Ahsg	chr16	5.41354129	-0.9425466	8.11597943	4.43E-06	0.00130544
Wnt4	chr4	2.89310387	5.03311767	8.06192403	4.57E-06	0.00132035
Srrm4os	chr5	2.83746235	3.61138985	8.01069135	4.84E-06	0.00137073
Apoa1	chr9	6.68102512	-2.4434213	7.80253925	6.50E-06	0.0018055
Elovl6	chr3	3.14304008	3.37792107	7.70931392	6.83E-06	0.00186108
G6pc	chr11	5.60315183	-3.3951407	7.70806009	7.06E-06	0.00187982
Cps1	chr1	5.23974239	-3.3848414	7.6779702	7.26E-06	0.00187982
Aldob	chr4	6.32598423	-2.8991635	7.69579537	7.29E-06	0.00187982
Arg1	chr10	4.44362967	-1.6097834	7.59014941	7.93E-06	0.00201138
Epn3	chr11	3.69453485	-1.5891827	7.53325568	8.41E-06	0.00209514
Hba-a1	NA	-2.2700334	6.2456611	-7.4965888	8.73E-06	0.00213896
Ddn	chr15	-3.398565	0.55130958	-7.2073462	1.24E-05	0.00298201
Fa2h	chr8	-3.2130863	1.20608522	-7.032031	1.53E-05	0.00363058
Cadm3	chr1	-2.051734	3.75756436	-6.9210036	1.75E-05	0.00403033
Ppp1r3b	chr8	2.57692007	0.40173032	6.92011989	1.76E-05	0.00403033
Pzp	chr6	5.50500956	-3.2228911	6.84788224	1.99E-05	0.00449761
Nfasc	chr1	-2.6849138	1.86436923	-6.7931454	2.06E-05	0.00458172
Orm2	chr4	4.38077136	-2.5305119	6.70807897	2.33E-05	0.00506111
Sox10	chr15	-3.2987824	1.71005186	-6.6733195	2.41E-05	0.00506111
Hpx	chr7	5.41271902	-1.3730015	6.697118	2.41E-05	0.00506111
Prkar2b	chr12	2.93206413	3.13121334	6.65445212	2.46E-05	0.00506111
Bmp7	chr2	-2.8365493	-0.1703501	-6.65022	2.47E-05	0.00506111
Ces5a	chr8	2.72830868	0.55171618	6.64349268	2.49E-05	0.00506111
Serpina1d	chr12	6.18935314	-3.6602287	6.67534697	2.52E-05	0.00506111
Dnajb13	chr7	4.93453349	-1.8392095	6.49479192	3.09E-05	0.00612482
Serpinc1	chr1	3.93724606	-1.3533652	6.28356394	4.02E-05	0.0078515
Dact2	chr17	-2.5238997	0.9490797	-6.1621573	4.69E-05	0.00902999
NA	NA	2.77363083	-0.8576183	6.1509308	4.76E-05	0.0090517
Hpd	chr5	5.36870724	-3.386459	6.15556143	4.90E-05	0.0091922
Acsn3	chr7	4.79325628	-0.8979803	6.08513137	5.32E-05	0.00985906
Dusp15	chr2	-2.832713	1.31355423	-5.9744195	6.05E-05	0.0110674
Mag	chr7	-2.4835744	1.72185278	-5.8808295	6.88E-05	0.01242024
2310008N11	chr8	2.47762951	0.0223978	5.78103716	7.90E-05	0.0140903
Aph1c	chr9	2.02542717	1.4446743	5.69597944	8.90E-05	0.01567068
Alas2	chrX	-2.7381939	2.63862482	-5.6882468	9.00E-05	0.01567068
Aldh1a2	chr9	-3.5045852	1.81350194	-5.619936	9.96E-05	0.01713352
Rasgef1c	chr11	-2.7268056	-0.4788461	-5.5831253	0.00010446	0.01760109
Slc9b2	chr3	2.60867469	0.03741035	5.58003047	0.00010488	0.01760109
Sptbn5	NA	-3.1073231	2.1100215	-5.5741903	0.00010597	0.01760109
Aph1	chr11	2.6612831	-0.5038737	5.5623464	0.00010758	0.01766438
Plip	chr8	-2.6544819	2.15418714	-5.5538324	0.00010889	0.01767908
Mgat3	chr15	-2.522372	1.43835928	-5.538076	0.00011133	0.01787536
Spn	chr4	-2.0266173	3.73200949	-5.4989035	0.0001177	0.01857554
Mup11	chr4	6.5359933	-0.3747465	5.54577617	0.00011827	0.01857554
Hbb-bt	NA	-2.1214318	4.13274184	-5.4147076	0.00013289	0.02043237
Klf19a	chr11	-2.4525954	1.56428869	-5.414812	0.00013292	0.02043237
Hepacam	chr9	-3.1235403	-0.7331738	-5.3921781	0.00013769	0.02094399
9130230L23	chr5	2.99937564	-1.7029617	5.3379434	0.00014889	0.0224111
Apo3	chr9	3.89375921	-2.1511746	5.21346237	0.00018033	0.02686402
Scube1	chr15	-2.61022	0.15037746	-5.1910997	0.00018445	0.02719675
Gal3st1	chr11	-2.7637327	-0.2060749	-5.1771827	0.00018838	0.02749575
Gm4841	chr18	2.44516047	5.35415478	5.13981561	0.00019895	0.02874825

Supplemental Table 3. Top 100 genes differentially expressed in
SadCas9-2xVP64-no guide compared to "untreated"

SYMBOL	TXCHROM	logFC	AveExpr	t	P.Value	adj.P.Val
Hba-a1	NA	3.12223892	6.2456611	12.331034	8.43E-08	0.00121768
Mpz	chr1	3.67608141	7.72442679	10.6882893	3.34E-07	0.00241497
Hbb-bt	NA	3.16484723	4.13274184	9.89369815	6.93E-07	0.00244925
Hba-a2	chr11	3.23794544	4.91567042	9.85619618	7.18E-07	0.00244925
Cadm4	chr7	3.13536782	3.43404374	9.68323067	8.47E-07	0.00244925
Mbp	chr18	3.22961817	6.32564126	9.26281665	1.28E-06	0.00308948
Lama1	chr17	5.3774057	0.34708087	8.31143772	3.57E-06	0.0073592
Hbb-bs	chr7	2.8634898	6.55027296	8.1395833	4.19E-06	0.00756795
Prx	chr7	3.18266127	5.10990804	7.96374203	5.11E-06	0.00765789
Cldn19	chr4	3.06808525	2.7388015	7.89212771	5.53E-06	0.00765789
Tlr13	chrX	-2.5987846	1.26055872	-7.8376704	5.88E-06	0.00765789
1700001O22Rik	chr2	3.76078395	-0.8282985	7.77422963	6.36E-06	0.00765789
Ccl2	chr11	-2.7593297	1.1058906	-7.6753655	7.09E-06	0.00788188
Mx1	chr16	-3.4609353	0.74527914	-7.5588781	8.15E-06	0.00804522
Fa2h	chr8	3.32272314	1.20608522	7.53694641	8.35E-06	0.00804522
CSar1	chr7	-1.8891035	2.10571941	-7.258534	1.16E-05	0.01013989
Klf1a	chr1	2.32436318	1.72216362	7.23538702	1.19E-05	0.01013989
Fam178b	chr1	3.0042875	1.76045364	7.10719561	1.40E-05	0.01120779
Ddn	chr15	3.14993822	0.55130958	6.92964353	1.74E-05	0.01322915
Lctl	chr9	3.26770545	-0.3808795	6.88735709	1.83E-05	0.01325713
Plip	chr8	3.06580561	2.15418714	6.82876119	1.97E-05	0.01357527
Lrrc15	chr16	-2.4883765	0.74755506	-6.7582519	2.15E-05	0.01414289
NA	NA	-1.9501553	3.38202652	-6.6526	2.46E-05	0.01547083
Alas2	chrX	2.9899336	2.63862482	6.51840873	2.93E-05	0.01765268
Xist	chrX	-3.2767212	6.77693285	-6.4385364	3.26E-05	0.01864379
Ncmmap	chr4	3.08130356	2.86901463	6.39750506	3.43E-05	0.01864379
Gjb1	chrX	3.46954089	0.2018106	6.38853084	3.48E-05	0.01864379
Mag	chr7	2.56287891	1.72185278	6.33342043	3.73E-05	0.01922711
SncA	chr6	3.12063457	0.87022355	6.30932249	3.86E-05	0.01922711
Ilrl1	chr1	-2.5405179	0.79307145	-6.2535459	4.15E-05	0.01968275
Pvalb	chr15	1.9371664	12.8626777	6.2397791	4.22E-05	0.01968275
Dusp15	chr2	2.72326203	1.31355423	5.89258602	6.77E-05	0.02962619
Adam12	chr7	-2.0037212	2.41708454	-5.8606882	7.07E-05	0.02962619
Ccl7	chr11	-2.540678	0.66322051	-5.838118	7.30E-05	0.02962619
Cd300lb	chr11	-2.4326089	0.71920982	-5.8322784	7.35E-05	0.02962619
Bmp7	chr2	2.48796642	-0.1703501	5.82971426	7.38E-05	0.02962619
Arg1	chr10	-3.6796601	-1.6097834	-5.8042644	7.70E-05	0.03006634
Sfrp1	chr8	-1.9250563	3.06171827	-5.6945804	8.91E-05	0.03389067
Aoah	chr13	-2.230976	0.59804822	-5.5991203	0.00010202	0.0377995
Msr1	chr8	-2.5214491	1.6301269	-5.5421659	0.00011069	0.03942958
Cmtm5	chr14	2.84161366	-0.3319268	5.5243583	0.00011364	0.03942958
Clec4d	chr6	-2.2426926	-0.1450714	-5.5175924	0.0001146	0.03942958
Mmd2	chr5	4.10127424	-3.3145461	5.35972024	0.00014594	0.04854376
Gal3st1	chr11	2.75795131	-0.2060749	5.34218678	0.00014781	0.04854376
Sbspon	chr1	2.22808049	0.44090614	5.22117943	0.00017632	0.05546403
Adams6	chr13	-2.4536128	1.59657982	-5.2204485	0.00017656	0.05546403
Hhip1	chr12	-1.9118394	2.31203806	-5.2007135	0.00018171	0.05586471
Hepacam	chr9	2.91504751	-0.7331738	5.16779439	0.00019114	0.0574251
F2rl1	chr13	-2.2231625	1.71289475	-5.1068931	0.00020888	0.06109392
Gm42067	NA	1.85312529	2.04012111	5.09877039	0.0002114	0.06109392
Adcy7	chr8	-1.881753	4.6834493	-5.0461447	0.00022871	0.06480185
Klf19a	chr11	2.27988907	1.56428869	5.02136549	0.00023743	0.06597873
Tlr8	chrX	-4.0365645	-1.2570211	-4.9449509	0.00027029	0.07369187
Smox	chr2	1.81483923	5.4987099	4.89994085	0.00028521	0.07513181
C3ar1	chr6	-3.095913	2.52727176	-4.897357	0.00028728	0.07513181
Eps8l1	chr7	-2.4924048	2.20261789	-4.8867435	0.00029117	0.07513181
Adgre1	chr17	-1.5575189	3.45468859	-4.8714084	0.00029787	0.07551336
Sfrp4	chr13	-2.0811203	0.08490344	-4.8359387	0.00031448	0.07742878
Sfrn4	chr11	-2.7773743	-1.1861611	-4.8334332	0.00031615	0.07742878
Lilr4b	chr10	-3.0117057	2.19904976	-4.7560936	0.00035649	0.08523435
NA	NA	2.2188592	1.46909467	4.74477333	0.00036179	0.08523435
Dupd1	chr14	1.83532553	6.3387283	4.73767715	0.00036571	0.08523435
Cdh1	chr8	2.69435584	0.15753197	4.7105409	0.00038186	0.08758428
Ctbp100	chr6	2.68467385	-0.1085207	4.65308887	0.00041746	0.09425372
Chil3	chr3	-4.4041995	-0.8908455	-4.6368929	0.00043783	0.09733231
NA	NA	-2.032801	1.63865163	-4.5516428	0.00048859	0.10697147
Ms4a7	chr19	-2.0914278	2.7993614	-4.4397996	0.0005829	0.12571458
Pmp22	chr11	1.97966029	7.02265612	4.41974373	0.00060171	0.12720169
Sox10	chr15	2.30301682	1.71005186	4.41400578	0.0006074	0.12720169
Tnc	chr4	-2.1869894	6.29769631	-4.323846	0.00070125	0.14475757
Dclre1c	chr2	-1.798468	1.83339495	-4.2666147	0.00076856	0.15522687
Ccl6	chr11	-1.8144934	4.30576424	-4.2626793	0.00077345	0.15522687
A930016O22Rik	chr7	1.77831878	7.34616069	4.2491595	0.00079047	0.15646948
Spire2	chr8	2.46586051	-0.8626346	4.23290116	0.00081207	0.15857376
Pdpr	chr8	-1.5137564	3.29044627	-4.2177999	0.00083148	0.1601984
Ms4a4a	chr19	-3.3012903	2.04305875	-4.2086143	0.0008496	0.16153672
Retnla	chr16	1.70702394	3.83252031	4.18081523	0.00088276	0.16566021
Igsf6	chr7	-2.1161057	1.14725297	-4.1698631	0.00089871	0.16649164
Bmper	chr9	-2.3781522	3.01926601	-4.1196582	0.00097551	0.17808689
Kcnmb4	chr10	2.06573485	-0.0707184	4.1071686	0.00099511	0.17808689
Cenpi	chrX	-3.2736233	-0.9010077	-4.1092747	0.00099827	0.17808689
Tfec	chr6	-2.4975075	-0.9890424	-4.0905242	0.00102332	0.18032955
Ccr1	chr9	-3.5459938	0.74382016	-4.0436169	0.00111601	0.19199831
Serpine1	chr5	-1.6929341	4.20045863	-4.0366477	0.00111654	0.19199831
G530011O06Rik	chrX	-3.0056326	0.28110767	-4.0255271	0.00114176	0.19199831
Ptger2	chr14	-2.2297542	-0.7377989	-4.0227253	0.00114269	0.19199831
Lilrb4a	chr10	-4.0994493	0.16994375	-3.9912293	0.00123047	0.20437115
Rnf180	chr13	-1.9538557	-0.0378799	-3.9618891	0.00126259	0.20522254
Ccl12	chr11	-2.0439673	-0.1216011	-3.9612473	0.001264	0.20522254
Jmjd1c	chr10	-1.8859772	1.84643433	-3.942432	0.00130374	0.20875585
Myh4	chr11	1.82246949	13.9035537	3.93272021	0.00132479	0.20875585
Cd33	chr7	-2.5021255	1.96787566	-3.931379	0.0013291	0.20875585
Fam25c	chr14	2.96687101	-0.5378939	3.90989754	0.00138126	0.21461452
Ccr5	chr9	-3.1584851	1.7062355	-3.9018754	0.0014026	0.21561211
Sostdc1	chr12	2.54871926	-0.5067107	3.88711315	0.00143042	0.21757481
Ccnb1	chr13	-2.1828154	0.91827463	-3.8617101	0.0014905	0.22309683
Nuf2	chr1	-2.3761242	0.99193462	-3.8590906	0.00149761	0.22309683
Fgf16	chrX	-2.4858276	0.08167528	-3.850861	0.00151877	0.22347127
Naip6	chr13	-3.0919726	-0.73325	-3.8332285	0.00157101	0.22347127

Supplemental Table 4. Analysis showing distance from off-target site to differentially expressed gene body

