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DATABASES



Update of the Pompe variant database for the prediction of clinical phenotypes: Novel disease-associated variants, common sequence variants, and results from newborn screening

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Abstract

Pompe disease is an inherited disorder caused by disease-associated variants in the acid α -glucosidase gene (GAA). The Pompe disease GAA variant database (http:// www.pompevariantdatabase.nl) is a curated, open-source, disease-specific database, and lists disease-associated GAA variants, in silico predictions, and clinical phenotypes reported until 2016. Here, we provide an update to include 226 diseaseassociated variants that were published until 2020. We also listed 148 common GAA sequence variants that do not cause Pompe disease. GAA variants with unknown severity that were identified only in newborn screening programs were listed as a new feature to indicate the reason why phenotypes were still unknown. Expression studies were performed for common missense variants to predict their severity. The updated Pompe disease GAA variant database now includes 648 disease-associated variants, 26 variants from newborn screening, and 237 variants with unknown severity. Regular updates of the Pompe disease GAA variant database will be required to improve genetic counseling and the study of genotype-phenotype relationships.

KEYWORDS

database, disease-associated variants, GAA, NBS, Pompe disease, SNP

1 | INTRODUCTION

Pompe disease (glycogen storage disease type II; MIM #232300) is an autosomal recessive disorder caused by disease-associated variants in the acid α -glucosidase (GAA) gene, resulting in a deficiency of the GAA enzyme, accumulation of lysosomal glycogen, and

progressive muscle weakness. The clinical spectrum of Pompe disease is broad (Güngör & Reuser, 2013). The most severe classic infantile phenotype presents shortly after birth with hypertrophic cardiomyopathy and generalized muscle weakness. These patients die in the first year of life due to cardiorespiratory insufficiency if left untreated. The slower progressing phenotype is characterized by

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muscle weakness that can appear at any age from <1 year into adulthood. These patients are generally spared from cardiac symptoms (Kohler et al., 2018; van der Ploeg & Reuser, 2008). Enzyme replacement therapy (ERT) with intravenously applied recombinant human GAA is available since 2006. ERT normalizes hypertrophic cardiomyopathy, improves motor function, and extends survival.

The differences between phenotypes in Pompe disease can, in part, be attributed to the severity of the disease-associated variants present in the GAA gene. Classic infantile patients carry two diseaseassociated variants that completely disrupt the function of GAA (i.e., null alleles). This group of patients can be subdivided based on their cross-reactive immunological material (CRIM) status, which is defined by the disease-associated variants involved. When two GAA variants are present that do not result in GAA protein expression, the patient is classified as CRIM-negative. When at least one GAA variant gives rise to GAA protein expression (in which the GAA protein can be enzymatically inactive), the patient is classified as CRIM-positive. The clinical importance of CRIM status is highlighted by the fact that CRIM-negative classic infantile patients have a poorer prognosis compared with CRIM-positive classic infantile patients, possibly due to the formation of high sustained anti-GAA antibody titers upon treatment with ERT (Bali et al., 2012; van Gelder et al., 2015). Patients who do not have the classic infantile phenotype carry at least one disease-associated variant that allows some residual enzymatic activity. These patients are, by definition, CRIM-positive (Kroos et al., 2012b; Kulessa et al., 2020).

The "Pompe disease GAA variant database" (http://www. pompevariantdatabase.nl) is an open-access database that lists and classifies all reported variants in the GAA gene. We recently revised this database to include clinical data from patients collected from the literature, adapted the classification system for variant severity, and added (predicted) CRIM status for disease-associated variants. The database included literature up to May 2016, resulting in a total of 561 variants (Niño et al., 2019). In recent years, many new patients and GAA variants have been reported. These include findings from large patient populations, such as the French nationwide study (246 patients with late-onset Pompe disease) and the Pompe registry (1079 patients from 26 countries; Reuser et al., 2019; Semplicini et al., 2018).

In addition, various countries, including Taiwan, the United States, Italy, Brazil, and Japan, have implemented newborn screening (NBS) programs for Pompe disease, resulting in an increase of variants of unknown significance (VUS; Bravo et al., 2017; Burlina et al., 2018; Chien et al., 2019; Elliott et al., 2016; Momosaki et al., 2019; Yang et al., 2014). For variants associated with late onset, the associated phenotypes from NBS cases are still unknown as symptom onset could, in principle, be delayed until (late) adulthood. It will be important to monitor the onset and progress of symptoms in patients identified via NBS programs closely to determine the severity of the newly identified genetic variants.

Public databases, such as dbSNP (https://www.ncbi.nlm.nih.gov/ snp) and gnomAD (https://gnomad.broadinstitute.org), provide a source of variants that have been detected in various genome-wide studies (Karczewski et al., 2020; Sherry et al., 2001). A large percentage of these variants represent common sequence variants that have a minor allele frequency (MAF) \geq 1%. Several of these variants have already been reported for the GAA gene and have been ruled out to cause Pompe disease (Kroos et al., 2007; Labrousse et al., 2010; Turaça et al., 2015). However, most of the common sequence variants in these databases are listed as VUSs and may lead to misinterpretation during molecular diagnostics.

In this study, we provide an update of the Pompe disease GAA variant database with variants and patients described in the literature up to January 2020. We included information on novel GAA variants that were identified via NBS and for which no phenotype was yet known. Known common sequence variants in the GAA gene that do not cause Pompe disease have now also been added to prevent misdiagnosis. In addition, selected common missense variants were tested in expression studies and also this information was added to the updated database. The database provides a curated up-to-date reference source for the molecular diagnosis of Pompe disease.

2 | METHODS

The Pompe disease GAA variant database is publicly available at http://www.pompevariantdatabase.nl. The previous version of the database included literature until 2016; the update described here contains variants from publications up to January 2020. Additionally, NBS studies that screened for Pompe disease were now included if the authors provided the genotypes of the described cases. Novel variants were analyzed as described in Niño et al. (2019). Variants were annotated based on the reference sequences NM 000152.3 for GAA messenger RNA (mRNA), LRG 673 genomic sequence for describing variants in intronic sequences, and NP_000143.2 for GAA protein. Exon annotations were based on the human genomic build (GRCH37/hg19) for exons 2-20; however, changes were made to the annotation of exon 1 to reflect the findings of (GRCH38/hg38). Within this region, a new 195-bp intron was identified at positions c.-112 and c.-113. Therefore, the region that was previously annotated as exon 1 has been split between exons 1A and 1B, which are separated by intron 1A. Intron 1 has been renamed to intron 1B. This numbering was made to maintain the same numbering of subsequent exons compared with existing literature.

Common sequence variants in the GAA gene (hg38 Chr17:80,101,556-80,119,881) were extracted from gnomAD and were categorized as "not disease-associated." Combined Annotation-Dependent Depletion (CADD) in silico predictions were performed using the CADD (https://cadd.gs.washington.edu) platform, which compiles different tools for analysis of intronic insertion and deletion variants (Rentzsch et al., 2019). The MAF and CADD scores were obtained in April 2020. Predictions of effect on pre-mRNA splicing were performed using Alamut Visual v.2.15 (Interactive Biosoftware).

Functional studies were performed using site-directed mutagenesis (SDM) to generate complementary DNA (cDNA) expression

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TABLE 1 Novel disease-associated variants added to the Pompe variant database

DNA nomenclature	Phenotype combined with a null allele	DNA nomenclature	Phenotype combined with a null allele
Ch37/hg19 chr17:78,059,821_ 78,076,592del	Unknown (disease-associated)	c.1057C>T	Unknown (disease-associated)
c113+2T>A	Unknown (disease-associated)	c.1057del	Unknown (disease-associated)
c32-1732-10delins(30)	Classic infantile	c.1099T>G	Unknown (disease-associated)
c32-1G>C	Unknown (disease-associated)	c.1106T>A	Unknown (disease-associated)
c.40_47del	Classic infantile	c.1109G>A	Unknown (disease-associated)
c.104T>C	Classic infantile	c.1114C>G	Unknown (disease-associated)
c.169C>T	Classic infantile	c.1114C>T	Unknown (disease-associated)
c.205C>T	Unknown (disease-associated)	c.1121G>A	Unknown (disease-associated)
c.258C>A	Unknown (disease-associated)	c.1127_1130del	Unknown (disease-associated)
c.265C>T	Unknown (disease-associated)	c.1129G>A	Unknown (disease-associated)
c.295_314del	Unknown (disease-associated)	c.1153del	Unknown (disease-associated)
c.323G>C	Unknown (disease-associated)	c.1192del	Unknown (disease-associated)
c.365del	Unknown (disease-associated)	c.1193del	Unknown (disease-associated)
c.380G>A	Unknown (disease-associated)	c.1201C>A	Unknown (disease-associated)
c.397T>G	Unknown (disease-associated)	c.1209C>A	Unknown (disease-associated)
c.437del	Classic infantile	c.1211A>C	Unknown (disease-associated)
c.445A>C	Unknown (disease-associated)	c.1211A>T	Classic infantile
c.484A>C	Classic infantile	c.1212C>G	Unknown (disease-associated)
c.502C>T	Unknown (disease-associated)	c.1216G>A	Childhood
c.505C>A	Unknown (disease-associated)	c.1219T>C	Unknown (disease-associated)
c.517_519del	Childhood	c.1221C>A	Classic infantile
c.541_545del	Classic infantile	c.1221del	Unknown (disease-associated)
c.547-1G>C	Unknown (disease-associated)	c.1226_1227insG	Classic infantile
c.568C>T	Unknown (disease-associated)	c.1231del	Unknown (disease-associated)
c.665T>G	Classic infantile	c.1240T>C	Unknown (disease-associated)
c.686G>C	Unknown (disease-associated)	c.1241del	Classic infantile
c.691C>T	Unknown (disease-associated)	c.1242C>A	Unknown (disease-associated)
c.692T>C	Unknown (disease-associated)	c.1249A>C	Unknown (disease-associated)
c.692+1G>T	Unknown (disease-associated)	c.1281G>T	Classic infantile
c.693-2A>C	Classic infantile	c.1292_1295dup	Classic infantile
c.693-1G>C	Unknown (disease-associated)	c.1293_1326+57del	Unknown (disease-associated)
c.715_716del	Unknown (disease-associated)	c.1298A>C	Classic infantile
c.730C>T	Classic infantile	c.1311_1312ins(26)	Classic infantile
c.736del	Unknown (disease-associated)	c.1320_1322del	Classic infantile
c.756_757insT	Unknown (disease-associated)	c.1327-54_1437+178del	Classic infantile
c.759del	Unknown (disease-associated)	c.1358_1361del	Classic infantile
c.766_784del	Unknown (disease-associated)	c.1378G>T	Unknown (disease-associated)
c.781G>A	Classic infantile	c.1388_1406del	Unknown (disease-associated)
c.784G>C	Unknown (disease-associated)	c.1396dup	Unknown (disease-associated)

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DNA nomenclature	Phenotype combined with a null allele		Phenotype combined with a null allele
c.796C>A	Childhood	c.1402A>T	Unknown (disease-associated)
c.799_803delinsA	Unknown (disease-associated)	c.1409A>G	Unknown (disease-associated)
c.837G>C	Unknown (disease-associated)	c.1431del	Classic infantile
c.841C>T	Unknown (disease-associated)	c.1441del	Unknown (disease-associated)
c.876C>G	Classic infantile	c.1447G>T	Unknown (disease-associated)
c.878G>T	Unknown (disease-associated)	c.1456G>T	Unknown (disease-associated)
c.883C>A	Unknown (disease-associated)	c.1464dup	Classic infantile
c.930_932del	Classic infantile	c.1470C>A	Childhood
c.942C>A	Unknown (disease-associated)	c.1477C>T	Unknown (disease-associated)
c.947A>G	Classic infantile	c.1493G>A	Classic infantile
c.950C>T	Unknown (disease-associated)	c.1501_1515del	Unknown (disease-associated)
c.955+1G>A	Classic infantile	c.1507del	Classic infantile
c.971dup	Classic infantile	c.1526A>T	Unknown (disease-associated)
c.982_988del	Classic infantile	c.1531C>A	Unknown (disease-associated)
c.983T>C	Classic infantile	c.1537G>A	Unknown (disease-associated)
c.994_995insTT	Unknown (disease-associated)	c.1538A>G	Classic infantile
c.1000G>T	Classic infantile	c.1551+3A>T	Unknown (disease-associated)
c.1004_1005dup	Unknown (disease-associated)	c.1551+5G>A	Unknown (disease-associated)
c.1047del	Unknown (disease-associated)	c.1559A>G	Unknown (disease-associated)
c.1560C>G	Unknown (disease-associated)	c.2096T>C	Unknown (disease-associated)
c.1579_1580del	Classic infantile	c.2109del	Unknown (disease-associated)
c.1583G>C	Unknown (disease-associated)	c.2131A>C	Classic infantile
c.1594G>A	Adult	c.2146G>C	Unknown (disease-associated)
c.1597T>G	Classic infantile	c.2153_2156delinsACGCCG	Classic infantile
c.1602_1605delinsAGG	Classic infantile	c.2182_2183del	Unknown (disease-associated)
c.1610del	Unknown (disease-associated)	c.2190-345A>G	Unknown (disease-associated)
c.1627T>G	Unknown (disease-associated)	c.2205dup	Classic infantile
c.1629C>G	Unknown (disease-associated)	c.2213G>A	Classic infantile
c.1636G>C	Unknown (disease-associated)	c.2221G>A	Classic infantile
c.1636+5G>A	Classic infantile	c.2222A>T	Unknown (disease-associated)
c.1650del	Unknown (disease-associated)	c.2234T>C	Classic infantile
c.1657C>T	Classic infantile	c.2235dup	Classic infantile
c.1681_1699dup	Unknown (disease-associated)	c.2237G>T	Unknown (disease-associated)
c.1688A>T	Unknown (disease-associated)	c.2240G>A	Unknown (disease-associated)
c.1716C>A	Classic infantile	c.2261dup	Unknown (disease-associated)
c.1721T>C	Unknown (disease-associated)	c.2294G>A	Classic infantile
c.1753_2799del	Classic infantile	c.2296T>A	Classic infantile
- c.1754+1dup	Unknown (disease-associated)	c.2297A>C	Classic infantile
c.1754+2T>C	Unknown (disease-associated)	c.2304del	Unknown (disease-associated)
c.1780C>T	Unknown (disease-associated)	c.2320G>A	Unknown (disease-associated)
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DNA nomenclature	Phenotype combined with a null allele	DNA nomenclature	Phenotype combined with a null allele
c.1784C>T	Unknown (disease-associated)	c.2331+5G>C	Classic infantile
c.1799G>C	Unknown (disease-associated)	c.2331+102del	Unknown (disease-associated)
c.1822del	Unknown (disease-associated)	c.2334_2335dup	Unknown (disease-associated)
c.1825T>G	Unknown (disease-associated)	c.2377_2378insAC	Classic infantile
c.1835A>C	Unknown (disease-associated)	c.2380dup	Unknown (disease-associated)
c.1835A>G	Unknown (disease-associated)	c.2395C>T	Unknown (disease-associated)
c.1837T>G	Unknown (disease-associated)	c.2407C>T	Unknown (disease-associated)
c.1839G>C	Unknown (disease-associated)	c.2411G>A	Classic infantile
c.1844_1846del	Unknown (disease-associated)	c.2459_2461del	Unknown (disease-associated)
c.1844G>T	Classic infantile	c.2460dup	Unknown (disease-associated)
c.1844G>A	Classic infantile	c.2474C>G	Unknown (disease-associated)
c.1847dup	Unknown (disease-associated)	c.2480A>G	Unknown (disease-associated)
c.1859C>A	Unknown (disease-associated)	c.2515C>T	Unknown (disease-associated)
c.1879_1881del	Classic infantile	c.2537C>A	Unknown (disease-associated)
c.1888+2_1888+15del	Classic infantile	c.2544del	Unknown (disease-associated)
c.1895T>C	Unknown (disease-associated)	c.2563G>C	Classic infantile
c.1895T>G	Classic infantile	c.2578G>A	Unknown (disease-associated)
c.1903A>G	Unknown (disease-associated)	c.2584G>A	Childhood
c.1913G>A	Classic infantile	c.2585del	Classic infantile
c.1944_1950del	Unknown (disease-associated)	c.2596del	Unknown (disease-associated)
c.1952dup	Unknown (disease-associated)	c.2619C>G	Unknown (disease-associated)
c.1961C>G	Unknown (disease-associated)	c.2636T>C	Classic infantile
c.2004C>A	Unknown (disease-associated)	c.2655_2656del	Unknown (disease-associated)
c.2015G>T	Unknown (disease-associated)	c.2716G>A	Unknown (disease-associated)
c.2020C>G	Unknown (disease-associated)	c.2720T>C	Unknown (disease-associated)
c.2020C>T	Unknown (disease-associated)	c.2725G>A	Unknown (disease-associated)
c.2024A>G	Classic infantile	c.2740dup	Unknown (disease-associated)
c.2040+2dup	Unknown (disease-associated)	c.2742dup	Classic infantile
c.2040+29_2190-270del	Classic infantile	c.2757del	Unknown (disease-associated)
c.2041-2A>G	Classic infantile	c.2799+5G>A	Unknown (disease-associated)
c.2051C>A	Unknown (disease-associated)	c.2800-1G>C	Classic infantile
c.2051C>G	Unknown (disease-associated)	c.2843dup	Classic infantile
c.2051C>T	Classic infantile	c.2845_2847del	Unknown (disease-associated)
c.2056_2057delinsCC	Unknown (disease-associated)		
c.2084dup	Unknown (disease-associated)		

