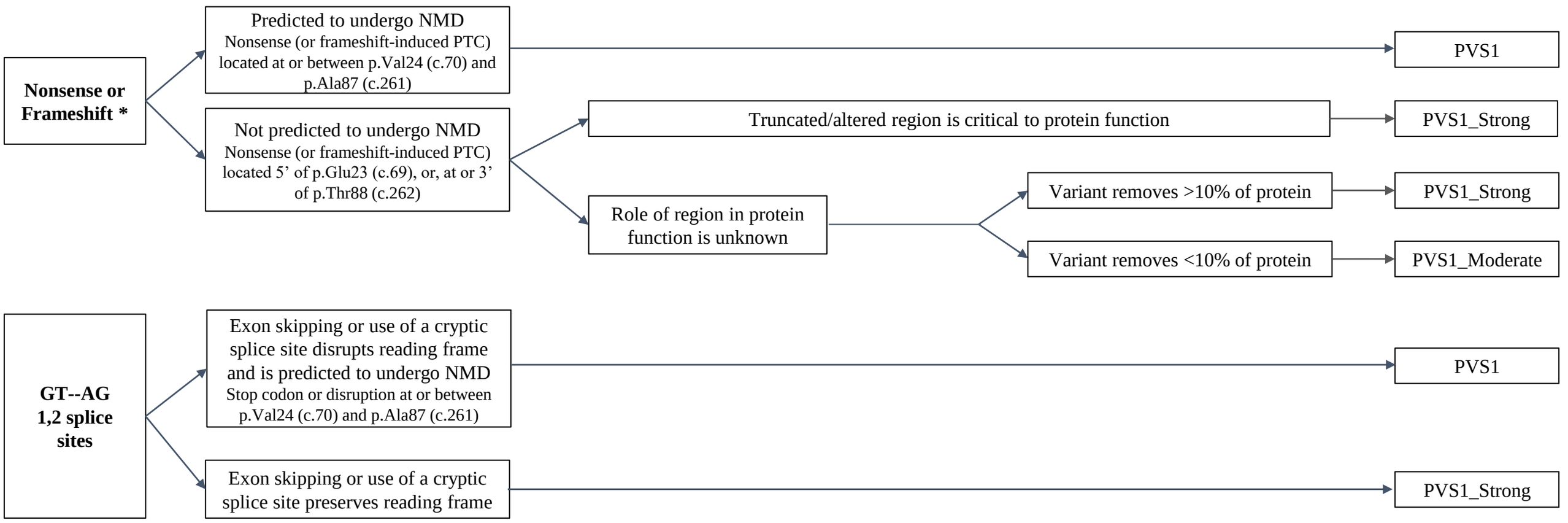
## *HBB*-specific recommendations for application of PVS1. Adapted from Tayoun et al. 2018




*NMD boundaries for *HBB* are published by Peixeiro et al. 2011 (doi: 10.3324/haematol.2010.039206) and shown below.

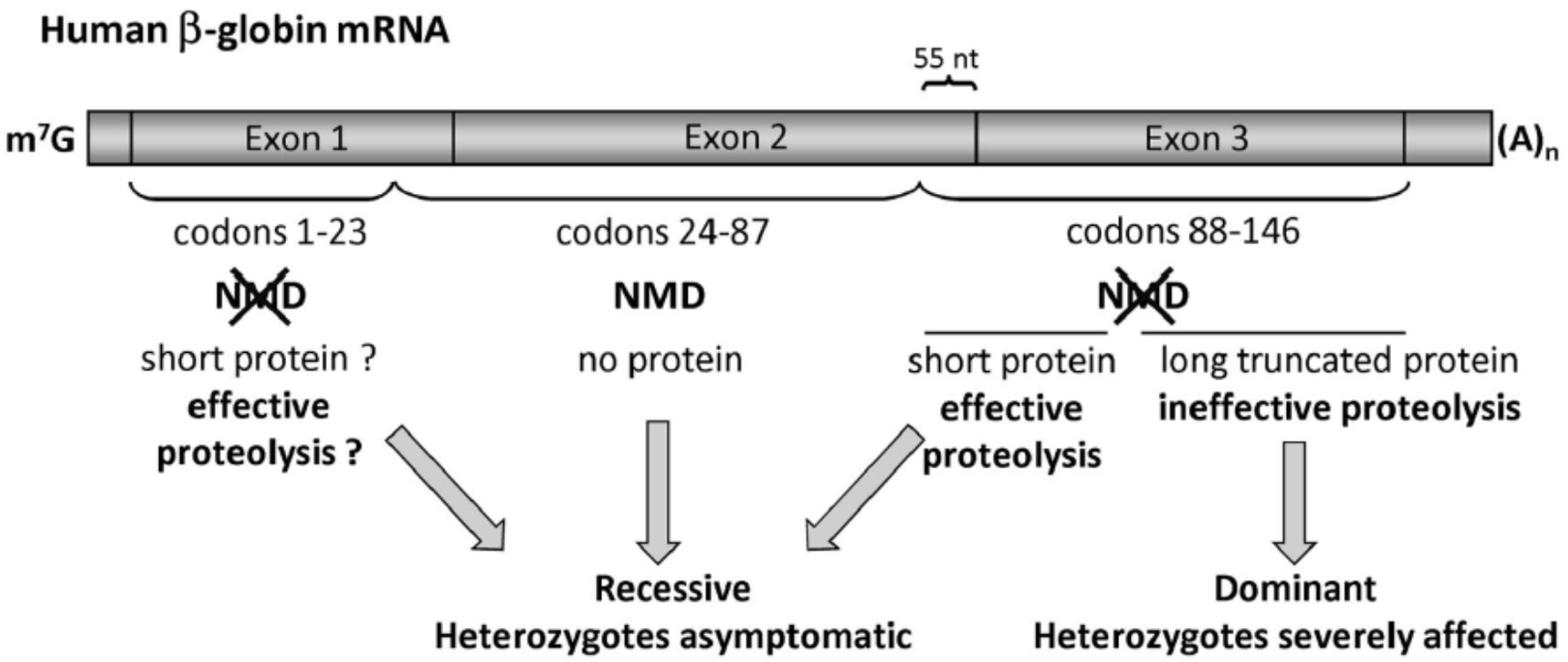



Figure 2. Nonsense-mediated mRNA decay (NMD) modulates disease phenotype. Map of the NMD-resistant and NMD-sensitive regions in the human β-globin mRNA. AUG-proximal premature termination codons (PTCs) do not trigger NMD but heterozygotes are asymptomatic, as the translated short β-globin peptides, along with the α-globin chains in excess, are effectively degraded. If the PTC is located downstream of codon 23 and more than 55 nucleotides (nt) upstream of the last exon-exon junction, the corresponding transcript is targeted for NMD and heterozygotes are also protected from thalassemia. In contrast, transcripts bearing PTCs located less than 55 nt upstream of the last exon-exon junction, or in the 5'-part of exon 3, escape NMD, resulting in the production of truncated proteins that are small enough to be efficiently degraded, along with the α-globin chains in excess, and heterozygotes are still asymptomatic for β-thalassemia. However, if the PTC is located further downstream, the encoded truncated nonfunctional β-globin proteins overwhelm the cellular proteolytic system and cause toxic precipitation of insoluble globin chains. Heterozygotes with these mutations are affected with dominantly inherited β-thalassemia intermedia.