SYMBOL	TXCHROM	logFC	adj.P.Val	Gene start (bp)	Gene end (bp)	Off-Target Sequence	Chromosome	Site (bp)	to Gene (bp)	matches	Strand	Lama1 Guide
Ces1c	chr8	8.06720501	1.10E-06	93099015	93131283	aggggGCaGgGaGCaGCGaGAGGCGCTGAGT	chr8	90241651	2857364	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Slc7a4	chr16	5.4202681	6.76E-06	17572018	17583214	tgCaAAGaGatTctGtCCaGAGGAGGAAT	chr16	8237459	9345359	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Fabp1	chr6	7.92790193	6.76E-06	71199827	71205023	CagCtGcGagCTGtGtCgCAGGCTGGAAT	chr6	72989175	1784152	8	-	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
1700003G13Rik	chr9	7.85466761	6.76E-06	45318265	45322079	ataCaAaGcGCTaGtGtCgCAGGCTGAGAT	chr9	43733430	1584835	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Cyp2c69	chr19	6.79884139	6.76E-06	39842660	39867668	gACgGCaGgGcCtGtGtGtGtGAGGCGGAT	chr19	40318577	413808	9	-	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
1810053B23Rik	chr16	10.5369617	6.76E-06	9342504	93366668	CACtAGaTtCctGtGtCAGGCTGGGAT	chr16	93058037	284467	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Apoa2	chr1	5.40091384	7.51E-06	171225054	171226379	tgGCaCgGGGCaGCGggaGAGGCTAGAT	chr1	171081055	143999	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Ttr	chr18	8.97781797	7.51E-06	20665250	20674324	AtgGCaGcagGCaGgCAGGCGAGGAAGAT	chr18	24331496	3657172	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Lama1	chr17	8.98321051	1.37E-05	67697265	67822645	N/A	N/A	N/A	N/A	N/A	N/A	N/A
ApoB	chr12	7.80234986	1.82E-05	7977648	8016835	CAggcgGcgagGCaGgCAGGCGAGGCTGGGT	chr12	8858012	841177	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Tat	chr8	5.3144494	2.07E-05	109990437	109999803	aACcttGgGcCacTctGtCtCAGGCGCTGAGT	chr8	110878472	878669	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Fam25c	chr14	7.39071119	2.27E-05	34351881	34355433	aACtGACCCCAgtGgGtGgGAGGCTGGGAT	chr14	32562154	1789727	8	+	CACCGACCCCACTGGAGTGAGGCTNNNNN
Serpina1c	chr12	8.17091474	2.27E-05	103894926	103904887	ttCacActtgAAtTGAAGgGAGGCTGGGGT	chr12	105870943	1966056	9	+	CACCGACCCCACTGGAGTGAGGCTNNNNN
Serpina3k	chr12	9.29387622	2.81E-05	104338486	104346144	ttCacActtgAAtTGAAGgGAGGCTGGGGT	chr12	105870943	1524799	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Igf1bp1	chr11	6.69308056	3.26E-05	7197782	7202546	cttttAAAGGgATgAGcCtCAGGCTGGGAT	chr11	7421018	218472	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Mup3	chr4	7.59035385	3.35E-05	62083476	62087342	CAgaGAAGGGCTgaactcCAGGCTAGGAT	chr4	63232225	1144883	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Gys2	chr6	4.5777154	4.07E-05	142422613	142473109	CAggtAATGtTtGtACGACCGAGCTGGAAT	chr6	143140787	667678	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Wnt4	chr4	4.20831956	4.07E-05	137277489	137299726	tcCTtggGgGcCaGcCtCAGGCTGGGAT	chr4	137673872	374146	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Rdh7	chr10	6.12757345	4.07E-05	127884028	127888817	ccCGCAACagcTCaGcCtCAGGCTGGGAT	chr10	128913811	1024994	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Gc	chr5	8.24180382	4.07E-05	89417522	89457898	CAgggttTGtGgGgGcCaGcCtCAGGAGGAAT	chr5	101591226	12133328	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Klrf2	chr6	4.59908759	4.07E-05	38625693	38637242	CggggcGtGtGcGgGgCAGGCGAGGCGGGGT	chr6	38551799	73894	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Srrm4os	chr5	4.04588484	4.94E-05	116438720	116465487	CcCaGACtCtAtgTGAAGgGAGGCTGAGAT	chr5	116290065	148655	8	-	CACCGACCCCACTGGAGTGAGGCTNNNNN
Matta	chr14	7.36113899	5.47E-05	41105035	41124412	gACCaCGGGCtAtGtCAGGCGAGGAGGAT	chr14	41192606	68194	7	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Alb	chr5	9.586285	6.01E-05	90460897	90476602	CAgggttTGtGgGgGcCaGcCtCAGGAGGAAT	chr5	101591226	11114624	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Serpina1a	chr12	6.95305368	6.01E-05	103853589	103863562	ttCacActtgAAtTGAAGgGAGGCTGGGGT	chr12	105870943	2007381	9	+	CACCGACCCCACTGGAGTGAGGCTNNNNN
Azgp1	chr5	6.86136461	6.46E-05	137981520	137990233	CAgtAATGtGtGtGtGtCAGGAGGCTGGAAT	chr5	139375397	1385164	8	-	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Fgb	chr3	7.48821994	8.50E-05	83040141	83049863	CAcCagctGagTCaCaCtCAGGCGAGAAT	chr3	87805463	4755600	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Cidea	chr18	5.07510399	8.82E-05	67343564	67367794	CAcTtAGtGtGtGtGtGtGtGtGGGGT	chr18	69341102	1973308	9	+	CACCGACCCCACTGGAGTGAGGCTNNNNN
9030619P08Rik	chr15	9.92341807	9.42E-05	75427606	75431829	aACtAGaGCaGgGgGtGtCAGGCGAGGAAT	chr15	76107757	675928	8	-	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Fgg	chr3	7.33395248	0.00011613	83007724	83015049	CAcCagctGagTCaCaCtCAGGCGAGGAAT	chr3	87805463	4790414	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Mmd2	chr5	6.67105341	0.00012028	142562358	142608800	cttTGaAGGAGGAGGgaaACACGCGCTGGAT	chr5	142010113	552245	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Cyp3a11	chr5	8.17360921	0.00013811	145854426	145879964	aggCGGGGgagTGAAGCAGGCGAGGATAGAT	chr5	145646342	208084	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Slc27a2	chr2	6.6827	0.00014295	126552407	126588243	cttCAACtCacTcAGgGgGAGGCTGGGGT	chr2	126819405	231162	9	+	CACCGACCCCACTGGAGTGAGGCTNNNNN
Ambp	chr4	6.13758126	0.00015057	63143275	63154799	CAggaGAAGGGCTgaactcCAGGCTGGGAT	chr4	63232225	77426	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Fga	chr3	7.62720297	0.00016636	83026076	83033627	CAcCagctGagTcAaCtCtCAGGCGAGAAT	chr3	87805463	4771836	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Samd11	chr4	4.11178696	0.00017075	156246966	156255657	CACAGCagTgGaaaaaCAAGGCGAGGAGGAT	chr4	155503906	743060	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Fabp4	chr3	2.15462805	0.00018279	10204088	10208576	cttGCaAaGcCtGtGtGtCAGGCTCGAT	chr3	10144068	60020	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Pvalb	chr15	2.52928689	0.00018754	78191114	78206400	cttCtAaCtTAgTCTGAGCgAGGCTGGGAAT	chr15	78664606	458206	9	+	CACCGACCCCACTGGAGTGAGGCTNNNNN
Lct1	chr9	4.2237416	0.00018754	64117147	64138118	aACtGtGAGcCacTctGtCtCAGGCTGGAT	chr9	64028355	88792	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
NA	NA	8.46471745	0.00018754	113325240	113330523	CgCtAGtAGtGatCaaGACACGAGTGAAT	chr12	118303251	4972728	8	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Apoa5	chr9	6.60004476	0.00018765	46268633	46271919	aACaaAGGCGCTGgGaACAGGCGCAGGAT	chr9	46913410	641491	6	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Vps13c	chr9	2.1668504	0.00018765	67840396	67995638	aACCGtGAGGcCacTctCtCAGGCTGGGT	chr9	69433414	1437776	8	-	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Kom4841	chr18	3.86778458	0.00025932	60268301	60273267	AtaCGtAaagGCTGtCtCtCAGGCTAGGAT	chr18	61821232	1547965	7	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Pkr2b	chr12	3.97928004	0.00026274	31958476	32061296	acCtttGgGgGgGgGgGgGgGgGgGgGAT	chr12	30913970	1044506	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Pdpr	chr8	-2.203443	0.00033107	111094630	111137074	aACtCtGtGcCacTctGtCtCAGGCTGAGT	chr8	110878472	216158	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Hhsp11	chr12	-2.7573871	0.00034623	108306270	108330869	tgaCTGGtGtGtCaGAGGCGAGGCTAGGAT	chr12	106527437	1778833	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Epn3	chr11	4.58955952	0.00036635	94489599	94499974	CACaCaCaCaCaGtAGaGAGGCTTGGGGT	chr11	95563937	1063963	8	-	CACCGACCCCACTGGAGTGAGGCTNNNNN
Aldh1a7	chr19	3.50528717	0.00038529	20629253	20727562	attCAGGtGGGCTcagGtCtCAGGCGAGGAT	chr19	17118070	3574883	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Apoa1	chr9	8.39321713	0.00042593	46228580	46230466	aACaaAGGCGCTGgGaACAGGCGCAGGAT	chr9	46913410	682944	6	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Aspg	chr12	4.61360624	0.0004273	112106679	112127559	cttCGtGGtGtCaGAGGCGAGGCTAGGAT	chr12	106527437	5579242	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Adgrd1	chr5	-2.427791	0.00048062	129096750	129204599	CAgCaATGcCtCaGtCtCAGGCTGGGGT	chr5	128632023	464727	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Kng1	chr16	5.95942358	0.00048561	23057865	23082075	gACtAGtGtGtGtGtGtGtGtGtGtGAT	chr16	29714613	6632535	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Ucp1	chr8	11.3963873	0.000504	129330352	12938455	gggtGAGtCCaCtCTaAGTGGAGGCTAAGAT	chr8	80336949	2953403	8	-	CACCGACCCCACTGGAGTGAGGCTNNNNN
Cps1	chr1	5.87067723	0.00055915	67123026	67231259	CAtgTGgGgGgGtGtGtGtGtGtGtGAT	chr1	74579710	7348451	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Sfrp4	chr13	-3.0424407	0.00058511	19623104	19632994	gACaaAAGtGtCtGtggCAGGCTAGGAT	chr13	20914228	1281234	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Jmjd1c	chr10	-3.2313535	0.00071562	67096125	67256322	tggaagCtGtGtGtCtGgCAGGCGAGGCTGGAT	chr10	70037110	2780784	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Orm2	chr4	6.27368915	0.00071836	63362449	63365878	CAgaGAAGGGCTgaactcCAGGCTAGGAT	chr4	63232225	130224	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
AtPv1g1	chr4	1.90407218	0.00072976	63544772	63550750	CAggaGAAGGGCTgaactcCAGGCTAGGAT	chr4	63232225	312547	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
NA	NA	3.54327015	0.00079375	21753529	21800773	CAtgTgTGGGgGCaGtGgGAGGCGAGGAAT	chr3	19951663	1801866	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Elovl6	chr17	3.24419618	0.00091886	129532355	129638493	cttCGtGtGtGtGtGtGtGtGtGtGAT	chr17	130060315	421820	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Plg	chr3	4.98742747	0.00098659	12378609	12414384	cttCCagAGaGaaTgGgAaCAGGCGAGGAAT	chr3	11877092	501517	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Gp2	chr6	6.39214719	0.00103185	128483567	128526720	gACaaaGGGtGtGtGtGtGtGtGtGtGtGAT	chr6	122781164	5702403	8	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
FtFt12	chr11	-2.0366214	0.00113135	49794178	49838613	gAtCtGtGtTCTGtGtACCAAGGCTAGGAT	chr11	48443528	1350650	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Thrsp	chr7	3.26554164	0.00113135	97412938	97417730	CActagTgGgGcagGaaCAGGCGAGGCTGGGT	chr7	98123842	706112	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Bhmt	chr13	6.2546489	0.00145525	93616675	93637961	ccCtCAaGtGgaGtGgaGAGGCGAGGAT	chr13	93929752	291791	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