constructs containing the missense variant of interest as described (in 't Groen et al., 2020). The activity of the GAA protein produced by the constructs was measured using 4-methylumbelliferyl- α -D-glucopyranoside (4-MU) as a substrate in transfected COS-7 cells, as

described in Kroos et al. (2008). Statistical analysis was performed using one-way analysis of variance with Tukey honestly significant difference post hoc multiple testing corrections. p < .05 was considered significant.

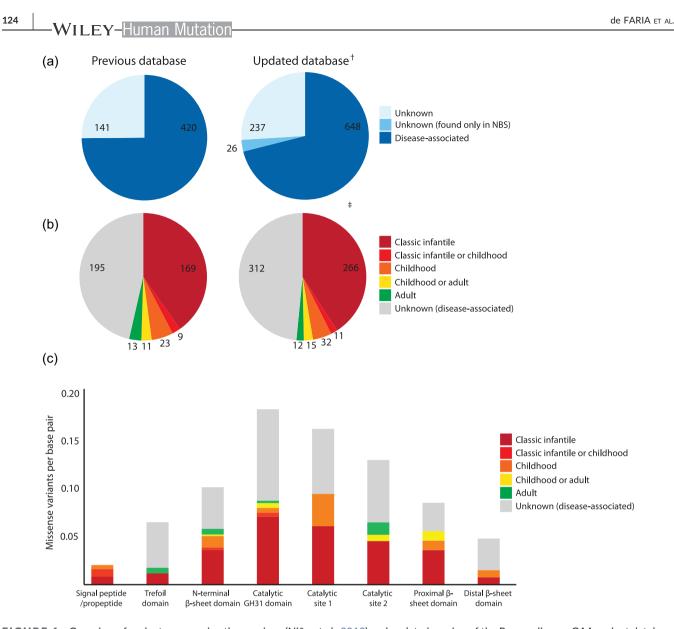


FIGURE 1 Overview of variants, comparing the previous (Niño et al., 2019) and updated version of the Pompe disease GAA variant database (http://www.pompevariantdatabase.nl). (a) Number of disease-associated and unknown variants in the previous database (left) and the updated version of the database (right). (b) Number of disease-associated variants classified based on the predicted clinical phenotype when combined with a null allele in the previous database (left) and in the updated version of the database (right). (c) Distribution of disease-associated missense variants listed in the updated database, based on the protein domains of GAA and the predicted clinical phenotype when combined with a null allele. Numbers are corrected for the length of each domain. †Two entries in the previous version of the database were removed as the variants were described twice using different nomenclatures. ‡For 36 variants listed in the previous version of the database, a reclassification of the phenotypic severity was performed due to the addition of novel patients included in this update

3 | RESULTS AND DISCUSSION

Table 1 provides an overview of the novel variants. We performed a literature search covering the past 4 years and identified 80 publications (listed in the updated database and Table S1) that described 350 novel variants, of which 226 were considered to be disease-associated (Table 1 and Figure 1a). Seventy-six novel variants (33%) were present in combination with a null allele, which allowed prediction of the clinical severity of these variants (Table 1 and Figure 1b). In addition, the inclusion of new patient information allowed us to classify the severity of 55 variants that were already present in the database. This resulted in a new total of 911 GAA variants, of which 648 were disease-associated (71%). In total, 336 out of 648 diseaseassociated variants (52%) could be associated with a clinical phenotype. The geographical or ethnical distribution of reported patients remained similar to what was described previously. The majority of patients had a Caucasian background or were of Caucasian descent (data not shown). This introduces a bias in the current version of the database and indicates the necessity of extending the database to patients of other descent. Mapping of missense variants to GAA protein domains revealed an even

TABLE 2 List of common sequence variants located within the boundaries of the GAA gene

