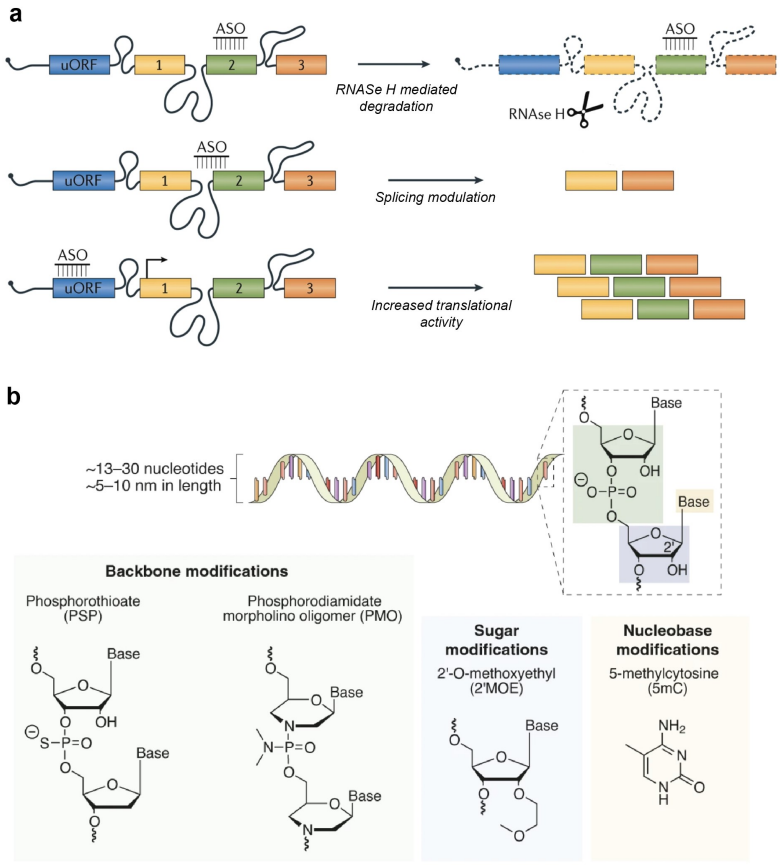


Figure 1: a) Mechanisms of action of antisense oligonucleotides (ASOs). When bound to the target RNA, ASOs can form a hybrid double helix that becomes a substrate for RNase H, which results in target mRNA degradation. ASO binding to splice sites can trigger skipping or inclusion of the targeted exon. Alternatively, when translation of the upstream open reading frames (uORFs) inhibits the expression of the primary ORF, ASOs binding to the uORFs can increase the amounts of protein translated from a downstream ORF. Adapted from [8]. b) Backbone, sugar, nucleobase modifications of approved ASOs enhance affinity to target RNA, improve nuclease resistance, and modulate immunological properties. Adapted from [9].

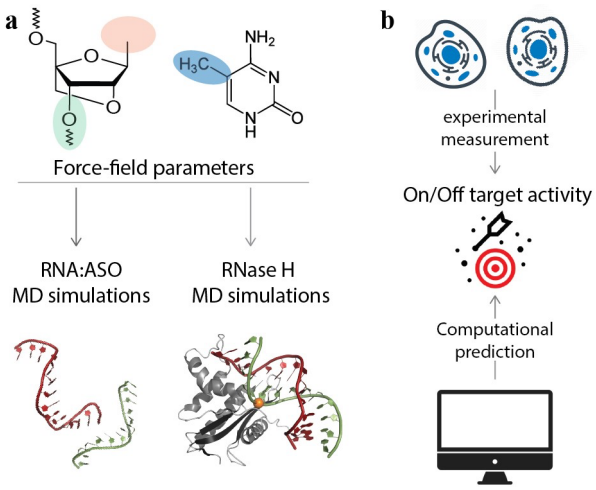


Figure 2: Schematic overview of the proposed project. a) Force-field parameters for chemically modified nucleic acids will be calculated for the most common modifications in commercially available ASOs. These parameters will enable the use of atomistic MD simulations to study structure and dynamics of generic biomolecular systems including RNA:ASO double helices in solution and bound to RNase H enzyme. b) In vitro activity of ASOs on intended and unintended target genes will be measured by RNA sequencing. At the same time, we will develop modeling techniques to predict these quantities computationally.

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