ApoC3	chr9	6.95422106	0.00146687	46232933	46235636	aACaaAGGCGCTGgGaACAGGCGCAGGAT	chr9	46913410	677774	6	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Proca1	chr11	2.63255929	0.00150381	78193392	78205763	tgtaaaAAGGCaTgTGtGtGtGtGtGtGAT	chr11	80531260	2325497	8	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Hpd	chr5	6.45914605	0.00156002	123171807	123182727	tcCtCAGgGtGtCtGtGtACAGGCTAGGAT	chr5	120699033	2472774	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Pcsk6	chr7	-1.8923239	0.00160525	58661734	6050386	tAggGgAaTgCtCaGcCtCAGGCGAGGAT	chr7	18273824	2073438	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Dnajb13	chr7	5.86226504	0.0018516	100501716	100514960	aAGaGAGaGcCCTGtggCAGGCGTGGAT	chr7	101044600	529640	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
NA	NA	-2.8570014	0.00187895	59672814	59676700	CAggcagGcgagGCaGgCAGGCGAGGATGAT	chr9	60247801	571101	9	+	CACCGCGGGGCGCGCAGGCGAGGCGNNNNN
Myo5b	chr18	2.30425852	0.00188746	74440936	74771493	attCtGAcAGGtGtGtGtGtCAGGCGGGGT	chr18	75554852	783359	9	+	CACCGAAGGGCTGCGCAGCAGGCGNNNNN
Fam213b	chr4	2.52777314										

Supplemental Table 5. List of computationally predicted off-target sites

Number	3'UTR Guide	Chromosome	Off-Target Locus	Off-Target Sequence	Strand	Mismatches
1	CACGCCCACTGAGTGGAGGTNNNNNN	chr1	16312174	CCAGCTCggtgTgAGTGGAGGGTGGGAAT	+	9
2	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	21512452	CACAGTgGacatTCGtCCAGGCTCGAGT	-	9
3	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	38624935	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
4	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	52143894	gTtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
5	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	55058127	CTCAGAGGtGacatTCAGCAGGCTAGT	+	9
6	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	57261402	gggCAAGGCTCTGCTCCAGGACAGAGT	-	8
7	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	57934949	CAGtGatGatGatTCtCAAGCAGGACAGGAT	-	8
8	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	57934949	CAGtGatGatGatTCtCAAGCAGGACAGGAT	-	8
9	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	57934626	CAGtGatGatGatTCtCAAGCAGGACAGGAT	-	8
10	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	57979710	CAGtGatGatGatTCtCAAGCAGGCTGGGAT	+	9
11	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	82782223	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	9
12	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	87961823	gTtCAAGGCTCggtgTgAGCAGGATAGGT	+	9
13	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	90031346	gggCAAGGCTCTGCTCCAGGACAGAGT	-	9
14	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	91995313	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	9
15	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	92255646	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
16	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	109694154	gggCAAGGCTCTGCTCCAGGACAGAGT	-	9
17	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	126488448	CAGtGatGatGatTCtCAAGCAGGACAGGAT	-	9
18	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	134277663	CTtGtAGGCTCTGCTCCAGGACAGAGT	-	9
19	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	135011972	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
20	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	141154277	CAGtGatGatGatTCtCAAGCAGGACAGGAT	-	8
21	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	150717933	gggCAAGGCTCTGCTCCAGGACAGAGT	-	9
22	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	157191498	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	9
23	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	159690338	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	9
24	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	161901332	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	9
25	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	16320275	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	9
26	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	171065806	gggCAAGGCTCTGCTCCAGGACAGAGT	+	9
27	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	171073441	gggCAAGGCTCTGCTCCAGGACAGAGT	+	9
28	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	171081075	gggCAAGGCTCTGCTCCAGGACAGAGT	+	9
29	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	17969166	gggCAAGGCTCTGCTCCAGGACAGAGT	-	9
30	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	179800672	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	8
31	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	180373924	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
32	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	180487362	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
33	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	181119150	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	9
34	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	182612878	gggCAAGGCTCTGCTCCAGGACAGAGT	-	9
35	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	182861831	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
36	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	185426468	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
37	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr1	185917332	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
38	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	6479941	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
39	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	174417051	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
40	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	25425905	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
41	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	25995977	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
42	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	28820200	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
43	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	31672867	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	8
44	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	32697101	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	8
45	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	33373871	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
46	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	33490722	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
47	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	33501232	CACCGAAGGCTCTGCTCCAGGACAGAGT	+	8
48	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	33951091	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	9
49	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	34571771	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
50	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	47931225	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
51	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	49708239	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
52	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	57243174	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	9
53	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	70241763	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
54	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	73434966	gggCAAGGCTCTGCTCCAGGACAGAGT	-	9
55	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	82053716	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	9
56	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	84994867	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
57	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	85791796	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
58	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	9364802	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	9
59	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	93611661	gggCAAGGCTCTGCTCCAGGACAGAGT	-	9
60	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	93645694	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
61	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	10508767	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
62	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	105117350	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	9
63	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	10860572	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
64	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	117143318	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	8
65	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	119080135	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
66	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	126819405	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
67	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	127089292	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
68	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	130267469	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
69	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	130540758	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
70	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	131044485	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
71	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	160713046	gggCAAGGCTCTGCTCCAGGACAGAGT	-	9
72	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	160802250	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	7
73	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	162278882	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
74	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	16474692	gggCAAGGCTCTGCTCCAGGACAGAGT	-	9
75	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	164089776	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	9
76	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	164618656	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	7
77	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	164852526	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
78	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	168041591	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