xborn 14, S'UTR c-200G>C rs2304849 16% No effect on splicing 8.996 xborn 14, S'UTR c-178G>A rs77514632 2% No effect on splicing 9.948 xborn 18, S'UTR c-727G-G rs80020206 0.%K (3% in African population) No effect on splicing 9.999 ntron 18 c-33121XG>C rs889761 75% No effect on splicing 9.799 ntron 18 c-3313TC>T rs8077056 20% No effect on splicing 4.579 ntron 18 c-33147C>T rs5751636 31% No effect on splicing 4.564 ntron 18 c-331407L>C rs24913147 5% No effect on splicing 0.664 ntron 18 c-33141267A rs142315 5% No effect on splicing 0.644 ntron 18 c-33141007T rs12002593 10% No effect on splicing 2.604 ntron 18 c-321420471 rs4202650 33% No effect on splicing 3.993 ntron 18 c-32.2716>C rs153666739 2% No effect on splicing 3.9	Location	Variant	Variant ID	Global allele frequency (GnomAD)	Predictions of pre-mRNA splicing	CADD score PHRED
Scon 1A, S UTR c178G>A rs77514632 2% No effect on splicing 9,949 Scon 1A, S'UTR c75C>G rs80020204 0.9% (3% in African population) No effect on splicing 9,999 Intron 1B c334:316C>C rs489791 75% No effect on splicing 9,079 Intron 1B c334:316C>C rs8077055 20% No effect on splicing 8,579 Intron 1B c334:316C>C rs55751634 31% No effect on splicing 4,564 Intron 1B c334:172C> rs244:13147 5% No effect on splicing 4,974 Intron 1B c334:1172C> rs12450199 34% No effect on splicing 6,964 Intron 1B c334:1172C> rs1246219 33% No effect on splicing 2,664 Intron 1B c334:1190C>T rs1240219 33% No effect on splicing 2,684 Intron 1B c32:1298G>C rs1240210 33% No effect on splicing 2,694 Intron 1B c32:4298G>C rs12402314 76% No effect on s	Exon 1A, 5' UTR	c338C>G	rs144639114	2%	No effect on splicing	6.524
Storn 18, S ¹ UTR c7SC-G rs8002026 0.9% (3% in African population) No effect on splicing 9.989 ntron 18 c33+219C-C rs4889961 75% No effect on splicing 0.866 ntron 18 c33+317C-T rs8077055 20% No effect on splicing 9.799 ntron 18 c33+671A-C rs5575153 20% No effect on splicing 4.574 ntron 18 c33+776-A rs24451347 5% No effect on splicing 4.574 ntron 18 c33+104A-G rs1142019 34% No effect on splicing 6.76 ntron 18 c33+1172C-A rs1442315 5% No effect on splicing 1.784 ntron 18 c33+1190C>C rs1442315 5% No effect on splicing 1.784 ntron 18 c33+1905C>C rs12602510 33% No effect on splicing 2.592 ntron 18 c32+196C>C rs1260250 2% No effect on splicing 3.993 ntron 18 c32+404C>T rs12602610 33% No effect on splicing	Exon 1A, 5' UTR	c260G>C	rs2304849	16%	No effect on splicing	8.996
ntron 1B c-33+219G>C rs4889961 75% No effect on splicing 0.866 ntron 1B c-33+316C>A rs8077055 20% No effect on splicing 9.079 ntron 1B c-33+316C>A rs8077055 20% No effect on splicing 8.579 ntron 1B c-33+757C>A rs2811317 5% No effect on splicing 4.976 ntron 1B c-33+104A>G rs1150841 75% No effect on splicing 6.976 ntron 1B c-33+104A>G rs11260253 10% No effect on splicing 1.744 ntron 1B c-33+107C> rs12602593 10% No effect on splicing 1.752 ntron 1B c-33+107C> rs142214 76% No effect on splicing 1.752 ntron 1B c-32+129G>C rs16202610 33% No effect on splicing 3.993 ntron 1B c-32+129G>C rs142214 76% No effect on splicing 3.993 ntron 1B c-32-129G>C rs1520026 0.7% (3% in African population) No effect on splicing 3.993	Exon 1A, 5' UTR	c178G>A	rs77514632	2%	No effect on splicing	9.948
ntron 18 c-33+316C>A rs8077055 20% No effect on splicing 9.079 ntron 18 c-33+317C>T rs8077056 20% No effect on splicing 8.579 ntron 18 c-33+671A>C rs55751636 31% No effect on splicing 4.74 ntron 18 c-33+757C>A rs28113147 5% No effect on splicing 4.74 ntron 18 c-33+104A>C rs11450199 34% No effect on splicing 6.76 ntron 18 c-33+1102A>C rs1442315 5% No effect on splicing 1.782 ntron 18 c-33+1190C>T rs142016 33% No effect on splicing 2.825 ntron 18 c-32+1120C>T rs1420260 0.7% (3% in African population No effect on splicing 3.993 ntron 18 c-32+1120C>T rs14502606 0.7% (3% in African population No effect on splicing 3.993 ntron 18 c-32-7120>C rs147264695 0.3% (1% in Finnish population No effect on splicing 0.34 ntron 18 c-32-460A>C rs147264695 0.3% (1% in Fin	Exon 1B, 5' UTR	c75C>G	rs80020206	0.9% (3% in African population)	No effect on splicing	9.989
ntron 18 c-33+317C>T n8077054 20% No effect on splicing 8.579 ntron 18 c-33+671A>C rs55751636 31% No effect on splicing 4.974 ntron 18 c-33+7576>A rs28413147 5% No effect on splicing 4.974 ntron 18 c-33+104A>C rs12450199 34% No effect on splicing 6.976 ntron 18 c-33+1102A>C rs1150841 75% No effect on splicing 6.976 ntron 18 c-33+1102A>C rs1442315 5% No effect on splicing 1.784 ntron 18 c-33+1090A>C rs142314 76% No effect on splicing 2.604 ntron 18 c-32+1298C>C rs142314 76% No effect on splicing 2.604 ntron 18 c-32-1298C>C rs14532064 0.9% (3% in African population) No effect on splicing 4.941 ntron 18 c-32-7210>C rs5754966 2% No effect on splicing 4.349 ntron 18 c-32-240C>T rs147264095 0.3% (15 in Finnish population) No effect on splici	Intron 1B	c33+219G>C	rs4889961	75%	No effect on splicing	0.866
ntron 18c-33+671A>Crs5575163631%No effect on splicing1.456ntron 18c-33+7576>Ars284131475%No effect on splicing4.974ntron 18c-33+903A>Crs1245019934%No effect on splicing6.076ntron 18c-33+11706>Ars1115084175%No effect on splicing6.076ntron 18c-33+11702>Ars1423155%No effect on splicing1.764ntron 18c-33+11702>Ars14231476%No effect on splicing1.764ntron 18c-33+11905>Crs1260259310%No effect on splicing2.604ntron 18c-32+1296>Crs1260261033%No effect on splicing3.993ntron 18c-32-1298C>rs145262060.9% (3% in African population)No effect on splicing3.993ntron 18c-32-7216>Crs155667392%No effect on splicing4.041ntron 18c-32-7216>Crs175754662%No effect on splicing0.366ntron 18c-32-7216>Crs175754662%No effect on splicing0.364ntron 18c-32-240C>Trs12600855%No effect on splicing0.364ntron 18c-32-240C>Ars1002951%No effect on splicing0.226ntron 18c-32-447C>Grs1403255722%No effect on splicing0.226xon 2c2476>Ars14023665%No effect on splicing0.226xon 2c2476>Ars14023665%No effect on splicing <td>Intron 1B</td> <td>c33+316C>A</td> <td>rs8077055</td> <td>20%</td> <td>No effect on splicing</td> <td>9.079</td>	Intron 1B	c33+316C>A	rs8077055	20%	No effect on splicing	9.079
ntron 1Bc-33+757G>Ars284131475%No effect on splicing4.974ntron 1Bc-33+903A>Crs1245019934%No effect on splicing6.976ntron 1Bc-33+1170A>Grs1115084175%No effect on splicing0.064ntron 1Bc-33+1190G>Trs1260259310%No effect on splicing1.784ntron 1Bc-33+1190G>Trs1260259310%No effect on splicing1.784ntron 1Bc-33+1190G>Trs1260259310%No effect on splicing1.784ntron 1Bc-32+1246>Trs1895960020%No effect on splicing3.993ntron 1Bc-32-884T>Crs1453620660.9% (3% in African population)No effect on splicing3.993ntron 1Bc-32-793C>Grs55667392%No effect on splicing1.008ntron 1Bc-32-640A>Grs1472640950.3% (1% in Finnish population)No effect on splicing1.036ntron 1Bc-32-640C>Trs1200084513%No effect on splicing0.364ntron 1Bc-32-494C>Grs140325722%No effect on splicing0.226ntron 1Bc-32-494C>Grs140325722%No effect on splicing0.226ntron 1Bc-32-494C>Grs140325722%No effect on splicing0.226ntron 1Bc-32-494C>Grs140325722%No effect on splicing0.226attron 1Bc-32-494C>Grs140325722%No effect on splicing0.226attron 1Bc-32-494C>G	Intron 1B	c33+317C>T	rs8077056	20%	No effect on splicing	8.579
thron 1Bc33+903A-Crs1245019934%No effect on splicing8.196ntron 1Bc33+1104A-Grs1115084175%No effect on splicing0.064ntron 1Bc33+1190C>Trs1240257310%No effect on splicing1.784ntron 1Bc33+1190C>Trs1240257310%No effect on splicing1.784ntron 1Bc33+1097>Crs144231476%No effect on splicing2.604ntron 1Bc32-1298C>Crs1260261033%No effect on splicing2.604ntron 1Bc32-1298C>Crs1453020660.5% (3% in African population)No effect on splicing3.993ntron 1Bc32-793C>Grs556667392%No effect on splicing4.041ntron 1Bc32-793C>Grs556667392%No effect on splicing4.041ntron 1Bc32-701C>Crs757549662%Selfect on splicing4.349ntron 1Bc32-640C>Trs120004551%No effect on splicing0.366ntron 1Bc32-494C>Grs14702404950.3% (1% in Finnish population)No effect on splicing0.256oton 2c.32-494C>Grs140325722%No effect on splicing0.256oton 2c.32-494C>Grs140325722%No effect on splicing0.256oton 2c.32-494C>Grs140325722%No effect on splicing0.256oton 2c.32-494C>Grs140325722%No effect on splicing1.252ntron 1Bc32-494C>G <t< td=""><td>Intron 1B</td><td>c33+671A>C</td><td>rs55751636</td><td>31%</td><td>No effect on splicing</td><td>1.456</td></t<>	Intron 1B	c33+671A>C	rs55751636	31%	No effect on splicing	1.456
ntron 18 c33+1104A-G rs11150841 75% No effect on splicing 6.976 ntron 18 c33+1172G-A rs142315 5% No effect on splicing 0.064 ntron 18 c33+1190G>T rs12602593 10% No effect on splicing 1.784 ntron 18 c33+1309T>C rs142314 76% No effect on splicing 2.004 ntron 18 c32+124G>T rs5895960 20% No effect on splicing 3.973 ntron 18 c32-1724G>C rs1450666 0.9% (3% in African population) No effect on splicing 3.973 ntron 18 c32-721G>C rs755666739 2% No effect on splicing 4.041 ntron 18 c32-721G>C rs75754966 2% Generates a new cryptic 0.036 ntron 18 c32-40C>T rs11506025 1% No effect on splicing 0.39 ntron 18 c32-440C>G rs140325572 2% No effect on splicing 0.236 ntron 18 c32-440C>G rs14032066 5% No effect on splicing	Intron 1B	c33+757G>A	rs28413147	5%	No effect on splicing	4.974
ntron 18 c33+1172G>A rs1442315 5% No effect on splicing 0.064 ntron 18 c33+1190G>T rs12602593 10% No effect on splicing 1.782 ntron 18 c33+1309T>C rs1442314 76% No effect on splicing 2.604 ntron 18 c32-1298G>C rs12602610 33% No effect on splicing 2.604 ntron 18 c32-1124C>T rs58959600 20% No effect on splicing 3.993 ntron 18 c32-2884T>C rs145362066 0.9% (3% in African population) No effect on splicing 3.993 ntron 18 c32-793C>G rs55666739 2% No effect on splicing 4.041 ntron 18 c32-272G>C rs5754966 2% Cenesplace accepter splice 1.008 ntron 18 c32-494C>G rs147264695 0.3% (1% in Finnish population) No effect on splicing 0.439 ntron 18 c32-494C>G rs140325572 2% No effect on splicing 0.226 ntron 18 c32-494C>G rs140325572 2%	Intron 1B	c33+903A>C	rs12450199	34%	No effect on splicing	8.196
ntron 1B c-33+1190G>T rs12602593 10% No effect on splicing 1.784 ntron 1B c-33+1309T>C rs1442314 76% No effect on splicing 1.752 ntron 1B c-32-1298G>C rs12602610 33% No effect on splicing 5.825 ntron 1B c-32-1124C>T rs58959690 20% No effect on splicing 3.993 ntron 1B c-32-793C>G rs145362066 0.9% (3% in African population) No effect on splicing 3.993 ntron 1B c-32-793C>G rs5566739 2% No effect on splicing 4.041 ntron 1B c-32-721G>C rs75754966 2% No effect on splicing 0.108 ntron 1B c-32-640C>T rs147264695 0.3% (1% in Finnish population) No effect on splicing 0.136 ntron 1B c-32-440C>G rs140325572 2% No effect on splicing 0.226 ntron 1B c-32-440C>G rs140325572 2% No effect on splicing 0.226 ntron 1B c-32-440C>G rs140325572 2% No effect o	Intron 1B	c33+1104A>G	rs11150841	75%	No effect on splicing	6.976
ntron 1B c -33+13097-C rs1442314 76% No effect on splicing 1.752 ntron 1B c -32-12986-C rs12602210 33% No effect on splicing 2.604 ntron 1B c -32-1124C>T rs58959690 20% No effect on splicing 3.993 ntron 1B c -32-384T>C rs145362066 0.9% (3% in African population) No effect on splicing 3.993 ntron 1B c -32-721G>C rs75754966 2% Senerates a new cryptic 1.008 ntron 1B c -32-686A>G rs147264695 0.3% (1% in Finnish population) No effect on splicing 4.349 ntron 1B c -32-640C>T rs1200845 51% No effect on splicing 0.036 ntron 1B c -32-640C>T rs1200845 51% No effect on splicing 0.036 ntron 1B c -32-494C>G rs140325572 2% No effect on splicing 0.226 xon 2 c 271G>A rs1800299 2% No effect on splicing 0.226 xon 2 c 2442S>A rs74003606 5% No effect on sp	Intron 1B	c33+1172G>A	rs1442315	5%	No effect on splicing	0.064
Arton 18 c32-1298G>C rs12602610 33% No effect on splicing 5.825 Intron 18 c32-1124C>T rs58959690 20% No effect on splicing 5.825 Intron 18 c32-884T>C rs145362066 0.9% (3% in African populatio) No effect on splicing 3.993 Intron 18 c32-793C>G rs5566739 2% No effect on splicing 4.041 Intron 18 c32-721G>C rs75754966 2% Generates a new cryptic 1.008 Intron 18 c32-686A>G rs147264095 0.3% (1% in Finnish populatio) No effect on splicing 4.349 Intron 18 c32-640C>T rs12600845 51% No effect on splicing 0.036 Intron 18 c32-440C>G rs110500925 1% No effect on splicing 0.226 intron 18 c32-440C>G rs1002097 2% No effect on splicing 0.226 ixon 2 c.2447G>A rs1300300 72% No effect on splicing 1.252 ixon 2 c.546+293G>A rs8476710 20% No e	Intron 1B	c33+1190G>T	rs12602593	10%	No effect on splicing	1.784
ntron 18 c32.1124C>T rs58959690 20% No effect on splicing 5.825 ntron 18 c32.884T>C rs145362066 0.9% (3% in African population) No effect on splicing 3.993 ntron 18 c32.793C>G rs55666739 2% No effect on splicing 4.041 ntron 18 c32.721G>C rs75754966 2% Generates a new cryptic splice accepter site 1.008 ntron 18 c32.640C>T rs147264695 0.3% (1% in Finnish population) No effect on splicing 4.349 ntron 18 c32.640C>T rs12600845 51% No effect on splicing 0.036 ntron 18 c32.440C>G rs140325572 2% No effect on splicing 0.226 exon 2 c.271G>A rs1000292 2% No effect on splicing 0.226 exon 2 c.324T>C rs100300 72% No effect on splicing 0.226 exon 2 c.3447C>A rs1800300 72% No effect on splicing 1.252 exon 2 c.3447C>A rs3289536 0.5% (3% in East Asian popu	Intron 1B	c33+1309T>C	rs1442314	76%	No effect on splicing	1.752
Arthon 1B c32-884T>C rs145362066 0.9% (3% in African population) No effect on splicing 3.993 Intron 1B c32-793C>G rs55666739 2% No effect on splicing 4.041 Intron 1B c32-721G>C rs75754966 2% Generates a new cryptic splice accepter site 1.008 Intron 1B c32-686A>G rs147264695 0.3% (1% in Finnish population) No effect on splicing 4.349 Intron 1B c32-640C>T rs12600845 51% No effect on splicing 0.036 Intron 1B c32-2494C>G rs140305572 2% No effect on splicing 0.036 Intron 1B c32-4492C>G rs14030297 2% No effect on splicing 0.226 Intron 1B c32-4492C>G rs1800300 72% No effect on splicing 0.226 Intron 1B c32-4492C>G rs14764710 20% No effect on splicing 1.891 Intron 1B c.547-243C>G rs805426 67% No effect on splicing 1.252 Intron 2 c.547-243C>G rs806491	Intron 1B	c32-1298G>C	rs12602610	33%	No effect on splicing	2.604
ntron 1B c32-793C>G rs55666739 2% No effect on splicing 4.041 ntron 1B c32-721G>C rs75754960 2% Generates a new cryptic splice accepter site 1.008 ntron 1B c32-686A>G rs147264695 0.3% (1% in Finnish population) No effect on splicing 4.349 ntron 1B c32-640C>T rs12600845 51% No effect on splicing 0.136 ntron 1B c32-521G>T rs115060925 1% Generates a new cryptic splice donor site 0.639 ntron 1B c32-494C>G rs140325572 2% No effect on splicing 0.226 cxon 2 c.271G>A rs1800299 2% No effect on splicing 0.226 cxon 2 c.324+7C rs1800300 72% No effect on splicing 8.391 cxon 2 c.547-243C rs34746710 20% No effect on splicing 1.252 cxon 2 c.547-243C>G rs34746710 20% No effect on splicing 1.337 ntron 2 c.547-243C>G rs3065426 67% No effect o	Intron 1B	c32-1124C>T	rs58959690	20%	No effect on splicing	5.825
ntron 1B c32-721G>C rs75754966 2% Generates a new cryptic splice accepter site 1.008 ntron 1B c32-686A>G rs147264695 0.3% (1% in Finnish population) No effect on splicing 4.349 ntron 1B c32-640C>T rs12600845 51% No effect on splicing 0.036 ntron 1B c32-521G>T rs115060925 1% Generates a new cryptic splice donor site 0.639 ntron 1B c32-494C>G rs140325572 2% No effect on splicing 0.226 ntron 1B c32-494C>G rs140325572 2% No effect on splicing 0.226 ntron 1B c32-494C>G rs140325572 2% No effect on splicing 0.226 ntron 1B c32-492C>G rs140325572 2% No effect on splicing 0.226 ntron 1B c32-492C>G rs1400320572 2% No effect on splicing 0.226 ntron 2 c.271G>A rs1800209 2% No effect on splicing 1.252 ntron 2 c.547-23A rs289536 0.5% (3% in East Asi	Intron 1B	c32-884T>C	rs145362066	0.9% (3% in African population)	No effect on splicing	3.993
ntron 1B c32-686A>G rs147264695 0.3% (1% in Finnish population) No effect on splicing 4.349 ntron 1B c32-640C>T rs12600845 51% No effect on splicing 0.639 ntron 1B c32-521G>T rs115060925 1% Septed conor site 0.639 ntron 1B c32-494C>G rs11000925 2% No effect on splicing 0.226 ntron 1B c32-494C>G rs140325572 2% No effect on splicing 0.226 ntron 1B c32-494C>G rs1800299 2% No effect on splicing 0.226 ixon 2 c.324T>C rs1800300 72% No effect on splicing 0.256 ixon 2 c.447G>A rs1800300 2% No effect on splicing 1.891 ntron 2 c.547-233C rs289536 2% No effect on splicing 1.891 ntron 2 c.547-233C rs8065426 67% No effect on splicing 1.337 ntron 2 c.547-67C>G rs8065421 67% No effect on splicing 1.337	Intron 1B	c32-793C>G	rs55666739	2%	No effect on splicing	4.041
ntron 1B c32-640C>T rs12600845 51% No effect on splicing 0.136 ntron 1B c32-521G>T rs115060925 1% Generates a new cryptic splice donor site 0.639 ntron 1B c32-494C>G rs140325572 2% No effect on splicing 0.036 ntron 1B c32-462G>A rs74003606 5% No effect on splicing 0.226 ixon 2 c.271G>A rs1800299 2% No effect on splicing 0.256 ixon 2 c.324T>C rs1800300 72% No effect on splicing 8.391 ixon 2 c.324T>C rs1800300 72% No effect on splicing 1.252 ixon 2 c.447G>A rs2289536 0.5% (3% in East Asian population) No effect on splicing 1.252 intron 2 c.547-2432C>G rs8065426 67% No effect on splicing 1.367 intron 2 c.547-238T>C rs12452263 20% No effect on splicing 1.337 intron 2 c.547-39T>G rs12452721 67% No effect on splicing <td< td=""><td>Intron 1B</td><td>c32-721G>C</td><td>rs75754966</td><td>2%</td><td></td><td>1.008</td></td<>	Intron 1B	c32-721G>C	rs75754966	2%		1.008
Intron 1B c32-521G>T rs115060925 1% Generates a new cryptic 0.639 Intron 1B c32-494C>G rs140325572 2% No effect on splicing 0.036 Intron 1B c32-462G>A rs74003606 5% No effect on splicing 0.226 ixon 2 c.271G>A rs1800299 2% No effect on splicing 0.256 ixon 2 c.324T>C rs1800300 72% No effect on splicing 8.391 ixon 2 c.447G>A rs2289536 0.5% (3% in East Asian population) No effect on splicing 1.252 ixon 2 c.546+293G>A rs34746710 20% No effect on splicing 1.899 ntron 2 c.547-243C>G rs8065426 67% No effect on splicing 1.337 ntron 2 c.547-39T>C rs12452263 20% No effect on splicing 1.337 ntron 2 c.547-39T>G rs12452721 67% No effect on splicing 2.56 ntron 2 c.547-39T>G rs18423721 67% No effect on splicing 4.721 xon 3 c.596A>G rs1042393 67% No effect	Intron 1B	c32-686A>G	rs147264695	0.3% (1% in Finnish population)	No effect on splicing	4.349
ntron 1B c32-494C>G rs140325572 2% No effect on splicing 0.036 ntron 1B c32-462G>A rs74003606 5% No effect on splicing 0.226 ixon 2 c.271G>A rs1800299 2% No effect on splicing 0.256 ixon 2 c.324T>C rs1800300 72% No effect on splicing 8.391 ixon 2 c.324T>C rs1800300 72% No effect on splicing 8.391 ixon 2 c.447G>A rs1800300 72% No effect on splicing 1.252 ixon 2 c.546+293G>A rs3249536 0.5% (3% in East Asian population) No effect on splicing 1.899 ntron 2 c.547-243C>G rs8065426 67% No effect on splicing 5.667 ntron 2 c.547-238T>C rs12452263 20% No effect on splicing 1.337 ntron 2 c.547-67C>G rs8069491 67% No effect on splicing 1.337 ntron 2 c.547-24SG rs12452721 67% No effect on splicing 4.721 ixon 3 c.59A>G rs1042393 67% No effect on splicing </td <td>Intron 1B</td> <td>c32-640C>T</td> <td>rs12600845</td> <td>51%</td> <td>No effect on splicing</td> <td>0.136</td>	Intron 1B	c32-640C>T	rs12600845	51%	No effect on splicing	0.136
ntron 1B c32-462G>A rs74003606 5% No effect on splicing 0.226 ixon 2 c.271G>A rs1800299 2% No effect on splicing 0.256 ixon 2 c.324T>C rs1800300 72% No effect on splicing 8.391 ixon 2 c.324T>C rs1800300 72% No effect on splicing 8.391 ixon 2 c.447G>A rs2289536 0.5% (3% in East Asian population) No effect on splicing 1.252 ntron 2 c.546+293G>A rs34746710 20% No effect on splicing 1.899 ntron 2 c.547-243C>G rs8065426 67% No effect on splicing 5.567 ntron 2 c.547-238T>C rs12452263 20% No effect on splicing 1.337 ntron 2 c.547-39T>G rs12452263 20% No effect on splicing 1.337 ntron 2 c.547-39T>G rs12452721 67% No effect on splicing 4.721 ntron 2 c.547-47C>G rs180301 67% No effect on splicing 5.486 x	Intron 1B	c.−32-521G>T	rs115060925	1%		0.639
ixon 2 c.271G>A rs1800299 2% No effect on splicing 0.256 ixon 2 c.324T>C rs1800300 72% No effect on splicing 8.391 ixon 2 c.447G>A rs289536 0.5% (3% in East Asian population) No effect on splicing 1.252 ixon 2 c.447G>A rs289536 0.5% (3% in East Asian population) No effect on splicing 1.252 intron 2 c.546+293G>A rs34746710 20% No effect on splicing 1.899 intron 2 c.547-243C>G rs8065426 67% No effect on splicing 5.667 intron 2 c.547-67C>G rs8069491 67% No effect on splicing 1.337 intron 2 c.547-39T>G rs12452721 67% No effect on splicing 4.721 intron 2 c.547-4C>G rs3816256 67% No effect on splicing 4.721 ixon 3 c.642C>T rs1800301 18% No effect on splicing 1.805 ixon 3 c.648G>A rs1042395 67% No effect on splicing 1.805 <td>Intron 1B</td> <td>c32-494C>G</td> <td>rs140325572</td> <td>2%</td> <td>No effect on splicing</td> <td>0.036</td>	Intron 1B	c32-494C>G	rs140325572	2%	No effect on splicing	0.036
Exon 2 c.324T>C rs1800300 72% No effect on splicing 8.391 Exon 2 c.447G>A rs2289536 0.5% (3% in East Asian population) No effect on splicing 1.252 Intron 2 c.546+293G>A rs34746710 20% No effect on splicing 1.899 Intron 2 c.547-243C>G rs8065426 67% No effect on splicing 2.529 Intron 2 c.547-238T>C rs12452263 20% No effect on splicing 5.667 Intron 2 c.547-67C>G rs8065426 67% No effect on splicing 1.337 Intron 2 c.547-67C>G rs8069491 67% No effect on splicing 1.337 Intron 2 c.547-39T>G rs12452721 67% No effect on splicing 4.721 Intron 2 c.547-4C>G rs3816256 67% No effect on splicing 0.548 Intron 2 c.547-4C>G rs1042393 67% No effect on splicing 0.548 Intron 2 c.547-4C>G rs1042393 67% No effect on splicing 1.805 </td <td>Intron 1B</td> <td>c32-462G>A</td> <td>rs74003606</td> <td>5%</td> <td>No effect on splicing</td> <td>0.226</td>	Intron 1B	c32-462G>A	rs74003606	5%	No effect on splicing	0.226
ixon 2 c.447G>A rs2289536 0.5% (3% in East Asian population) No effect on splicing 1.252 intron 2 c.546+293G>A rs34746710 20% No effect on splicing 1.899 intron 2 c.547-243C>G rs8065426 67% No effect on splicing 2.529 intron 2 c.547-238T>C rs12452263 20% No effect on splicing 5.667 intron 2 c.547-67C>G rs8069491 67% No effect on splicing 1.337 intron 2 c.547-67C>G rs8069491 67% No effect on splicing 1.337 intron 2 c.547-39T>G rs12452721 67% No effect on splicing 2.78 intron 2 c.547-4C>G rs3816256 67% No effect on splicing 4.721 ixon 3 c.596A>G rs1042393 67% No effect on splicing 0.548 ixon 3 c.648G>A rs1042395 67% No effect on splicing 1.805 ixon 3 c.668G>A rs1042395 67% No effect on splicing 1.46	Exon 2	c.271G>A	rs1800299	2%	No effect on splicing	0.256
ntron 2 c.546+293G>A rs34746710 20% No effect on splicing 1.899 ntron 2 c.547-243C>G rs8065426 67% No effect on splicing 2.529 ntron 2 c.547-238T>C rs12452263 20% No effect on splicing 5.667 ntron 2 c.547-238T>C rs12452263 20% No effect on splicing 1.337 ntron 2 c.547-67C>G rs8069491 67% No effect on splicing 1.337 ntron 2 c.547-39T>G rs12452721 67% Loss of cryptic splice donor site 2.78 ntron 2 c.547-4C>G rs3816256 67% No effect on splicing 4.721 exon 3 c.596A>G rs1042393 67% No effect on splicing 0.548 exon 3 c.648C>A rs1042395 67% No effect on splicing 1.805 exon 3 c.668G>A rs1042395 67% No effect on splicing 1.46	Exon 2	c.324T>C	rs1800300	72%	No effect on splicing	8.391
ntron 2 c.547-243C>G rs8065426 67% No effect on splicing 2.529 ntron 2 c.547-238T>C rs12452263 20% No effect on splicing 5.667 ntron 2 c.547-67C>G rs8069491 67% No effect on splicing 1.337 ntron 2 c.547-39T>G rs12452721 67% Loss of cryptic splice donor site 2.78 ntron 2 c.547-4C>G rs3816256 67% No effect on splicing 4.721 fixon 3 c.596A>G rs1042393 67% No effect on splicing 0.548 fixon 3 c.642C>T rs1800301 18% No effect on splicing 1.805 fixon 3 c.668G>A rs1042395 67% No effect on splicing 1.46	Exon 2	c.447G>A	rs2289536		No effect on splicing	1.252
ntron 2 c.547-238T>C rs12452263 20% No effect on splicing 5.667 ntron 2 c.547-67C>G rs8069491 67% No effect on splicing 1.337 ntron 2 c.547-39T>G rs12452721 67% Loss of cryptic splice donor site 2.78 ntron 2 c.547-4C>G rs3816256 67% No effect on splicing 4.721 atron 3 c.596A>G rs1042393 67% No effect on splicing 0.548 atron 3 c.642C>T rs1800301 18% No effect on splicing 1.805 atron 3 c.668G>A rs1042395 67% No effect on splicing 1.46	Intron 2	c.546+293G>A	rs34746710	20%	No effect on splicing	1.899
Intron 2 c.547-67C>G rs8069491 67% No effect on splicing 1.337 Intron 2 c.547-39T>G rs12452721 67% Loss of cryptic splice donor site 2.78 Intron 2 c.547-4C>G rs3816256 67% No effect on splicing 4.721 Exon 3 c.596A>G rs1042393 67% No effect on splicing 0.548 Exon 3 c.642C>T rs1800301 18% No effect on splicing 1.805 Exon 3 c.668G>A rs1042395 67% No effect on splicing 1.46	Intron 2	c.547-243C>G	rs8065426	67%	No effect on splicing	2.529
Intron 2c.547-39T>Grs1245272167%Loss of cryptic splice donor site2.78Intron 2c.547-4C>Grs381625667%No effect on splicing4.721Exon 3c.596A>Grs104239367%No effect on splicing0.548Exon 3c.642C>Trs180030118%No effect on splicing1.805Exon 3c.668G>Ars104239567%No effect on splicing1.46	Intron 2	c.547-238T>C	rs12452263	20%	No effect on splicing	5.667
Intron 2 c.547-4C>G rs3816256 67% No effect on splicing 4.721 Exon 3 c.596A>G rs1042393 67% No effect on splicing 0.548 Exon 3 c.642C>T rs1800301 18% No effect on splicing 1.805 Exon 3 c.668G>A rs1042395 67% No effect on splicing 1.46	Intron 2	c.547-67C>G	rs8069491	67%	No effect on splicing	1.337
Exon 3 c.596A>G rs1042393 67% No effect on splicing 0.548 Exon 3 c.642C>T rs1800301 18% No effect on splicing 1.805 Exon 3 c.668G>A rs1042395 67% No effect on splicing 1.46	Intron 2	c.547-39T>G	rs12452721	67%		2.78
Exon 3 c.642C>T rs1800301 18% No effect on splicing 1.805 Exon 3 c.668G>A rs1042395 67% No effect on splicing 1.46	Intron 2	c.547-4C>G	rs3816256	67%	No effect on splicing	4.721
Exon 3c.668G>Ars104239567%No effect on splicing1.46	Exon 3	c.596A>G	rs1042393	67%	No effect on splicing	0.548
	Exon 3	c.642C>T	rs1800301	18%	No effect on splicing	1.805
ntron 3 c.692+38C>T rs2304848 3% 5.574	Exon 3	c.668G>A	rs1042395	67%	No effect on splicing	1.46
	Intron 3	c.692+38C>T	rs2304848	3%		5.574