79	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	170015101	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
80	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	179914167	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	7
81	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	180212667	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
82	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr2	18144088	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
83	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	11081365	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
84	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	18621066	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	7
85	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	19951163	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	9
86	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	21671508	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	9
87	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	34729203	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	8
88	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	38817310	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	8
89	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	48228320	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
90	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	5179594	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
91	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	52184374	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
92	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	55076356	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
93	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	57982882	CACCGAAGGCTCTGCTCCAGGACAGAGT	+	8
94	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	60211721	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
95	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	69549643	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
96	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	87805461	CACCGAAGGCTCTGCTCCAGGACAGAGT	+	9
97	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	89390004	CACCGAAGGCTCTGCTCCAGGACAGAGT	-	9
98	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	89911373	CACCGAAGGCTCTGCTCCAGGACAGAGT	+	9
99	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	98329680	CACCGAAGGCTCTGCTCCAGGACAGAGT	+	8
100	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	108428269	CACCGAAGGCTCTGCTCCAGGACAGAGT	-	9
101	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	115947255	CACCGAAGGCTCTGCTCCAGGACAGAGT	-	9
102	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	116888897	CACCGAAGGCTCTGCTCCAGGACAGAGT	-	9
103	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	117537453	CACCGAAGGCTCTGCTCCAGGACAGAGT	-	9
104	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	119127176	CACCGAAGGCTCTGCTCCAGGACAGAGT	-	9
105	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	127683845	CACCGAAGGCTCTGCTCCAGGACAGAGT	+	9
106	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	130680135	CTtGtAGGCTCTGCTCCAGGACAGAGT	+	9
107	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	130315796	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
108	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	131116757	gggCAAGGCTCTGCTCCAGGACAGAGT	-	9
109	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	131471628	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	8
110	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	147117145	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
111	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	153279735	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
112	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	153488946	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
113	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr3	154251972	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	9
114	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	8579745	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	9
115	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	18021813	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
116	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	24631445	CACCGAAGGCTCTGCTCCAGGACAGAGT	-	9
117	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	31636941	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	9
118	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	37414488	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
119	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	37947240	CTtGtAGGCTCTGCTCCAGGACAGAGT	-	7
120	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	37063381	CTtGtAGGCTCTGCTCCAGGACAGAGT	-	9
121	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	38565722	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
122	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	41611043	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	8
123	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	41631617	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
124	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	43631260	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	8
125	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	46610409	CTtGtAGGCTCTGCTCCAGGACAGAGT	-	9
126	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	48027877	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	9
127	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	49151863	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	9
128	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	53545042	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
129	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	56611614	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	9
130	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	57847249	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	8
131	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	62212225	CAGtGatGatGatTCtCAAGCAGGCTGGAT	+	9
132	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	63877939	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	9
133	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	103500698	TCtCAAGGCTCggtgTgAGCAGGATAGGT	+	9
134	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	107496668	TCtCAAGGCTCggtgTgAGCAGGATAGGT	-	9
135	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	120025417	CACCGAAGGCTCTGCTCCAGGACAGAGT	-	9
136	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	109802772	CACCGAAGGCTCTGCTCCAGGACAGAGT	-	9
137	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	114586084	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	9
138	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr4	114597689	CAGtGatGatGatTCtCAAGCAGGCTGGAT	-	9
139	CACCGAAGGCGCTGGCAGCAG					

148	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	13119993	TGTGAAGGCTGAGTGGACAGCGTGAAT		9
149	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	13499917	mACGCGGCGGAGTCAAGCAGCGAAT		9
150	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	13626299	AAACGAGCGATCTAACAGCAGCGGAT	+	9
151	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	13797382	TCGAGGAGGCGGACAGCAGGTCAGAT	+	9
152	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	13774204	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
153	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	13907468	AGAGGAGGCTGGGCGACAGCGGGGT	+	9
154	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	13921803	AGAGGAGGCTGGGCGACAGCGAGGAT	+	9
155	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	13996800	AGGAAAGGCGGCGGTCAGCGAGGCGT	+	8
156	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14132582	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
157	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14183300	CACGAGGATCTAGGCGACAGCGTGAAT	+	9
158	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14203812	CACGAGGATCTAGGCGACAGCGTGAAT	+	9
159	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	14451823	TCGAGGAGGCGGAGTCAAGCAGCGAAT	+	9
160	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14471448	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
161	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	14835578	TCGAGGAGGCGGAGTCAAGCAGCGAAT	+	9
162	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	14960529	CACGCGGGGTCGAGCAGCGAGGAT	+	8
163	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	14984826	CAGTGGTGGTGGGCGACAGCGTGAAT	+	9
164	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	15071282	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
165	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	15128712	hGCAAGGAGGATGTGGACCGAGCGTGAAT	+	9
166	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	15204503	CAGTGGATCTGGGAGCGACAGCGGGT	+	8
167	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	15427399	CACTGAATCTGGGCGACAGCGAGGAT	+	8
168	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	15694774	CAGGAGGAGGCGGAGTCAAGCAGCGAAT	+	9
169	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	15700000	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
170	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	17375110	CGAGCAAGATCTAGGACAGCGCTGAAT	+	8
171	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	18085515	CACAGAGGATCTGGGCGACAGCGAGAT	+	9
172	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	18377137	TCGAGGAGGCGGAGTCAAGCAGCGAAT	+	9
173	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	18377190	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
174	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	18100183	AGTGAAGGCTGGAGCGACAGCAAGAT	+	6
175	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	4378870	CACGAGGATCTAGGCGACAGCGTGAAT	+	9
176	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	10519126	CAGGAGGAGGCGGAGTCAAGCAGCGAAT	+	9
177	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	10399454	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
178	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	10474000	AGTCCGACGCTGGTCTGAGGCTGGGAT	+	9
179	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	10646403	TCGAGGAGGCGGAGTCAAGCAGCGAAT	+	8