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Location Variant ID Clobal allele frequency (GenomAD) Predictions of pre-mNA splica donue state CADD Splica donue state Intron 3 c.692-144A-5C rs204497 67% No effect on splicing 3.633 Intron 3 c.692-144A-5C rs806205 66% No effect on splicing 3.633 Intron 3 c.692-167A-5C rs806305 67% No effect on splicing 2.363 Intron 3 c.692-15715-C rs806635 67% No effect on splicing 2.363 Intron 3 c.693-3657-C rs1060515 67% No effect on splicing 2.363 Intron 3 c.693-4915-A rs12402422 67% No effect on splicing 3.629 Intron 3 c.693-441C-G rs1240249 66% No effect on splicing 3.629 Intron 3 c.693-441C-G rs1241269 66% No effect on splicing 4.416 Intron 3 c.693-441C-G rs1241269 66% No effect on splicing 2.66 Intron 3 c.693-441C-G rs1241269 66% No effect on splicing						
Intron 3 C492 H44A-G rs230447 67% No effect on splicing 3.651 Intron 3 C492-5974C-G rs0082005 67% No effect on splicing 2.301 Intron 3 C492-5974C-G rs0082051 67% No effect on splicing 2.301 Intron 3 C492-5975C- rs0082051 67% No effect on splicing 2.311 Intron 3 C493-5807C- rs1200422 67% No effect on splicing 2.321 Intron 3 C493-5957C- rs120422 67% No effect on splicing 3.629 Intron 3 C493-4910-A rs1294201 67% No effect on splicing 3.629 Intron 3 C493-414C-G rs129022 67% No effect on splicing 2.301 Intron 3 C493-414C-G rs1291249 66% No effect on splicing 2.302 Intron 3 C493-414C-G rs1291249 67% No effect on splicing 2.302 Intron 3 C493-214T-A rs119044 77% No effect on splicing 2.302 I	Location	Variant	Variant ID		•	CADD score PHRED
Intron 3 c.692+5071-C rs8082405 66% No effect on splicing 3.271 Intron 3 c.692+674G×C rs8078350 67% No effect on splicing 2.363 Intron 3 c.692+674G×C rs8078350 67% No effect on splicing 2.364 Intron 3 c.693-5867-C rs806850 67% No effect on splicing 4.133 Intron 3 c.693-5867-C rs1202422 67% No effect on splicing 3.627 Intron 3 c.693-441C-G rs12202420 67% No effect on splicing 4.143 Intron 3 c.693-441C-G rs12941289 66% No effect on splicing 4.146 Intron 3 c.693-414C-G rs1297590 67% Loss of a cryptic splice splice acceptor site 0.077 Intron 3 c.693-2167-A rs11150844 67% No effect on splicing 4.134 Intron 3 c.693-2167-A rs74003611 67% No effect on splicing 2.374 Intron 4 c.893-376-C rs7003611 67% No effect on splicing 2.37					, · ·	
Intron 3 C.692.4674C-C rs8078350 67% No effect on splicing 4.501 Intron 3 C.692.751T-C rs8068051 67% No effect on splicing 2.363 Intron 3 C.693.586C-A rs112308142 3% No effect on splicing 2.71 Intron 3 C.693.585T-C rs806855 67% No effect on splicing 3.62 Intron 3 C.693.586T-C rs1260242 67% No effect on splicing 3.62 Intron 3 C.693.441C-C rs1240240 67% No effect on splicing 4.104 Intron 3 C.693.414C-C rs12941289 66% No effect on splicing 4.104 Intron 3 C.693.413A-C rs1291289 66% No effect on splicing 4.104 Intron 3 C.693.413A-C rs12937590 67% No effect on splicing 9.666 Intron 3 C.693.413A-C rs1290251 C.597 No effect on splicing 1.54 Intron 3 C.693.413A-C rs1290251 C.597 No effect on splicing 2.374 <t< td=""><td>Intron 3</td><td>c.692+144A>G</td><td>rs2304847</td><td>67%</td><td>No effect on splicing</td><td>3.653</td></t<>	Intron 3	c.692+144A>G	rs2304847	67%	No effect on splicing	3.653
Intron 3 c.492+7511>C rs8068051 67% No effect on splicing 2.363 Intron 3 c.693-586G>A rs112308142 3% No effect on splicing 4.133 Intron 3 c.693-586T>C rs8068555 67% No effect on splicing 4.133 Intron 3 c.693-491G>A rs12908421 67% No effect on splicing 4.20 Intron 3 c.693-441C>G rs129082420 67% No effect on splicing 7.59 Intron 3 c.693-441C>G rs12941269 66% No effect on splicing 4.416 Intron 3 c.693-414C>G rs12937590 67% Loss of a cryptic splice 1.544 Intron 3 c.693-413A>G rs11937590 67% No effect on splicing 4.13 Intron 3 c.693-413A>G rs1193042 67% No effect on splicing 4.13 Intron 3 c.693-413A>G rs11930507 7% No effect on splicing 4.13 Intron 3 c.693-413A>G rs11920321 67% No effect on splicing 0.66	Intron 3	c.692+509T>C	rs8082405	66%	No effect on splicing	3.271
Intron 3 C.693-586S-A ris12308142 3% No effect on splicing 2.71 Intron 3 C.693-585T-C rs8068555 67% No effect on splicing 4.133 Intron 3 C.693-585T-C rs12602422 67% No effect on splicing 3.229 Intron 3 C.693-491G-A rs12602420 67% No effect on splicing 3.229 Intron 3 C.693-441C-G rs12602440 67% No effect on splicing 4.14 Intron 3 C.693-441C-G rs12941269 66% No effect on splicing 4.14 Intron 3 C.693-413A-G rs1297590 67% No effect on splicing 4.13 Intron 3 C.693-413A-G rs1150844 67% No effect on splicing 4.13 Intron 3 C.693-410C-T rs79849256 0.2% (3% in East Asian population) No effect on splicing 0.66 Intron 4 C.693-49C-T rs79849256 0.2% (3% in East Asian population) No effect on splicing 0.67 Intron 4 C.693-49C-T rs74984950 7% No effect	Intron 3	c.692+674G>C	rs8078350	67%	No effect on splicing	4.501
Intron 3 c.693-5857>C rs8068555 67% No effect on splicing 4.13 Intron 3 c.693-559C>T rs12602422 67% No effect on splicing 3.629 Intron 3 c.493-491C>A rs12948631 67% No effect on splicing 3.629 Intron 3 c.493-441C>G rs12941269 66% No effect on splicing 4.140 Intron 3 c.693-414C>G rs12941269 66% No effect on splicing 4.141 Intron 3 c.693-413A>G rs12937590 67% Sos of a cryptic splice 1.544 Intron 3 c.693-2167>A rs1150844 67% No effect on splicing 4.13 Intron 3 c.693-340C>T rs7984256 0.2% (3% in East Asian population) No effect on splicing 2.57 Intron 3 c.693-78C>T rs7984256 0.2% (3% in East Asian population) No effect on splicing 0.66 Intron 4 c.852G>A rs142626724 0.6% (1% in East Asian population) No effect on splicing 0.107 Intron 5 c.921A>T rs1800303 8	Intron 3	c.692+751T>C	rs8068051	67%	No effect on splicing	2.363
Intron 3 c.693.559C>T rs12602422 67% No effect on splicing 1.879 Intron 3 c.693.491G>A rs12948631 67% No effect on splicing 3.629 Intron 3 c.693.441C>G rs12941269 66% No effect on splicing 4.416 Intron 3 c.693.434C>A rs12941289 66% No effect on splicing 4.416 Intron 3 c.693.413A>G rs12937590 67% No effect on splicing 4.13 Intron 3 c.693.413A>G rs1150844 67% No effect on splicing 4.13 Intron 3 c.693.413A>G rs12937590 62% (3% in East Asian population) No effect on splicing 9.666 Intron 3 c.693.49C>T rs74003611 6% No effect on splicing 0.67 Intron 4 c.8526>A rs142626724 0.6% (1% in East Asian population) No effect on splicing 0.67 Intron 5 c.921A>T rs7400301 6% No effect on splicing 9.61 Intron 5 c.921A>T rs7400303 6% No effect on splicin	Intron 3	c.693-586G>A	rs112308142	3%	No effect on splicing	2.71
Intron 3 c. 693.491G>A rs12948631 67% No effect on splicing 3.629 Intron 3 c. 693.441C>G rs12602440 67% Loss of a cryptic splice acceptor site 7.559 Intron 3 c. 693.441C>G rs12941289 66% No effect on splicing 4.416 Intron 3 c. 693.414C>G rs12947289 66% Loss of a cryptic splice acceptor site 0.007 Intron 3 c. 693.413A>G rs1150844 67% No effect on splicing 4.13 Intron 3 c. 693.413A>G rs1150844 67% No effect on splicing 4.13 Intron 3 c. 693.416C>T rs70849256 0.2% (3% in East Asian population) No effect on splicing 0.06 Intron 4 c. 693.49C>T rs7085517 % No effect on splicing 0.06 Intron 4 c. 852G>A rs142626724 0.6% (1% in European population) No effect on splicing 0.06 Intron 4 c. 852G>A rs1290319 6% No effect on splicing 0.101 Intron 5 c. 955+12G>A rs2524188	Intron 3	c.693-585T>C	rs8068555	67%	No effect on splicing	4.133
Intron 3c.693.441C>Grs1260244067%Loss of a cryptic splice acceptor site7.559Intron 3c.693.434C>Ars1294126966%No effect on splicing4.416Intron 3c.693.414C>Grs1294128966%Loss of a cryptic splice acceptor site0.077Intron 3c.693.413A>Grs1293759067%Loss of a cryptic splice acceptor site1.544Intron 3c.693.216T>Ars1115084467%No effect on splicing4.13Intron 3c.693.44C>Trs798492560.2% (3% in East Asian population)No effect on splicing0.66Intron 3c.693.44C>Trs740036116%No effect on splicing0.66Intron 4c.852G>Ars1426267240.66(1% in European population)No effect on splicing0.067Exon 4c.852G>Ars1426267240.66(1% in European population)No effect on splicing0.067Exon 5c.921A>Trs18003038%No effect on splicing0.981Intron 5c.955+13C>Ars22524556%No effect on splicing0.716Intron 5c.955+13C>Ars222418873%No effect on splicing0.716Intron 5c.956-107G>Ars22418873%No effect on splicing0.431Intron 5c.956-107G>Ars224188767%No effect on splicing5.835Intron 6c.1075+13C>Trs71271640.7% (1% in East Asian population)No effect on splicing5.835Intron 5c.956-84C>T<	Intron 3	c.693-559C>T	rs12602422	67%	No effect on splicing	1.879
Intron 3 C.693.434C>A rs12941269 66% No effect on splicing 4.416 Intron 3 C.693.414C>G rs12941289 66% Coss of a cryptic splice acceptor site 0.077 Intron 3 C.693.414C>G rs12937590 67% Loss of a cryptic splice acceptor site 1.544 Intron 3 C.693.216T>A rs11150844 67% No effect on splicing 4.13 Intron 3 C.693.78C>T rs70949256 0.2% (3% in East Asian population) No effect on splicing 0.06 Intron 3 C.693.78C>T rs7003611 6% No effect on splicing 0.06 Intron 4 C.693.78C>T rs7003611 6% No effect on splicing 0.06 Intron 5 C.693.49C>T rs7003611 6% No effect on splicing 0.067 Intron 4 C.893.49C>T rs7805075 7% No effect on splicing 0.067 Intron 5 C.951.40C>T rs203080 6% No effect on splicing 0.067 Intron 5 C.955.15C>A rs2901190 5% No effect on spli	Intron 3	c.693-491G>A	rs12948631	67%	No effect on splicing	3.629
Intron 3c.693.414C>Grs1294128966%Loss of a cryptic splice acceptor site0.077 acceptor siteIntron 3c.693.413A>Grs1293759067%Loss of a cryptic splice acceptor site1.544Intron 3c.693.216T>Ars1115084467%No effect on splicing4.13Intron 3c.693.94C>Trs789492560.2% (3% in East Asian population)No effect on splicing9.666Intron 3c.693.94C>Trs78050757%No effect on splicing0.06Intron 3c.693.49C>Trs78050757%No effect on splicing0.067Exon 4c.8526>Ars1226267240.2% (1% in European population)No effect on splicing0.067Exon 5c.921A>Trs18003038%No effect on splicing0.981Intron 5c.955+12G>Ars22545569%No effect on splicing0.981Intron 5c.955+136C>Trs20148473%No effect on splicing0.438Intron 5c.956-107G>Ars224188673%No effect on splicing7.196Intron 6c.1075+13C>Trs1292021%No effect on splicing7.496Exon 8c.1226A>Grs200948820.07% (1% in East Asian population)No effect on splicing7.496Intron 6c.1075+13C>Trs140030467%No effect on splicing7.496Intron 5c.956-84C>Trs224188673%No effect on splicing7.496Intron 6c.1075+13C>Trs140030467%No effe	Intron 3	c.693-441C>G	rs12602440	67%	<i>,</i> , ,	7.559
Intron 3c.693-413A-Grs1293759067%Loss of a cryptic splice acceptor site1.544Intron 3c.693-216T-Ars1115084467%No effect on splicing4.13Intron 3c.693-24C-Trs798492560.22% (3% in East Asian population)No effect on splicing9.666Intron 3c.693-78C-Trs740036116%No effect on splicing0.06Intron 3c.693-78C-Trs78050757%No effect on splicing0.06Intron 4c.858-307-Crs230484566%No effect on splicing0.067Exon 4c.858+307-Crs20484566%No effect on splicing0.067Intron 5c.921A>Trs18003038%No effect on splicing0.061Intron 5c.955+12C>Ars20245569%No effect on splicing0.981Intron 5c.955+13C>Ars99011905%No effect on splicing0.981Intron 5c.955+167C>Trs224188773%No effect on splicing6.348Intron 6c.1075+13C>Trs224188773%No effect on splicing7.496Intron 6c.1075+13C>Trs412920213%No effect on splicing9.972Intron 6c.1032+Ars8180030467%No effect on splicing9.692Intron 6c.1032+Ars8140030467%No effect on splicing9.692Intron 6c.1032+Ars8140030467%No effect on splicing9.724Intron 7c.1286A/SCrs8140030467%<	Intron 3	c.693-434C>A	rs12941269	66%	No effect on splicing	4.416
intron 3 c.693-216T>A rs1150844 67% No effect on splicing 4.13 Intron 3 c.693-94C>T rs79849256 0.2% (3% in East Asian) population) No effect on splicing 9.666 Intron 3 c.693-78C>T rs74003611 6% No effect on splicing 0.06 Intron 3 c.693-78C>T rs74003611 6% No effect on splicing 0.374 Intron 4 c.893-78C>T rs74855075 7% No effect on splicing 0.374 Exon 4 c.8526>A rs124262724 0.6%(1% in European population) No effect on splicing 0.067 Intron 4 c.858+30T>C rs204845 66% No effect on splicing 0.067 Intron 5 c.921A>T rs1800303 8% No effect on splicing 9.014 Intron 5 c.955+15C>A rs9901190 5% No effect on splicing 9.081 Intron 5 c.955+167C>T rs77717164 0.7% (6% in East Asian population) No effect on splicing 6.348 Intron 5 c.956+107C>A rs2241887 73%	Intron 3	c.693-414C>G	rs12941289	66%	<i></i>	0.077
Intron 3c.693-94C>Trs798492560.2% (3% in East Asian population)No effect on splicing9.666Intron 3c.693-78C>Trs740036116%No effect on splicing0.04Intron 3c.693-78C>Trs788550757%No effect on splicing2.374Exon 4c.852G>Ars1426267240.6% (1% in European population)No effect on splicing0.067Exon 5c.921A>Trs18003038%No effect on splicing0.067Exon 5c.955+12G>Ars25245569%No effect on splicing0.981Intron 5c.955+15C>Ars90011905%No effect on splicing7.196Intron 5c.955+167C>Trs777171640.7% (6% in East Asian population)No effect on splicing6.348Intron 5c.956-107G>Ars224188773%No effect on splicing0.661Intron 6c.1075+13C>Trs72171641%No effect on splicing5.835Intron 6c.1075+13C>Trs224188773%No effect on splicing0.641Intron 6c.1075+13C>Trs180030467%No effect on splicing5.972Exon 8c.1226A>Grs81033467%No effect on splicing1.999Intron 8c.1326+430C>Trs74073770.7% (6% in East Asian 	Intron 3	c.693-413A>G	rs12937590	67%	,, ,	1.544
populationpopulationNo effect on splicing0.06Intron 3c.693-78C>Trs740036116%No effect on splicing2.374Intron 3c.693-49C>Trs788550757%No effect on splicing2.374Exon 4c.852G>Ars1426267240.6% (1% in European population)No effect on splicing0.067Intron 4c.858+30T>Crs20484566%No effect on splicing0.067Exon 5c.921A>Trs18003038%No effect on splicing9.101Intron 5c.955+13C>Ars22245569%No effect on splicing0.681Intron 5c.955+13C>Ars9011905%No effect on splicing6.348Intron 5c.955+167C>Trs77171640.7% (6% in East Asian population)No effect on splicing6.348Intron 5c.956-107G>Ars22188767%No effect on splicing6.348Intron 6c.1075+13C>Trs12920201%No effect on splicing5.772Exon 8c.1203G>Ars81030467%No effect on splicing0.061Intron 8c.1326+132G>Ars8943667%No effect on splicing1.999Intron 8c.1326+132G>Ars8143070.7% (6% in East Asian population)No effect on splicing0.061Intron 6c.1075+13C>Trs12920201%O/7% (1% in East Asian population)No effect on splicing0.061Intron 6c.1326+132G>Ars81403067%No effect on splicing0.061Intro	Intron 3	c.693-216T>A	rs11150844	67%	No effect on splicing	4.13
Intron 3 c.693.49C>T rs78855075 7% No effect on splicing 2.374 Exon 4 c.852G>A rs142626724 0.6% (1% in European population) No effect on splicing 0.095 Intron 4 c.858+30T>C rs204845 66% No effect on splicing 0.067 Exon 5 c.921A>T rs1800303 8% No effect on splicing 0.911 Intron 5 c.955+12G>A rs252455 69% No effect on splicing 0.981 Intron 5 c.955+15SC>A rs901190 5% No effect on splicing 0.348 Intron 5 c.955+167C>T rs7717164 0.7% (6% in East Asian population) No effect on splicing 0.348 Intron 5 c.956+40 rs2241887 73% No effect on splicing 5.835 Intron 6 c.1075+13C>T rs1292402 1% No effect on splicing 5.835 Intron 6 c.1075+13C>T rs1800304 67% No effect on splicing 5.835 Intron 6 c.1075+13C>T rs1800304 67% No effect on splicing	Intron 3	c.693-94C>T	rs79849256		No effect on splicing	9.666
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intron 4 c.858+30T>C rs2304845 66% No effect on splicing 0.067 Exon 5 c.921A>T rs1800303 8% No effect on splicing 9.101 Intron 5 c.955+12G>A rs2252455 69% No effect on splicing 0.981 Intron 5 c.955+15C>A rs9901190 5% No effect on splicing 0.981 Intron 5 c.955+167C>T rs77717164 0.7% (6% in East Asian population) No effect on splicing 5.835 Intron 5 c.956-107G>A rs2241887 67% No effect on splicing 0.061 Intron 5 c.956-84C>T rs129202 1% No effect on splicing 5.835 Intron 6 c.1075+13C>T rs120294882 0.07% (1% in East Asian population) No effect on splicing 5.972 Exon 8 c.1203G>A rs1800304 67% No effect on splicing 0.068 Intron 5 c.1326+43C>T rs1800304 67% No effect on splicing 1.999 Exon 8 c.1326+43G>A rs894306 67% No effect on splicing <td>Intron 3</td> <td>c.693-49C>T</td> <td>rs78855075</td> <td>7%</td> <td>No effect on splicing</td> <td>2.374</td>	Intron 3	c.693-49C>T	rs78855075	7%	No effect on splicing	2.374
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	Intron 8	c.1326+460G>A	rs12150323	2%	No effect on splicing	0.322
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	Intron 8	c.1327-356G>T	rs6565640	73%	No effect on splicing	0.258