180	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	11317482	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
181	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	11316283	TCGAGGAGGCGGAGTCAAGCAGCGAAT	+	9
182	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	114313628	CAGCGAGGCTCAGGCGACAGCGAGGAT	+	9
183	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	114313628	CAGCGAGGCTCAGGCGACAGCGAGGAT	+	9
184	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	11478328	CACCGAGGCTCAGGCGACAGCGCGGAT	+	9
185	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	11539281	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
186	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	12603906	CACTGAATCTGGGCGACAGCGAGGAT	+	9
187	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	11715752	TCGAGGAGGCGGAGTCAAGCAGCGAAT	+	7
188	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	11729617	TCGAGGAGGCTGGGCGACAGCGTGAAT	+	9
189	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	12117400	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
190	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	12810829	CACTGAAGGCTGGGCGACAGCGAGGAT	+	7
191	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	12609933	CTCAAGGCTCTGGTACAGCGCATGAAT	+	9
192	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	12861203	CACTGAATCTGGGCGACAGCGAGGAT	+	9
193	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	13006488	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
194	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	13061722	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
195	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	13070786	CAAGAGCTCTCAGGAGGAGGAGGCTGGGAT	+	8
196	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	13168701	TCGAGGAGGCTGGGCGACAGCGTGAAT	+	9
197	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	13171783	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
198	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	13171783	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
199	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	13544943	CACTGAAGGCTGGGCGACAGCGAGGAT	+	7
200	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	13979397	AGTGAAGGCTGGAGCGACAGCAAGAT	+	6
201	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	40670003	CTCGAATCTGGGCGACAGCGAGGAT	+	9
202	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	40761782	TCGAGGAGGCGGAGTCAAGCAGCGAAT	+	9
203	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	42010123	CTTCAAGGAGGCTGGGCGACAGCGTGAAT	+	9
204	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	140049950	CAGGAGGAGGCGGAGTCAAGCAGCGAAT	+	9
205	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	144881996	AGTATAGGCTCTGGTCAAGCGCAAGAT	+	9
206	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14582182	mBGCCTGGGCTGGGCGACAGCGTGAAT	+	9
207	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14582182	mBGCCTGGGCTGGGCGACAGCGTGAAT	+	9
208	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	15059495	ACMGCTGGAGGCTGGGCGACAGCGTGAAT	+	9
209	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	17817896	CACGAGGCTCTCAGGCTCAGGCTGGGAT	+	9
210	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	18390394	mACCCATCTCAACCTGGTCTGAGGCTGGGAT	+	7
211	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	29663904	CACTGAAGGCTCTGGTCAAGCGCATGAAT	+	9
212	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	35551789	CTGAGGAGGCGGACAGCAGGTCAGAT	+	9
213	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	36371617	hGAAGGAGGCTGGGCGACAGCGAGGAT	+	8
214	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	38551799	CGGGGCTGGGCTGGGCGACAGCGGGAT	+	9
215	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	38898496	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
216	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	38898496	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
217	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	38898496	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
218	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	48713013	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
219	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	50339773	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
220	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	51408307	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
221	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	51408307	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
222	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	58019878	hAGAGGAGGCTGGGCGACAGCGAGGAT	+	9
223	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	61961946	hAGAGGAGGCTGGGCGACAGCGAGGAT	+	9
224	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	72989175	CAGTGGCTGGTCTGGGCGACAGCGTGAAT	+	8
225	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	72989175	CAGTGGCTGGTCTGGGCGACAGCGTGAAT	+	8
226	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	82876317	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
227	CACGAGGGGCGGGCAGCAGCGNNNNN	chr4	82876317	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
228	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	82815450	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
229	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	84367590	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
230	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	84367590	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
231	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	90571883	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
232	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	90571883	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
233	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	90599564	CTTGAAGGCTGGGCGACAGCGTGAAT	+	7
234	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	90667517	hAGAGGAGGCTCTCAACCTGGTCTGAGGAT	+	9
235	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	90667517	hAGAGGAGGCTCTCAACCTGGTCTGAGGAT	+	9
236	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	90667517	hAGAGGAGGCTCTCAACCTGGTCTGAGGAT	+	9
237	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	103079000	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
238	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	106128458	hACAGGAGGCTGGGCGACAGCGAGGAT	+	9
239	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	106128458	hACAGGAGGCTGGGCGACAGCGAGGAT	+	9
240	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
241	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
242	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
243	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
244	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
245	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
246	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
247	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
248	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
249	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
250	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
251	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
252	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
253	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
254	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
255	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
256	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
257	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
258	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
259	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
260	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
261	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
262	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
263	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
264	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
265	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
266	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
267	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
268	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
269	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
270	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
271	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
272	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
273	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
274	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
275	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
276	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
277	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
278	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
279	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
280	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
281	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
282	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
283	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
284	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
285	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
286	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
287	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
288	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
289	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
290	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
291	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
292	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
293	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
294	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
295	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
296	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
297	CACGAGGGGCTGGCGACAGGCGNNNN	chr4	14687633	CACTGAAGGCTGGGCGACAGCGAGGAT	+	9
298						

298	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	31150434	hCCGAGGCGaGgigACCAGGCAAGAT	+	9
299	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	38878074	CAhCGGCGGCTGGgCAGGCTTGAT	+	8
300	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	38883608	ChCAGGCGGCTGGgACAGGCTTGAT	+	9