backbainVariant D(Jonamb)splicingsplicingPHEDIntron 8c1327-32144ri403851447%No effect on splicing0.88Intron 8c1327-297A-Xri5269A-X7%No effect on splicing0.121Intron 8c1327-297A-Xri5269A-X20%No effect on splicing0.434Intron 8c1327-178A-Xri52062A20%No effect on splicing0.124Intron 8c1327-18A-Xri52005A7%No effect on splicing0.204Intron 9c1327-178A-Xri52005A7%No effect on splicing0.204Intron 9c1438-106C-Xri52042A7%No effect on splicing0.204Intron 9c1438-106C-Xri52042A7%No effect on splicing1.521Intron 10c15142C-Xri114275180% (% in African population)No effect on splicing1.732Intron 10c15144C-Xri114275180%No effect on splicing1.732Intron 11c163641174Fri11972712%No effect on splicing1.731Intron 11c163641174Fri199782013%No effect on splicing1.731Intron 11c163641174Fri199782013%No effect on splicing1.731Intron 11c163641174Fri199782013%No effect on splicing1.731Intron 11c163641174Fri199782013%No effect on splicing1.732Intron 11c16364205CFri1162513%No effect on splicing1.7		aca,				
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population population population Intron 8 c.1327.179CrA rs.2278620 20% No effect on splicing 0.143 Intron 8 c.1327.118A>G rs.200305 7% No effect on splicing 0.124 Exon 9 c.1372.18A>G rs.2278618 67% No effect on splicing 0.006 Intron 9 c.1438.108C> rs.2278618 67% No effect on splicing 6.607 Intron 9 c.1438.108C> rs.2278618 67% No effect on splicing 5.521 Intron 9 c.1438.108C> rs.220484 67% No effect on splicing 7.131 Intron 10 c.151442C>A rs.1042396 23% No effect on splicing 6.781 Exon 11 c.1636+172C>T rs.204842 5% No effect on splicing 0.045 Intron 11 c.1636+172C>T rs.2904842 5% No effect on splicing 0.811 Intron 11 c.1636+172C>T rs.19294584 11% No effect on splicing 0.811 Intron 11 c.1636+240C>T rs.	Intron 8	c.1327-269A>G	rs6565641	67%	No effect on splicing	4.207
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Intron 9 c.1438-19C>C rs2304844 67% No effect on splicing 5.29 Intron 10 c.1551+42C>A rs115427918 0.9% (3% in African population) No effect on splicing 5.792 Intron 10 c.1551+49C>A rs204843 67% No effect on splicing 6.758 Intron 11 c.1636+43G>T rs204842 5% Cenerates a new cryptic splice accepter splice 6.859 Intron 11 c.1636+117del rs19788201 59% No effect on splicing 0.045 Intron 11 c.1636+117CFT rs12945868 11% No effect on splicing 0.181 Intron 11 c.1636+117CFT rs12945868 11% No effect on splicing 0.181 Intron 11 c.1636+205C>T rs79673008 3% No effect on splicing 0.181 Intron 11 c.1636+205C>T rs71487884 5% No effect on splicing 0.181 Intron 11 c.1636+389C>G rs721675 63% No effect on splicing 0.573 Intron 11 c.1636+404A>G rs2004936 7%209921	Intron 9	c.1438-220A>G	rs2278618	67%	No effect on splicing	6.607
Intron 10 c.1551+42G>A rs115427918 0.9% (3% in African population) No effect on splicing 5.792 Intron 10 c.1551+49C>A rs2304843 67% No effect on splicing 7.131 Exon 11 c.1581G>A rs1042396 23% No effect on splicing 6.859 Intron 11 c.1636+137del rs199788201 59% No effect on splicing 0.045 Intron 11 c.1636+117del rs199788201 59% No effect on splicing 0.181 Intron 11 c.1636+117G>T rs12945868 11% No effect on splicing 0.013 Intron 11 c.1636+117G>T rs12945868 13% No effect on splicing 0.181 Intron 11 c.1636+205C>T rs79673008 3% No effect on splicing 0.131 Intron 11 c.1636+205C>T rs79487884 5% No effect on splicing 0.328 Intron 11 c.1636+389C>G rs721675 63% No effect on splicing 0.573 Intron 11 c.1636+390A>G rs7209721 63% No effect on splicin	Intron 9	c.1438-108G>A	rs12944802	67%	No effect on splicing	0.013
Intron 10 c.1551+49C>A rs204843 67% No effect on splicing 7.131 Exon 11 c.1581-SA rs1042396 23% No effect on splicing 6.859 Intron 11 c.1636+43G>T rs204842 5% Generates a new cryptic splice accepter site 6.859 Intron 11 c.1636+117cel rs199788201 59% No effect on splicing 0.045 Intron 11 c.1636+117c>T rs12945868 11% No effect on splicing 0.181 Intron 11 c.1636+118C>T rs4889817 59% No effect on splicing 0.163 Intron 11 c.1636+205C>T rs79478840 5% No effect on splicing 1.81 Intron 11 c.1636+205C>T rs11162584 2% No effect on splicing 0.573 Intron 11 c.1636+204C>C rs11151014 2% No effect on splicing 0.573 Intron 11 c.1636+204C>C rs112555 55% No effect on splicing 0.576 Intron 11 c.1637+185A>G rs2304840 6% No effect on splicing	Intron 9	c.1438-19G>C	rs2304844	67%	No effect on splicing	3.529
Exon 11 c.1581G>A rs1042396 23% No effect on splicing 6.758 Intron 11 c.1636+43G>T rs2304842 5% Generates a new cryptic splice accepter site 6.859 Intron 11 c.1636+117Cel rs199788201 59% No effect on splicing 0.045 Intron 11 c.1636+117C>T rs199788201 59% No effect on splicing 0.181 Intron 11 c.1636+117C>T rs199788201 59% No effect on splicing 0.181 Intron 11 c.1636+118C>T rs4889817 59% No effect on splicing 0.013 Intron 11 c.1636+205C>T rs7948784 5% No effect on splicing 0.828 Intron 11 c.1636+269C>T rs11152584 2% No effect on splicing 0.573 Intron 11 c.1636+284C>C rs111551014 2% No effect on splicing 0.573 Intron 11 c.1636+404A>G rs4889818 74% No effect on splicing 0.576 Intron 11 c.1637+185A>G rs12951255 55% No effect on splicing <td>Intron 10</td> <td>c.1551+42G>A</td> <td>rs115427918</td> <td>0.9% (3% in African population)</td> <td>No effect on splicing</td> <td>5.792</td>	Intron 10	c.1551+42G>A	rs115427918	0.9% (3% in African population)	No effect on splicing	5.792
Intron 11 c.1636+43G>T rs2304842 5% Generates a new cryptic splice accepter site 6.859 Intron 11 c.1636+117del rs199788201 59% No effect on splicing 0.045 Intron 11 c.1636+117C>T rs12945868 11% No effect on splicing 0.181 Intron 11 c.1636+118G>T rs4889817 59% No effect on splicing 0.103 Intron 11 c.1636+210G>A rs79673008 3% No effect on splicing 0.131 Intron 11 c.1636+228G>T rs711625854 2% No effect on splicing 3.828 Intron 11 c.1636+284G>C rs111551014 2% No effect on splicing 0.573 Intron 11 c.1636+3980 rs721675 63% No effect on splicing 0.573 Intron 11 c.1636+3980 rs720921 63% No effect on splicing 0.576 Intron 11 c.1637+185A>G rs12951255 55% No effect on splicing 0.576 Intron 11 c.1637+185A>G rs12904826 0.9% (3% in African population) <	Intron 10	c.1551+49C>A	rs2304843	67%	No effect on splicing	7.131
Intron 11 c.1636+117del rs199788201 59% No effect on splicing 0.045 Intron 11 c.1636+117C>T rs12945868 11% No effect on splicing 0.181 Intron 11 c.1636+118C>T rs489817 59% No effect on splicing 0.103 Intron 11 c.1636+210C>T rs79673008 3% No effect on splicing 0.013 Intron 11 c.1636+220C>T rs7947884 5% No effect on splicing 0.828 Intron 11 c.1636+240C>T rs11162584 2% No effect on splicing 0.573 Intron 11 c.1636+284C>C rs11151014 2% No effect on splicing 0.573 Intron 11 c.1636+284C>C rs11551014 2% No effect on splicing 0.573 Intron 11 c.1636+3430C>C rs21255 55% No effect on splicing 0.576 Intron 11 c.1637+185A rs190307 2% No effect on splicing 0.268 Intron 12 c.1756A rs230480 6% No effect on splicing 0.763	Exon 11	c.1581G>A	rs1042396	23%	No effect on splicing	6.758
Intron 11 c.1636+117C>T rs12945868 11% No effect on splicing 0.181 Intron 11 c.1636+118G>T rs4889817 59% No effect on splicing 0.013 Intron 11 c.1636+205C>T rs79673008 3% No effect on splicing 0.013 Intron 11 c.1636+205C>T rs79673008 3% No effect on splicing 0.013 Intron 11 c.1636+205C>T rs71478784 5% No effect on splicing 1.463 Intron 11 c.1636+269C>T rs111525854 2% No effect on splicing 3828 Intron 11 c.1636+389C>G rs7221675 63% No effect on splicing 0.573 Intron 11 c.1636+390A>G rs720921 63% No effect on splicing 1.829 Intron 11 c.1636+404A>G rs4889818 74% No effect on splicing 0.573 Intron 11 c.1637+185A>G rs12951255 55% No effect on splicing 0.576 Exon 12 c.1754+104A>G rs201480 6% No effect on splicing 1.422	Intron 11	c.1636+43G>T	rs2304842	5%		6.859
Intron 11 c.1636+118G>T rs4889817 59% No effect on splicing 3.161 Intron 11 c.1636+205C>T rs79673008 3% No effect on splicing 0.013 Intron 11 c.1636+205C>T rs79487884 5% No effect on splicing 1.463 Intron 11 c.1636+206C>T rs111625854 2% No effect on splicing 3.828 Intron 11 c.1636+284G>C rs111551014 2% No effect on splicing 0.573 Intron 11 c.1636+390A>G rs7221675 63% No effect on splicing 0.573 Intron 11 c.1636+390A>G rs7209921 63% No effect on splicing 0.576 Intron 11 c.1636+404A>G rs4889818 74% No effect on splicing 0.576 Intron 11 c.1637-185A>G rs12951255 55% No effect on splicing 0.576 Exon 12 c.1754+126>A rs204840 6% No effect on splicing 4.325 Intron 12 c.1754+100 <t< td=""> rs13686855 0.9% (3% in African population) No effect on splicing</t<>	Intron 11	c.1636+117del	rs199788201	59%	No effect on splicing	0.045
Intron 11 c.1636+205C>T rs79673008 3% No effect on splicing 0.013 Intron 11 c.1636+210G>A rs79487884 5% No effect on splicing 1.463 Intron 11 c.1636+269C>T rs111625854 2% No effect on splicing 3.828 Intron 11 c.1636+284G>C rs111551014 2% No effect on splicing 0.573 Intron 11 c.1636+389C>G rs720921 63% No effect on splicing 0.573 Intron 11 c.1636+404A>G rs489818 74% No effect on splicing 1.902 Intron 11 c.1637-185A>G rs12951255 55% No effect on splicing 0.573 Intron 11 c.1637-185A>G rs12951255 55% No effect on splicing 0.576 Exon 12 c.1726C>A rs1800307 2% Generates a new cryptic splice acceptor 0.268 Intron 12 c.1754+12G>A rs204840 6% No effect on splicing 0.432 Intron 12 c.1754+140C>G rs204836 0.9% (3% in African population) No effect	Intron 11	c.1636+117C>T	rs12945868	11%	No effect on splicing	0.181
Intron 11 c.1636+210G>A rs79487884 5% No effect on splicing 1.463 Intron 11 c.1636+269C>T rs111625854 2% No effect on splicing 3.828 Intron 11 c.1636+284G>C rs111551014 2% No effect on splicing 1.81 Intron 11 c.1636+284G>C rs721675 63% No effect on splicing 0.573 Intron 11 c.1636+389C>G rs7221675 63% No effect on splicing 0.573 Intron 11 c.1636+4390A>G rs720921 63% No effect on splicing 1.829 Intron 11 c.1636+404A>G rs4889818 74% No effect on splicing 0.576 Intron 11 c.1637-185A>G rs12951255 55% No effect on splicing 0.576 Exon 12 c.1754+10C>T rs1800307 2% Generates a new cryptic 0.268 Intron 12 c.1754+10C>T rs13068685 0.9% (3% in African population) No effect on splicing 8.142 Intron 12 c.1754+140C>T rs2304837 5% No effect on splicing	Intron 11	c.1636+118G>T	rs4889817	59%	No effect on splicing	3.161
Intron 11 c.1636+269C>T rs111625854 2% No effect on splicing 3.828 Intron 11 c.1636+284G>C rs111551014 2% No effect on splicing 1.81 Intron 11 c.1636+389C>G rs7221675 63% No effect on splicing 0.573 Intron 11 c.1636+390A>G rs720921 63% No effect on splicing 1.829 Intron 11 c.1636+404A>G rs4889818 74% No effect on splicing 0.576 Intron 11 c.1637-185A>G rs12951255 55% No effect on splicing 0.576 Exon 12 c.1726G>A rs1800307 2% Generates a new cryptic splice acceptor 0.576 Intron 12 c.1754+12G>A rs2304840 6% No effect on splicing 4.325 Intron 12 c.1754+100C>T rs113688685 0.9% (3% in African population) No effect on splicing 0.763 Intron 12 c.1754+104C>G rs2304839 5% No effect on splicing 2.327 Intron 12 c.1754+144C>T rs2304837 6% No effect	Intron 11	c.1636+205C>T	rs79673008	3%	No effect on splicing	0.013
Intron 11 c.1636+284G>C rs111551014 2% No effect on splicing 1.81 Intron 11 c.1636+389C>G rs7221675 63% No effect on splicing 0.573 Intron 11 c.1636+390A>G rs7209921 63% No effect on splicing 1.829 Intron 11 c.1636+404A>G rs489818 74% No effect on splicing 1.902 Intron 11 c.1637-185A>G rs12951255 55% No effect on splicing 0.576 Exon 12 c.1726G>A rs1800307 2% Generates a new cryptic splice acceptor 0.268 Intron 12 c.1754+12G>A rs2304840 6% No effect on splicing 4.325 Intron 12 c.1754+100C>T rs113686855 0.9% (3% in African population) No effect on splicing 0.763 Intron 12 c.1754+104C>G rs204830 5% No effect on splicing 1.787 Intron 12 c.1754+144C>T rs204836 61% No effect on splicing 3.378 Intron 12 c.1755-186A>G rs204836 72% No effect on	Intron 11	c.1636+210G>A	rs79487884	5%	No effect on splicing	1.463
Intron 11 c.1636+389C>G rs7221675 63% No effect on splicing 0.573 Intron 11 c.1636+390A>G rs7209921 63% No effect on splicing 1.829 Intron 11 c.1636+404A>G rs4889818 74% No effect on splicing 1.902 Intron 11 c.1637+185A>G rs12951255 55% No effect on splicing 0.576 Exon 12 c.1726G>A rs1800307 2% Generates a new cryptic splice acceptor 0.268 Intron 12 c.1754+12G>A rs2304840 6% No effect on splicing 4.325 Intron 12 c.1754+100C>T rs11368685 0.9% (3% in African population) No effect on splicing 6.142 Intron 12 c.1754+144C>T rs2304839 5% No effect on splicing 2.032 Intron 12 c.1754+144C>T rs2304839 61% No effect on splicing 3.378 Intron 13 c.188+21G>A rs2304837 6% No effect on splicing 3.378 Intron 14 c.2040+66C>T rs2304836 72% No effect on	Intron 11	c.1636+269C>T	rs111625854	2%	No effect on splicing	3.828
Intron 11 c.1636+390A>G rs7209921 63% No effect on splicing 1.829 Intron 11 c.1636+404A>G rs4889818 74% No effect on splicing 1.902 Intron 11 c.1637-185A>G rs12951255 55% No effect on splicing 0.576 Exon 12 c.1726G>A rs1800307 2% Generates a new cryptic splice acceptor 0.268 Intron 12 c.1754+10C>T rs1368685 0.9% (3% in African population) No effect on splicing 4.325 Intron 12 c.1754+10C>T rs2304840 6% No effect on splicing 6.142 Intron 12 c.1754+10C>T rs11368685 0.9% (3% in African population) No effect on splicing 8.142 Intron 12 c.1754+140C>G rs2304837 5% No effect on splicing 0.763 Intron 12 c.1755+186A>G rs62075593 2% No effect on splicing 3.378 Intron 13 c.1888+21G>A rs2304837 6% No effect on splicing 3.378 Intron 14 c.2040+66C>T rs2304836 72% <td>Intron 11</td> <td>c.1636+284G>C</td> <td>rs111551014</td> <td>2%</td> <td>No effect on splicing</td> <td>1.81</td>	Intron 11	c.1636+284G>C	rs111551014	2%	No effect on splicing	1.81
Intron 11 c.1636+404A>G rs4889818 74% No effect on splicing 1.902 Intron 11 c.1637-185A>G rs12951255 55% No effect on splicing 0.576 Exon 12 c.1726G>A rs1800307 2% Generates a new cryptic splice acceptor 0.268 Intron 12 c.1754+12G>A rs2304840 6% No effect on splicing 4.325 Intron 12 c.1754+100C>T rs113688685 0.9% (3% in African population) No effect on splicing 8.142 Intron 12 c.1754+104C>G rs2304839 5% No effect on splicing 0.763 Intron 12 c.1754+104C>G rs2304837 61% No effect on splicing 1.787 Intron 12 c.1755+186A>G rs62075593 2% No effect on splicing 3.378 Intron 13 c.1888+21G>A rs2304837 6% No effect on splicing 3.378 Intron 14 c.2040+60A>T rs2304837 7% No effect on splicing 3.54 Intron 14 c.2040+66A>T rs2304834 6% No effect on sp	Intron 11	c.1636+389C>G	rs7221675	63%	No effect on splicing	0.573
Intron 11c.1637-185A>Grs1295125555%No effect on splicing0.576Exon 12c.1726G>Ars18003072%Generates a new cryptic splice acceptor0.268Intron 12c.1754+12G>Ars23048406%No effect on splicing4.325Intron 12c.1754+100C>Trs1136886850.9% (3% in African population)No effect on splicing8.142Intron 12c.1754+100C>Trs23048395%No effect on splicing0.763Intron 12c.1754+144C>Trs230483861%No effect on splicing1.787Intron 12c.1755+186A>Grs620755932%No effect on splicing2.032Intron 13c.1888+21G>Ars230483672%No effect on splicing3.378Intron 14c.2040+20A>Grs23048367%No effect on splicing3.54Intron 14c.2040+66A>Trs23048346%No effect on splicing0.027Intron 14c.2040+66AArs23048346%No effect on splicing0.027	Intron 11	c.1636+390A>G	rs7209921	63%	No effect on splicing	1.829
Exon 12 c.1726G>A rs1800307 2% Generates a new cryptic splice acceptor 0.268 Intron 12 c.1754+12G>A rs2304840 6% No effect on splicing 4.325 Intron 12 c.1754+100C>T rs113688685 0.9% (3% in African population) No effect on splicing 8.142 Intron 12 c.1754+104C>G rs2304839 5% No effect on splicing 0.763 Intron 12 c.1754+144C>T rs2304838 61% No effect on splicing 1.787 Intron 12 c.1755+186A>G rs62075593 2% No effect on splicing 2.032 Intron 13 c.1888+21G>A rs2304837 6% No effect on splicing 3.378 Intron 14 c.2040+20A>G rs2304836 72% No effect on splicing 3.54 Intron 14 c.2040+66C>T rs2304836 6% No effect on splicing 0.027 Intron 14 c.2040+69A>G rs2304836 7% No effect on splicing 0.027 Intron 14 c.2040+69A>G rs2304836 6% No effect on splici	Intron 11	c.1636+404A>G	rs4889818	74%	No effect on splicing	1.902
Intron 12 c.1754+12G>A rs2304840 6% No effect on splicing 4.325 Intron 12 c.1754+100C>T rs113686855 0.9% (3% in African population) No effect on splicing 8.142 Intron 12 c.1754+104C>G rs2304839 5% No effect on splicing 0.763 Intron 12 c.1754+144C>T rs2304839 61% No effect on splicing 1.787 Intron 12 c.1755-186A>G rs62075593 2% No effect on splicing 2.032 Intron 13 c.1888+21G>A rs2304836 7% No effect on splicing 3.378 Intron 14 c.2040+20A>G rs2304836 7% No effect on splicing 2.163 Intron 14 c.2040+66C>T rs2304836 7% No effect on splicing 3.54 Intron 14 c.2040+66A>G rs2304836 6% No effect on splicing 0.027 Intron 14 c.2040+69A>G rs2304836 6% No effect on splicing 0.027 Intron 14 c.2040+69A>G rs2304831 6% No effect on splicing	Intron 11	c.1637-185A>G	rs12951255	55%	No effect on splicing	0.576
Intron 12 c.1754+100C>T rs113688685 0.9% (3% in African population) No effect on splicing 8.142 Intron 12 c.1754+104C>G rs2304839 5% No effect on splicing 0.763 Intron 12 c.1754+104C>T rs2304838 61% No effect on splicing 1.787 Intron 12 c.1755+186A>G rs62075593 2% No effect on splicing 2.032 Intron 13 c.1888+21G>A rs2304837 6% No effect on splicing 3.378 Intron 14 c.2040+20A>G rs2304835 72% No effect on splicing 3.54 Intron 14 c.2040+66C>T rs2304835 7% No effect on splicing 3.54 Intron 14 c.2040+66C>T rs2304834 6% No effect on splicing 0.027 Intron 14 c.2040+69A>G rs2304834 6% No effect on splicing 0.027 Intron 14 c.2040+69A>G rs2304834 6% No effect on splicing 0.027 Intron 14 c.2040+69A>G rs2304834 27% No effect on splicing	Exon 12	c.1726G>A	rs1800307	2%		0.268
Intron 12 c.1754+104C>G rs2304839 5% No effect on splicing 0.763 Intron 12 c.1754+144C>T rs2304838 61% No effect on splicing 1.787 Intron 12 c.1755+186A>G rs62075593 2% No effect on splicing 2.032 Intron 13 c.1888+21G>A rs2304837 6% No effect on splicing 3.378 Intron 14 c.2040+20A>G rs2304836 72% No effect on splicing 2.163 Intron 14 c.2040+66C>T rs2304835 7% No effect on splicing 3.54 Intron 14 c.2040+69A>G rs2304834 6% No effect on splicing 0.027 Intron 14 c.2040+66C>T rs2304834 6% No effect on splicing 0.027 Intron 14 c.2040+69A>G rs2304833 27% No effect on splicing 0.027 Intron 14 c.2041-64G>A rs2304833 27% No effect on splicing 0.371	Intron 12	c.1754+12G>A	rs2304840	6%	No effect on splicing	4.325
Intron 12 c.1754+144C>T rs2304838 61% No effect on splicing 1.787 Intron 12 c.1755-186A>G rs62075593 2% No effect on splicing 2.032 Intron 13 c.1888+21G>A rs2304837 6% No effect on splicing 3.378 Intron 14 c.2040+20A>G rs2304836 72% No effect on splicing 2.163 Intron 14 c.2040+66C>T rs2304835 7% No effect on splicing 3.54 Intron 14 c.2040+69A>G rs2304834 6% No effect on splicing 0.027 Intron 14 c.2040+69A>G rs2304833 27% No effect on splicing 0.027 Intron 14 c.2040+69A>G rs2304833 27% No effect on splicing 0.027 Intron 14 c.2041-64G>A rs2304833 27% No effect on splicing 0.371	Intron 12	c.1754+100C>T	rs113688685	0.9% (3% in African population)	No effect on splicing	8.142
Intron 12 c.1755-186A>G rs62075593 2% No effect on splicing 2.032 Intron 13 c.1888+21G>A rs2304837 6% No effect on splicing 3.378 Intron 14 c.2040+20A>G rs2304836 72% No effect on splicing 2.163 Intron 14 c.2040+66C>T rs2304835 7% No effect on splicing 3.54 Intron 14 c.2040+69A>G rs2304834 6% No effect on splicing 0.027 Intron 14 c.2040+69A>G rs2304833 27% No effect on splicing 0.027 Intron 14 c.2040+69A>G rs2304833 27% No effect on splicing 0.371	Intron 12	c.1754+104C>G	rs2304839	5%	No effect on splicing	0.763
Intron 13 c.1888+21G>A rs2304837 6% No effect on splicing 3.378 Intron 14 c.2040+20A>G rs2304836 72% No effect on splicing 2.163 Intron 14 c.2040+66C>T rs2304835 7% No effect on splicing 3.54 Intron 14 c.2040+69A>G rs2304834 6% No effect on splicing 0.027 Intron 14 c.2041-64G>A rs2304833 27% No effect on splicing 0.371	Intron 12	c.1754+144C>T	rs2304838	61%	No effect on splicing	1.787
Intron 14 c.2040+20A>G rs2304836 72% No effect on splicing 2.163 Intron 14 c.2040+66C>T rs2304835 7% No effect on splicing 3.54 Intron 14 c.2040+69A>G rs2304834 6% No effect on splicing 0.027 Intron 14 c.2041-64G>A rs2304833 27% No effect on splicing 0.371	Intron 12	c.1755-186A>G	rs62075593	2%	No effect on splicing	2.032
Intron 14 c.2040+66C>T rs2304835 7% No effect on splicing 3.54 Intron 14 c.2040+69A>G rs2304834 6% No effect on splicing 0.027 Intron 14 c.2041-64G>A rs2304833 27% No effect on splicing 0.371	Intron 13	c.1888+21G>A	rs2304837	6%	No effect on splicing	3.378
Intron 14 c.2040+69A>G rs2304834 6% No effect on splicing 0.027 Intron 14 c.2041-64G>A rs2304833 27% No effect on splicing 0.371	Intron 14	c.2040+20A>G	rs2304836	72%	No effect on splicing	2.163
Intron 14 c.2041-64G>A rs2304833 27% No effect on splicing 0.371	Intron 14	c.2040+66C>T	rs2304835	7%	No effect on splicing	3.54
	Intron 14	c.2040+69A>G	rs2304834	6%	No effect on splicing	0.027
Exon 15 c.2065G>A rs1800309 6% No effect on splicing 1.783	Intron 14	c.2041-64G>A	rs2304833	27%	No effect on splicing	0.371
	Exon 15	c.2065G>A	rs1800309	6%	No effect on splicing	1.783