301	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	37092813	CACCGAAGGCGCTGGgACAGGCTTGAT	+	9
302	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	38123045	CAGCAGGCGGCTGGgACAGGCTTGAT	-	9
303	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	44972382	hggAAGGCGCTGGgACAGGCTTGAT	-	7
304	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	61987142	hggAAGGCGCTGGgACAGGCTTGAT	-	9
305	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	68123824	hggAAGGCGCTGGgACAGGCTTGAT	+	9
306	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	79214171	CACCGAAGGCGCTGGgACAGGCTTGAT	+	9
307	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	79451983	hCAGGCGGCTGGgACAGGCTTGAT	+	8
308	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	79727951	hCAGGCGGCTGGgACAGGCTTGAT	+	9
309	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	79801854	CAGGCGGCGCTGGgACAGGCTTGAT	-	8
310	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	80062652	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
311	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	80994047	hCAGGCGGCTGGgACAGGCTTGAT	+	9
312	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	81468367	hCAGGCGGCTGGgACAGGCTTGAT	+	9
313	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	81578428	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
314	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	81778798	CAGGCGGCGCTGGgACAGGCTTGAT	+	8
315	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	82771251	hggAAGGCGCTGGgACAGGCTTGAT	-	9
316	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	82771352	hggAAGGCGCTGGgACAGGCTTGAT	-	9
317	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	82771740	hggAAGGCGCTGGgACAGGCTTGAT	-	9
318	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	82771840	hggAAGGCGCTGGgACAGGCTTGAT	-	9
319	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	87666635	CAGGCGGCGCTGGgACAGGCTTGAT	-	8
320	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	88123842	hCAGGCGGCTGGgACAGGCTTGAT	+	9
321	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	98410864	hCAGGCGGCTGGgACAGGCTTGAT	-	9
322	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	10104600	hCAGGCGGCTGGgACAGGCTTGAT	-	9
323	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	101349375	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
324	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	11167849	CAGGCGGCGCTGGgACAGGCTTGAT	+	8
325	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	111695118	hCAGGCGGCTGGgACAGGCTTGAT	-	9
326	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	112773259	CAGGCGGCGCTGGgACAGGCTTGAT	-	8
327	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	113018110	hCAGGCGGCTGGgACAGGCTTGAT	-	9
328	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	11484101	hCAGGCGGCTGGgACAGGCTTGAT	+	8
329	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	11515895	hCAGGCGGCTGGgACAGGCTTGAT	+	9
330	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	117001778	CAGGCGGCGCTGGgACAGGCTTGAT	+	7
331	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	117002279	CAGGCGGCGCTGGgACAGGCTTGAT	+	7
332	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	12741234	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
333	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	13413058	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
334	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	135510409	CAGGCGGCGCTGGgACAGGCTTGAT	+	8
335	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	138607181	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
336	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	140210641	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
337	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	143110786	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
338	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	142624184	CAGGCGGCGCTGGgACAGGCTTGAT	+	7
339	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	142960249	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
340	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	144513026	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
341	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr7	145295997	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
342	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	3624632	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
343	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	12853787	CAGGCGGCGCTGGgACAGGCTTGAT	-	8
344	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	14926525	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
345	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	15201178	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
346	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	26974740	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
347	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	31922609	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
348	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	44554008	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
349	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	45054404	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
350	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	46654050	CAGGCGGCGCTGGgACAGGCTTGAT	+	8
351	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	47635899	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
352	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	48818821	CAGGCGGCGCTGGgACAGGCTTGAT	-	7
353	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	70131798	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
354	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	71352363	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
355	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	78585857	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
356	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	80336949	hggAAGGCGCTGGgACAGGCTTGAT	-	8
357	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	90241651	hggAAGGCGCTGGgACAGGCTTGAT	+	9
358	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	101340162	hggAAGGCGCTGGgACAGGCTTGAT	-	8
359	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	104182220	hCAGGCGGCTGGgACAGGCTTGAT	+	9
360	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	104985257	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
361	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	105492218	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
362	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	10886788	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
363	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	110878472	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
364	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	114816077	hCAGGCGGCTGGgACAGGCTTGAT	-	9
365	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	119478932	CAGGCGGCGCTGGgACAGGCTTGAT	-	7
366	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	12068114	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
367	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	120860571	CAGGCGGCGCTGGgACAGGCTTGAT	-	8
368	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	120974938	hggAAGGCGCTGGgACAGGCTTGAT	+	9
369	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	123041378	hggAAGGCGCTGGgACAGGCTTGAT	-	9
370	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	12511189	hCAGGCGGCTGGgACAGGCTTGAT	+	9
371	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	125556642	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
372	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr8	126158536	CAGGCGGCGCTGGgACAGGCTTGAT	-	8
373	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	621347	CAGGCGGCGCTGGgACAGGCTTGAT	+	8
374	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	14751378	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
375	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	21916518	hCAGGCGGCTGGgACAGGCTTGAT	+	9
376	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	22034827	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
377	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	22617858	hCAGGCGGCTGGgACAGGCTTGAT	+	9
378	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	23021171	hCAGGCGGCTGGgACAGGCTTGAT	+	9
379	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	23123824	CAGGCGGCGCTGGgACAGGCTTGAT	+	8
380	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	26613462	hCAGGCGGCTGGgACAGGCTTGAT	-	9
381	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	27811492	hCAGGCGGCTGGgACAGGCTTGAT	+	8
382	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	40241451	hCAGGCGGCTGGgACAGGCTTGAT	+	9
383	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	42855882	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
384	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	43743430	hCAGGCGGCTGGgACAGGCTTGAT	+	9
385	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	46913410	hCAGGCGGCTGGgACAGGCTTGAT	+	6
386	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	49517223	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
387	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	52786660	CAGGCGGCGCTGGgACAGGCTTGAT	+	7
388	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	53321364	CAGGCGGCGCTGGgACAGGCTTGAT	+	8
389	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	57503474	CAGGCGGCGCTGGgACAGGCTTGAT	+	7
390	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	60247801	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
391	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	61384373	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
392	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	64028355	hCAGGCGGCTGGgACAGGCTTGAT	-	9
393	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	64321431	hCAGGCGGCTGGgACAGGCTTGAT	+	7
394	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	64744207	hCAGGCGGCTGGgACAGGCTTGAT	+	9
395	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	65228873	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
396	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	69431414	CAGGCGGCGCTGGgACAGGCTTGAT	-	8
397	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	77945476	hCAGGCGGCTGGgACAGGCTTGAT	+	8
398	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	86468771	hCAGGCGGCTGGgACAGGCTTGAT	+	9
399	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	87740653	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
400	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	90022393	hCAGGCGGCTGGgACAGGCTTGAT	-	9
401	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	96933884	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
402	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	96977120	CAGGCGGCGCTGGgACAGGCTTGAT	+	8
403	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	98334796	hCAGGCGGCTGGgACAGGCTTGAT	+	9
404	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	106552854	CAGGCGGCGCTGGgACAGGCTTGAT	+	7
405	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	106652201	hCAGGCGGCTGGgACAGGCTTGAT	+	9
406	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	107778977	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
407	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	109134664	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
408	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	111481582	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
409	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	115916828	CAGGCGGCGCTGGgACAGGCTTGAT	-	8
410	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	116019882	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
411	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr9	119728955	CAGGCGGCGCTGGgACAGGCTTGAT	+	7
412	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	3754863	hCAGGCGGCTGGgACAGGCTTGAT	-	8
413	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	4744761	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
414	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	5147787	hCAGGCGGCTGGgACAGGCTTGAT	+	8
415	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	10301849	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
416	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	12721008	hCAGGCGGCTGGgACAGGCTTGAT	+	8
417	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	21806329	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
418	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	29217698	CAGGCGGCGCTGGgACAGGCTTGAT	+	7
419	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	29534725	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
420	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	31382172	CAGGCGGCGCTGGgACAGGCTTGAT	-	8
421	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	39018711	hCAGGCGGCTGGgACAGGCTTGAT	+	9
422	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	39514818	hCAGGCGGCTGGgACAGGCTTGAT	+	8
423	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	62424916	hCAGGCGGCTGGgACAGGCTTGAT	+	9
424	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	62807998	hCAGGCGGCTGGgACAGGCTTGAT	+	8
425	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	70037110	hCAGGCGGCTGGgACAGGCTTGAT	+	9
426	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	70813707	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
427	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	71101122	CAGGCGGCGCTGGgACAGGCTTGAT	+	7
428	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	78196582	CAGGCGGCGCTGGgACAGGCTTGAT	-	7
429	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	78623512	hCAGGCGGCTGGgACAGGCTTGAT	+	8
430	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	78653456	CAGGCGGCGCTGGgACAGGCTTGAT	+	5
431	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	79923552	hCAGGCGGCTGGgACAGGCTTGAT	+	9
432	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	83675226	CAGGCGGCGCTGGgACAGGCTTGAT	+	9
433	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	11706251	CAGGCGGCGCTGGgACAGGCTTGAT	+	8
434	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	12094625	hCAGGCGGCTGGgACAGGCTTGAT	-	8
435	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	120738425	hCAGGCGGCTGGgACAGGCTTGAT	+	9
436	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr10	128913813	CAGGCGGCGCTGGgACAGGCTTGAT	-	9
437	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr11	5252790	hCAGGCGGCTGGgACAGGCTTGAT	-	9
438	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr11	7421018	hCAGGCGGCTGGgACAGGCTTGAT	-	9
439	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr11	8711999	hCAGGCGGCTGGgACAGGCTTGAT	+	9
440	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr11	20371071	hCAGGCGGCTGGgACAGGCTTGAT	-	9
441	CACCGAAGGCGCTGGCAGCAGGNNNNNN	chr11	26961809	hCAGGCGGCTGGg		

448	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	5886272	AtaGAAGGgTGtSggGCTAGCGGGGGAT	-	9
449	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	5897089	BAtaAAGGgTGtTGCACAGCGCTGGAT	-	9
450	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	6066293	BAtaAAGGgTGtTGCACAGCGCTGGAT	-	9
451	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	6061500	BAtaAAGGgTGtTGCACAGCGCTGGAT	-	9
452	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	6062472	BAtaAAGGgTGtTGCACAGCGCTGGAT	-	9
453	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	6064723	BAtaAAGGgTGtTGCACAGCGCTGGAT	-	9
454	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	6066344	BAtaAAGGgTGtTGCACAGCGCTGGAT	-	9
455	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	6153768	CACAGAGGCTTGAGGCTAGGCTGAAT	+	7
456	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr11	63110471	CACAGGCTTGAAGGAGGAGGAGGTTGGAT	+	9
457	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr11	6391474	BgCTGAGGCGAGGCTTGACAGGCTAGGAT	+	9
458	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	6796116	ChCAGAGGCTTGAGGCTAGGCTAGGAT	+	9
459	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	6965323	CcAGAGAGGAGGAGGAGCAGGAGCAAT	+	9
460	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	72116055	ctCTGAGAGGCTTGAGGCTAGGAGGAT	+	9
461	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	80511260	gAtaAAAGGCTTGAGGCTAGGAGGAT	+	8
462	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr11	81781637	hAGgAGGAGGCTTGAGGCTAGGAGGAT	+	9
463	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	83327345	CAGgggTGGGCTTGAGGCTAGGAGGAT	-	8
464	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr11	85710844	CAGtGGGAGGCTTGAGGCTAGGAGGAT	-	9
465	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	85753263	BgCAGAGGCTTGACACAGGCAAGAT	+	9
466	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	85812147	ChCAGAGGCTTGAGGCTAGGAGGAT	+	9
467	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	88504549	hCTGAGGCGCTTGAGGCTAGGAGGAT	-	8
468	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr11	91269261	hAtaACtAGCTTGAGGAGGAGGCTTGAT	+	9
469	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr11	95563937	CACaACtAGCTTGAGGAGGAGGAGGTTGGAT	-	8
470	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	99941086	CAGCTGGGAGGAGGAGCAGGAGTGGAT	+	9
471	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr11	9819292	CTCCAGGAGCTTGAGGAGGAGTGGAT	+	9
472	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	98185411	hTCCAAAGGCTTGAGGAGGAGTGGAT	-	9
473	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr11	98498861	CAGGAGGAGGCTTGAGGAGGAGTGGAT	+	9
474	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	98962560	BAtaAAGGCTTGAGGAGGAGTGGAT	+	9
475	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr11	99006518	CAGGAGGAGGCTTGAGGAGGAGTGGAT	+	9
476	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr11	102217537	CACAGGAGGCTTGAGGAGGAGTGGAT	+	7
477	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr11	102769375	hAGAGGCTTGAGGAGGAGGAGTGGAT	-	9
478	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	103682781	BAtaAAGGCTTGAGGAGGAGTGGAT	+	9
479	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr11	10263794	ChCAGAGGCTTGAGGAGGAGTGGAT	+	9
480	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	106361263	hACAGAGGAGGCTTGAGGAGGAGTGGAT	-	9
481	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	107518125	CtAGAGGAGGCTTGAGGAGGAGTGGAT	+	9
482	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	107951510	hCAGAGGCTTGAGGAGGAGTGGAT	-	9
483	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	112270871	hACAGAGGCTTGAGGAGGAGTGGAT	+	9
484	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	113070859	hACAGAGGCTTGAGGAGGAGTGGAT	+	8
485	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	114783454	CTCAGAGGCTTGAGGAGGAGTGGAT	+	9
486	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	118468890	CTCCAGAGGCTTGAGGAGGAGTGGAT	+	8
487	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr11	119019532	CTCCAGAGGCTTGAGGAGGAGTGGAT	+	9
488	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr11	120476342	CAGAGGAGGCTTGAGGAGGAGTGGAT	-	9
489	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr12	8858012	CAGGAGGAGGCTTGAGGAGGAGTGGAT	+	9
490	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr12	16463830	BgCAGAGGCTTGAGGAGGAGTGGAT	+	8
491	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr12	17695510	ChCAGAGGCTTGAGGAGGAGTGGAT	+	9
492	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr12	2153825	CAGGAGGCTTGAGGAGGAGTGGAT	-	8
493	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr12	24731791	CACAGGAGGCTTGAGGAGGAGTGGAT	-	9
494	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr12	25120352	hCTCAGAGGCTTGAGGAGGAGTGGAT	-	9
495	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr12	30913976	ChCAGAGGCTTGAGGAGGAGTGGAT	+	9
496	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr12	3936480	BgAGAGGCTTGAGGAGGAGTGGAT	+	9
497	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr12	37539562	hTCAAGAGCTTGAGGAGGAGTGGAT	+	9
498	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr12	45311226	CACAGAGGCTTGAGGAGGAGTGGAT	-	9
499	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr12	11564130	ChCAGAGGCTTGAGGAGGAGTGGAT	+	9
500	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr12	55776123	hACAGAGGCTTGAGGAGGAGTGGAT	-	9
501	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr12	66280707	CAGGAGGAGGCTTGAGGAGGAGTGGAT	+	9
502	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr12	71431937	BgAGAGGCTTGAGGAGGAGTGGAT	+	9