			Global allele frequency	Predictions of pre-mRNA	CADD score
Location	Variant	Variant ID	(GnomAD)	splicing	PHRED
Exon 15	c.2133A>G	rs1800310	27%	No effect on splicing	1.134
Intron 15	c.2189+95C>T	rs72850840	5%	No effect on splicing	3,771
Intron 15	c.2189+263G>A	rs7221604	66%	Generates a new cryptic splice donor site	0.563
Intron 15	c.2189+510T>G	rs4889963	5%	No effect on splicing	1.444
Intron 15	c.2189+607G>A	rs112710614	7%	No effect on splicing	0.189
Intron 15	c.2189+616T>C	rs139307163	5%	No effect on splicing	1.94
Intron 15	c.2189+723G>A	rs4889819	20%	No effect on splicing	0.367
Intron 15	c.2189+729A>G	rs74737410	5%	No effect on splicing	0.498
Intron 15	c.2189+859A>G	rs4889964	5%	No effect on splicing	1.503
Intron 15	c.2189+884G>A	rs4889965	5%	No effect on splicing	0.355
Intron 15	c.2189+1153A>G	rs72850844	5%	No effect on splicing	3.687
Intron 15	c.2189+1201C>A	rs72850846	5%	No effect on splicing	2.352
Intron 15	c.2189+1208A>G	rs72850847	5%	No effect on splicing	0.367
Intron 15	c.2189+1263A>G	rs74700450	5%	No effect on splicing	2.97
Intron 15	c.2189+1290A>G	rs74003630	5%	No effect on splicing	6.015
Intron 15	c.2189+1600C>T	rs60668271	5%	No effect on splicing	0.481
Intron 15	c.2190-1531G>A	rs74702528	0.9% (3% in African population)	No effect on splicing	0.489
Intron 15	c.2190-1463G>A	rs116416508	0.9% (3% in African population)	No effect on splicing	0.328
Intron 15	c.2190-1139A>G	rs184803352	0.7% (2% in African population	No effect on splicing	0.095
Intron 15	c.2190-1005A>G	rs4889820	5%	No effect on splicing	2.452
Intron 15	c.2190-686G>A	rs12452616	19%	No effect on splicing	2.725
Intron 15	c.2190-647G>A	rs59362713	10%	No effect on splicing	0.227
Intron 15	c.2190-536G>A	rs60429724	10%	No effect on splicing	0.454
Intron 15	c.2190-490G>A	rs111477580	1%	No effect on splicing	3.101
Intron 15	c.2190-444A>G	rs4889967	73%	No effect on splicing	1.059
Intron 15	c.2190-336C>T	rs76178719	3%	No effect on splicing	1.566
Intron 16	c.2331+20G>A	rs2304832	75%	No effect on splicing	5.346
Intron 16	c.2331+24T>C	rs2304831	15%	No effect on splicing	0.204
Intron 16	c.2331+151C>T	rs111537160	2%	No effect on splicing	0.608
Intron 16	c.2332-198A>T	rs2304830	73%	No effect on splicing	3.363
Exon 17	c.2338G>A	rs1126690	72%	No effect on splicing	2.675
Exon 17	c.2446G>A	rs1800314	5%	No effect on splicing	5.793
Intron 17	c.2482-132C>T	rs113824706	0.9% (3% in African population)	No effect on splicing	0.066
Exon 18	c.2553G>A	rs1042397	57%	Weakens a cryptic splice donor site	1.241
Intron 18	c.2647-71G>C	rs4889821	5%	No effect on splicing	3.473
Exon 19	c.2780C>T	rs1800315	2%	No effect on splicing	0.222
Intron 19	c.2800-227C>G	rs9890469	66%	No effect on splicing	0.661
Intron 19	c.2800-60G>A	rs55662462	0.7% (11% in Latino population)	No effect on splicing	2.209
Exon 20, 3' UTR	c.*3G>A	rs1800317	5%	No effect on splicing	0.03

Location	Variant	Variant ID	Global allele frequency (GnomAD)	Predictions of pre-mRNA splicing	CADD score PHRED
Exon 20, 3' UTR	c.*91G>A	rs2229221	12%	No effect on splicing	6.887
Exon 20, 3' UTR	c.*223C>T	rs8132	22%	No effect on splicing	3.025
Exon 20, 3' UTR	c.*419G>T	rs7567	19%	No effect on splicing	4.17

Abbreviations: CADD, Combined Annotation-Dependent Depletion; mRNA, messenger RNA; UTR, untranslated region.

stronger enrichment in the catalytic core compared with the mapping we performed previously (Niño et al., 2019; Figure 1c).

We included in the current version of the database common sequence variants that have a MAF \geq 1% and do not cause Pompe disease. This resulted in a relative increase in the number of nondiseaseassociated variants (Table 2). We decided to include common sequence variants in response to the misreporting of these variants as the principal cause of disease in several patients. Examples of this are the c.547-67C>G (rs8069491) and 547-39T>G (rs12452721) variants, which were reported as the cause of disease while having an allele frequency of 67% in the global population (Bekircan-Kurt et al., 2017; Guevara-Campos et al., 2019). In total, the database now includes 148 variants with a MAF \geq 1%. All variants had a low CADD score (<10; Table 2) and were classified as "unknown." We note that while these common sequence

(a)

Variant	Protein change	phenotype combined with a null allele	reported patients	Predictions on pre-mRNA splicing	CADD score PHRED
GAA + c.1597T>C	p.(Cys533Arg)	Classic infantile	1	no effect on splicing	25.5
GAA + c.307T>G	p.(Cys103Gly)	Classic infantile	11	loss of a cryptic splice donor site	25.1
GAA + c.309C>G	p.(Cys103Trp)	Unknown	1	no effect on splicing	5.6
GAA + c.655G>A	p.(Gly219Arg)	Classic infantile	14	no effect on splicing	28.2
GAA + c.670C>T	p.(Arg224Trp)	Classic infantile or Childhood	7	no effect on splicing	22.8
GAA + c.1655T>C	p.(Leu552Pro)	Classic infantile	41	no effect on splicing	29.9
GAA + c.1798C>T	p.(Arg600Cys)	Classic infantile	18	no effect on splicing	27.0

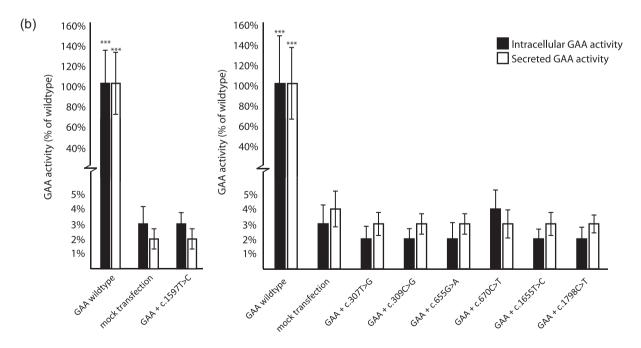


FIGURE 2 Expression study of seven disease-associated missense variants in the GAA gene. (a) Overview of basic information regarding the pathogenicity of selected variants. (b) Measured GAA activity in both cells and medium of COS-7 cultures after transfection with the generated constructs. Findings for the c.1597T>C variants are plotted separately as this was performed in a separate experiment. Data represent means, error bars represent *SD* (*n* = 3 biological replicates), ****p* < .001. CADD, Combined Annotation-Dependent Depletion; mRNA, messenger RNA

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Variant	Protein change	Location	Type of variant (protein)	MAF	Predictions on splicing-Align GVGD-SIFT-Mutation taster-[CADD score]	Experimental data	Country and reference
c.317G>A*	p.(Arg106His)	Exon 2	Missense	MAF <1%	No effect on splicing-Class CO-Deleterious-Disease causing-[25.9]		Japan; Momosaki et al. (2019)
c.365T>A	p.(Met122Lys)	Exon 2	Missense	MAF not reported	No effect on splicing-Class CO-Tolerated-Polymorphism-[14.17]		USA; Scott et al. (2013)
c.424_440del	p.(Ser142Leufs*29)	Exon 2	Frameshift	MAF not reported	No effect on splicing-Results in an out of frame product-[32]		Taiwan; Chien et al. (2011)
c.533G>A*	p.(Arg178His)	Exon 2	Missense	MAF <1%	No effect on splicing-Class CO-Tolerated-Disease causing-[31]	No effect on splicing of exon 2 in minigene construct (Goina, et al., 2019)	Taiwan; Chien et al. (2011)
c.546+5G>T*	p.?	Intron 2	No category (splicing)	MAF <1%	Weakens exon 2 splice donor and generates a cryptic splice donor-[23.7]	Affects splicing of exon 2 in minigene construct (Goina, et al., 2019)	Taiwan; Labrousse et al. (2010)
c.705G>A	p.(=)	Exon 4	Silent	MAF <1%	No effect on splicing-[0.534]		Japan; Momosaki et al. (2019)
c.811A>G*	p.(Thr 271Ala)	Exon 4	Missense	MAF not reported	No effect on splicing-Class CO-Tolerated-Polymorphism-[16.93]	71% residual activity of GAA in expression study (Kroos, et al., 2012a)	Taiwan; Labrousse et al. (2010)
c.1054C>T	p.(Gln352*)	Exon 6	Nonsense	MAF not reported	No effect on splicing-Introduces an early stop codon-[43]		Taiwan; Liao et al. (2014)
c.1080C>G	p.(Tyr360*)	Exon 7	Nonsense	MAF not reported	No effect on splicing-Introduces an early stop codon-[39]		Taiwan; Chien et al. (2011)
c.1082C>A	p.(Pro361Arg)	Exon 7	Missense	MAF <1%	No effect on splicing-Class C65-Deleterious-Disease causing-[25.5]		Japan; Momosaki et al. (2019)
c.1220A>G	p.(Tyr407Cys)	Exon 8	Missense	MAF <1%	No effect on splicing-Class C65-Deleterious-Disease causing-[25.9]		Mexico; Navarrete- Martínez et al. (2017)
c.1244C>T	p.(Thr415Met)	Exon 8	Missense	MAF <1%	No effect on splicing-Class C15-Deleterious-Disease causing-[24.6]		Japan; Momosaki et al. (2019)
c.1324G>A*	p.(Val442Met)	Exon 8	Missense	MAF <1%	No effect on splicing-Class CO-Deleterious-Disease causing-[23.8]		Taiwan; Chien et al. (2011)
c.1409A>C	p.(Asn470Thr)	Exon 9	Missense	MAF <1%	No effect on splicing-Class C25-Deleterious-Disease causing-[23.2]		Hungary; Witmann et al. (2012)

TABLE 3 Variants of unknown significance that were found only through newborn screening programs

Variant	Protein change	Location	Type of variant (protein)	MAF	Predictions on splicing-Align GVGD-SIFT-Mutation taster-[CADD score]	Experimental data	Country and reference
c.1574T>A	p.(Phe525Tyr)	Exon 11	Missense	MAF not reported	No effect on splicing-Class C15-Deleterious-Disease causing-[28.8]	10% residual activity of GAA in expression study (Kroos, et al., 2012a)	Taiwan; Chien et al. (2011)
c.1805C>T	p.(Thr 602lle)	Exon 13	Missense	MAF not reported	No effect on splicing-Class C0-Tolerated-Disease causing-[24.1]		USA; Elliott et al. (2016)
c.1840A>G	p.(Thr 614Ala)	Exon 13	Missense	MAF not reported	No effect on splicing-Class C55-Deleterious-Disease causing-[24.3]		Taiwan; Liao et al. (2014)
c.1925T>A	p.(Val642Asp)	Exon 14	Missense	MAF not reported	No effect on splicing-Class C45-Deleterious-Disease causing-[29.2]		USA; Scott et al. (2013)
c.1958C>A	p.(Thr 653Asn)	Exon 14	Missense	MAF <1%	No effect on splicing-Class C15-Tolerated-Disease causing-[25.4]		Taiwan; Chien et al. (2011)
c.2003A>G*	p.(Tyr668Cys)	Exon 14	Missense	MAF not reported	No effect on splicing-Class C65-Deleterious-Disease causing-[31]		Japan; Momosaki et al. (2019)
c.2055C>G	p.(Tyr685*)	Exon 15	Nonsense	MAF not reported	No effect on splicing-Introduces an early stop codon-[36]		Japan; Momosaki et al. (2019)
c.2174G>A	p.(Arg725Gln)	Exon 15	Missense	MAF <1%	No effect on splicing-Class C0-Tolerated-Disease causing-[32]		Hungary; Witmann et al. (2012)
c.2482-5T>C*	p.?	Intron 17	No category (splicing)	MAF not reported	No effect on splicing-[8.409]		Taiwan; Liao et al. (2014)
c.2482-2A>G	p.?	Intron 17	No category (splicing)	MAF <1%	Loss of exon 18 splice acceptor site-[35]		Hungary; Witmann et al. (2012)
c.2647-23del	p.?	Intron 18	No category (intron variant)	MAF <1%	No effect on splicing-[0.451]		Taiwan; Liao et al. (2014)
c.2843dup	p.(Val949Argfs*69) Exon 20	Exon 20	Frameshift	MAF not reported	No effect on splicing-Results in an out of frame product-[23.1]		Taiwan; Liao et al. (2014)
Abbraviations.	Abhraviations: CADD Combined Annotation Denendent Denletion: MAE minor allele frequency	notation De	mendent Denletion.	MAE minor alle			

Abbreviations: CADD, Combined Annotation-Dependent Depletion; MAF, minor allele frequency.

*Variants found in cis with the Asian pseudodeficiency allele c[1726G>A; 2065G>A].

TABLE 3 (Continued)

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variants do not result in clinical manifestation of Pompe disease, it remains possible that they might modify disease progression when present in cis with a disease-associated variant. In Pompe disease, this is the case for the Asian pseudodeficiency allele (c.[1726G>A (p.Gly576-Ser);2065G>A (p.Glu689Lys)]) and GAA2 (c.271G>A, (p.Asp91Asn)), which have a MAF of 14% for c.1726G>A, 23.5% for c.2065G>A (both East Asian), and 3.2% for GAA2 (European), and can be present in cis with known disease-associated variants (Kroos et al., 2006; Labrousse et al., 2010). Also, a variant with a low MAF in the general population, c.510C>T (p.=) (rs564758226), is known to be linked to the late-onset variant c.-32-13T>G (p.[=,0]) (IVS1). c.510C>T has a global MAF of 0.005%, but a MAF of 27.3% in compound heterozygous IVS1 patients with symptom onset at childhood. It worsens aberrant splicing caused by IVS1 and causes lower levels of leaky wild-type splicing and lower GAA enzyme activity, resulting in accelerated disease onset (Bergsma et al., 2019).

Figure 2a,b shows the results on the GAA variants we subjected to a more in-depth investigation. We selected the common missense variants c.307T>G (p.Cys103Gly), c.655G>A (p.Gly219Arg), c.670C>T (p.Arg224Trp), c.1655T>C (p.Leu552Pro), and c.1798C>T (p.Arg600-Cys) and performed in vitro analysis of their severity using SDM of GAA cDNA expression constructs. In addition, c.1597T>C (p.Cys533Arg) and c.309C>G (p.Cys103Trp) were tested due to a request for diagnostic purposes. All of these variants fully abrogated GAA enzymatic activity following transfection in COS-7 cells (Figure 2, compare mutant GAA with mock transfections). The c.309C>G variant was included because the patient that harbored this variant in combination with c.525del p.(Glu176Argfs*45) showed an atypical Pompe disease phenotype (Mori et al., 2017). This case report described an adult patient with cardiomyopathy. Molecular analysis of primary skin fibroblasts identified a reduction in GAA activity, although not at pathogenic levels, and GAA activity was in the normal range for skeletal muscle tissue (Mori et al., 2017). We note that the c.309C>G variant was not detected in DNA from either parent and was described as a de novo variant (Mori et al., 2017). This variant might have been introduced during embryonic development, resulting in mosaicism similar to, as described previously in Labrijn-Marks et al. (2019) and in 't Groen et al. (2020). This might explain the "uneven pattern" of glycogen accumulation in histological sections derived from cardiac tissue (Mori et al., 2017). The in vitro analysis indicated that the c.309C>G variant is fully deleterious and has a predicted classic infantile phenotype in combination with a null allele. A comprehensive genetic analysis would be necessary to confirm this hypothesis.

Novel variants that have been reported only through NBS studies, but for which no clinical phenotype has been provided, were classified as "Unknown (found only in NBS)". In the current version of the database, 26 variants have been classified as such (Table 3). Seven out of 26 variants were also present in *cis* with the Asian pseudodeficiency allele, indicating that additional testing is required because the Asian pseudodeficiency is known to result in false-positive outcomes in dried blood spot-based assays (Liao et al., 2014; Momosaki et al., 2019). It is currently unknown at what age symptoms will develop in neonates diagnosed with disease-associated variants that are potentially associated with a late-onset phenotype. Symptoms might be delayed until late adulthood or, for some genetic variants, might not even lead to disease. In these cases, further research on the effect of the genetic variants is essential to better inform patients, families, and doctors. As reported, in these cases, the uncertainty of the diagnosis, the possibility of an emerging disease, and the doubt on when to start treatment with ERT could lead to emotional stress (Bodamer et al., 2017). This underscores the importance of phenotype prediction for disease-associated variants, especially in the case of asymptomatic patients identified through NBS programs.

The sharp increase in reports on patients with Pompe disease and GAA disease-associated variants highlights the need for regular updates of the Pompe disease GAA variant database. Increased awareness and improved diagnostic technology with exome and genome sequencing and NBS programs are expected to further increase the number of entries in the database in the coming years. It will be important to link variants to clinical information and to test their deleterious effect in vitro using expression and splicing assays. Curated disease-specific databases such as the Pompe disease GAA variant database will be important to provide guidance to clinicians and clinical geneticists to establish an accurate molecular diagnosis.

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CONFLICT OF INTERESTS

Ans T. van der Ploeg has provided consulting services for various industries in the field of Pompe disease under an agreement between these industries and Erasmus MC, Rotterdam, the Netherlands. The remaining authors declare that there are no conflict of interests.

WEB RESOURCES

Pompe disease GAA variant database: http://www.pompevariant database.nl/

LOVD: http://gaa.lovd.nl/ GnomAD: https://gnomad.broadinstitute.org/ dbSNP: https://www.ncbi.nlm.nih.gov/snp/ CADD score: https://cadd.gs.washington.edu/

DATA AVAILABILITY STATEMENT

The data described in this study is available upon request from the corresponding authors, and new variants have been added to the Pompe disease GAA variant database (http://www.pompevariant database.nl/) and LOVD (http://gaa.lovd.nl/).

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SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.

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