503	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr12	7904969	ChCAGAGGCTTGAGGAGGAGTGGAT	+	9
504	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr12	79061547	hACAGAGGCTTGAGGAGGAGTGGAT	+	9
505	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr12	82361332	CACAGAGGCTTGAGGAGGAGTGGAT	+	8
506	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr12	82551317	CACAGAGGCTTGAGGAGGAGTGGAT	-	9
507	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr12	86398817	hAGAGAGGCTTGAGGAGGAGTGGAT	+	9
508	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr12	86621523	hAGAGAGGCTTGAGGAGGAGTGGAT	+	9
509	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr12	100325518	hAGAGAGGCTTGAGGAGGAGTGGAT	-	9
510	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr12	105870943	hTCAAGAGCTTGAGGAGGAGTGGAT	+	9
511	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr12	106577437	BgAGAGGCTTGAGGAGGAGTGGAT	-	9
512	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr12	118301253	CAGAGAGGCTTGAGGAGGAGTGGAT	+	9
513	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr13	12179661	hACAGAGGCTTGAGGAGGAGTGGAT	-	7
514	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	13964861	CAGAGAGGCTTGAGGAGGAGTGGAT	-	9
515	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	20914238	hACAGAGGCTTGAGGAGGAGTGGAT	-	9
516	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	24919802	hTCAAGAGCTTGAGGAGGAGTGGAT	+	9
517	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr13	34206479	CAGAGAGGCTTGAGGAGGAGTGGAT	-	9
518	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	37149519	hAGAGAGGCTTGAGGAGGAGTGGAT	-	9
519	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	42978966	BgAGAGGCTTGAGGAGGAGTGGAT	-	9
520	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	43421534	hAGAGAGGCTTGAGGAGGAGTGGAT	+	9
521	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr13	44055834	CTCCAGAGGCTTGAGGAGGAGTGGAT	-	9
522	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	44229402	hAGAGAGGCTTGAGGAGGAGTGGAT	+	8
523	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	46897173	CAGAGAGGCTTGAGGAGGAGTGGAT	-	8
524	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	51863896	ChCAGAGGCTTGAGGAGGAGTGGAT	+	9
525	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	52045981	CAGAGAGGCTTGAGGAGGAGTGGAT	+	9
526	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	52831465	CAGAGAGGCTTGAGGAGGAGTGGAT	+	8
527	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr13	53100751	hAGAGAGGCTTGAGGAGGAGTGGAT	-	9
528	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr13	54401068	CAGAGAGGCTTGAGGAGGAGTGGAT	+	9
529	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr13	55391654	hACAGAGGCTTGAGGAGGAGTGGAT	+	9
530	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	58899317	CACAGAGGCTTGAGGAGGAGTGGAT	+	9
531	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	70126600	hACAGAGGCTTGAGGAGGAGTGGAT	-	9
532	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr13	71070081	hAGAGAGGCTTGAGGAGGAGTGGAT	+	9
533	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	72366003	CACAGAGGCTTGAGGAGGAGTGGAT	-	9
534	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr13	73239445	hTCAAGAGCTTGAGGAGGAGTGGAT	-	8
535	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	73820815	CAGAGAGGCTTGAGGAGGAGTGGAT	-	8
536	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	79327572	CTCCAGAGGCTTGAGGAGGAGTGGAT	+	9
537	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	98475919	CAGAGAGGCTTGAGGAGGAGTGGAT	+	9
538	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr13	98754007	CAGAGAGGCTTGAGGAGGAGTGGAT	+	9
539	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr13	100689073	BgAGAGGCTTGAGGAGGAGTGGAT	-	9
540	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	10794961	hAGAGAGGCTTGAGGAGGAGTGGAT	+	9
541	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	102681498	CTCCAGAGGCTTGAGGAGGAGTGGAT	-	8
542	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr13	105444228	CACAGAGGCTTGAGGAGGAGTGGAT	+	7
543	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr13	111052405	CAGAGAGGCTTGAGGAGGAGTGGAT	-	8
544	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr13	11273181	hACAGAGGCTTGAGGAGGAGTGGAT	+	9
545	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr13	114136231	CACAGAGGCTTGAGGAGGAGTGGAT	-	7
546	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr14	11910722	CAGAGAGGCTTGAGGAGGAGTGGAT	+	9
547	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	12509076	CACAGAGGCTTGAGGAGGAGTGGAT	+	8
548	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	14986967	CACAGAGGCTTGAGGAGGAGTGGAT	+	9
549	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	23975031	CAGAGAGGCTTGAGGAGGAGTGGAT	+	9
550	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	25304711	CTCAGAGGCTTGAGGAGGAGTGGAT	-	8
551	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr14	25773728	CTCAGAGGCTTGAGGAGGAGTGGAT	-	9
552	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	26531051	CAGAGAGGCTTGAGGAGGAGTGGAT	+	7
553	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr14	28401010	CAGAGAGGCTTGAGGAGGAGTGGAT	+	9
554	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr14	30801201	CAGAGAGGCTTGAGGAGGAGTGGAT	-	9
555	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	30612628	CAGAGAGGCTTGAGGAGGAGTGGAT	-	9
556	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr14	32941154	hACAGAGGCTTGAGGAGGAGTGGAT	+	8
557	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr14	41192608	CACAGAGGCTTGAGGAGGAGTGGAT	+	9
558	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr14	53622914	CAGAGAGGCTTGAGGAGGAGTGGAT	+	9
559	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	57798611	CTCCAGAGGCTTGAGGAGGAGTGGAT	-	8
560	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	62801514	hAGAGAGGCTTGAGGAGGAGTGGAT	+	7
561	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	63491007	CTCAGAGGCTTGAGGAGGAGTGGAT	-	9
562	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	63703799	hAGAGAGGCTTGAGGAGGAGTGGAT	-	8
563	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	66153763	CAGAGAGGCTTGAGGAGGAGTGGAT	+	9
564	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr14	70861731	CAGAGAGGCTTGAGGAGGAGTGGAT	+	9
565	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr14	70895904	CACAGAGGCTTGAGGAGGAGTGGAT	+	8
566	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	72181539	CACAGAGGCTTGAGGAGGAGTGGAT	-	9
567	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	74495784	CAGAGAGGCTTGAGGAGGAGTGGAT	-	9
568	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	75613804	hAGAGAGGCTTGAGGAGGAGTGGAT	-	9
569	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	76617123	hACAGAGGCTTGAGGAGGAGTGGAT	+	9
570	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr14	76674124	CAGAGAGGCTTGAGGAGGAGTGGAT	+	9
571	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	80562162	CTCAGAGGCTTGAGGAGGAGTGGAT	-	9
572	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	8341636	CTCAGAGGCTTGAGGAGGAGTGGAT	+	9
573	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	84127278	CTCAGAGGCTTGAGGAGGAGTGGAT	+	9
574	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr14	86951751	hCTCAGAGGCTTGAGGAGGAGTGGAT	+	9
575	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	88169881	CCTCAGAGGCTTGAGGAGGAGTGGAT	+	9
576	CACCGACCCCACTGAGTGGAGGTTNNNNN	chr14	101214710	CTCAGAGGCTTGAGGAGGAGTGGAT	+	9
577	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr14	10514141	CTCAGAGGCTTGAGGAGGAGTGGAT	+	9
578	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	118049261	hAGAGAGGCTTGAGGAGGAGTGGAT	-	9
579	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr14	121666531	CTCAGAGGCTTGAGGAGGAGTGGAT	+	8
580	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr15	45397474	hACAGAGGCTTGAGGAGGAGTGGAT	+	8
581	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr15	45939662	hAGAGAGGCTTGAGGAGGAGTGGAT	+	9
582	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr15	12809175	CTCAGAGGCTTGAGGAGGAGTGGAT	-	9
583	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr15	35255002	CACAGAGGCTTGAGGAGGAGTGGAT	+	9
584	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr15	37068666	CACAGAGGCTTGAGGAGGAGTGGAT	+	9
585	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr15	47999921	CTCAGAGGCTTGAGGAGGAGTGGAT	+	9
586	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr15	95450985	CACAGAGGCTTGAGGAGGAGTGGAT	-	9
587	CACCGAAGGCGCTGGCACCAGCGNNNNNN	chr15	63954610	CAGAGAGGCTTGAGGAGGAGTGGAT	-	9
588	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr15	65109882	CACAGAGGCTTGAGGAGGAGTGGAT	+	9
589	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr15	78017171	hAGAGAGGCTTGAGGAGGAGTGGAT	+	9
590	CACCGGCGGGCGCGCCAGCAGCGNNNNNN	chr15	74723850	CAGAGAGGCTTGAGGAGGAGTGGAT	+	8
591	CACCGAAGGCGCTGGCACCAGCGNNNNNN					

Supplemental Table 6. Primers, plasmids and sgRNAs used in this study

	Name	Sequences (5' to 3')	ID Number
Primers	U6_gRNA_PCR_F	GAG GGC CTA TTT CCC ATG ATT CCT	RDC 255
	dTomato_guide_B	AAA CCA CTG TGG GGT GGA GGG GAC	RDC 1194
	mGAPDH F	TGTTTGTGATGGGTGTGAACC	RDC 345
	mGAPDH R	ACTGTGGTCATGAGCCCTTC	RDC 346
	Lama1 Primer FW	GGAAGGTTACAAAGTTCGATTGG	RDC 977
	Lama1 Primer RV	ACGTGAAATAAGACCTTGCCATC	RDC 978
	Lama1 qPCR 1 KL F	TTTCTTTGAAGGAAGCGGATATG	RDC 1918
	Lama1 qPCR 2 KL F	TGGGTCCATTAACAGAAGGAAAG	RDC 1919
	Lama1 qPCR 2 KL R	ATGGGTGAAGTCCAAAAGTTGAG	RDC 1920
	Vector genome F	ATGGTGAGCAAGGGCGAGG	RDC 1687
	Vector genome R	AGCTTGCCGTAGGTGGCATC	RDC 1679

	Name	ID number
Plasmids	minCMV-tdTomato	RDC 262
	px601 SadCas9-VP64	RDC 153
	3-guide cassettes	RDC 367

	Name	Sequences (5' to 3')	PAM
sgRNAs	<i>tdTomato</i> sgRNA Top	CACCGTCCCCTCCACCCACAGTG	AAGAAT
	<i>tdTomato</i> sgRNA Bottom	AAACCACTGTGGGGTGGAGGGGAC	
	<i>Lama1</i> sgRNA1 Top	CACCGAAGGGCCTGCGCACCAGGC	CAGGGT
	<i>Lama1</i> sgRNA1 Bottom	AAACGCCTGGTGCAGGCCCTTC	
	<i>Lama1</i> sgRNA2 Top	CACCGCGGGGCGCGGCCAGGCAGGC	GGGGGT
	<i>Lama1</i> sgRNA2 Bottom	AAACGCCTGCCTGGCCGCGCCCCGC	
	<i>Lama1</i> sgRNA3 Top	CACCGTCCAGGGGGAGCCCCGCCGTG	CGGAGT
	<i>Lama1</i> sgRNA3 Bottom	AAACCACGGCGGGGCTCCCCCTGGAC	
	<i>Lama1</i> sgRNA4 Top	CACCGGTGGTGGCGGGGTGCCTCTC	CTGGGT
	<i>Lama1</i> sgRNA4 Bottom	AAACGAGAGGCACCCCGCCACCACC	
	<i>Lama1</i> sgRNA5 Top	CACCGACCCCACTGGAGTGGAGGGT	CAGAGT
	<i>Lama1</i> sgRNA5 Bottom	AAACACCCTCCACTCCAGTTGGGGTC	