

# KORRO BIO

Unlocking the OPERA Platform

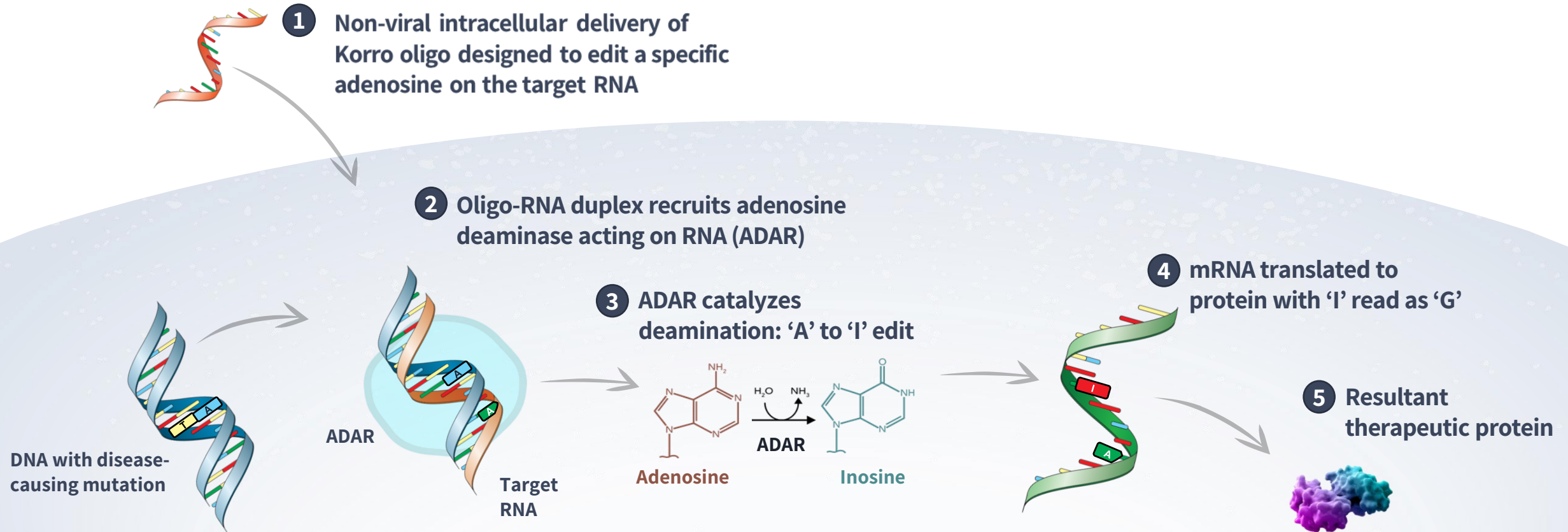
**Development and Application of Graph  
Neural Networks to Design Guide  
Oligonucleotides that Promote RNA Editing**



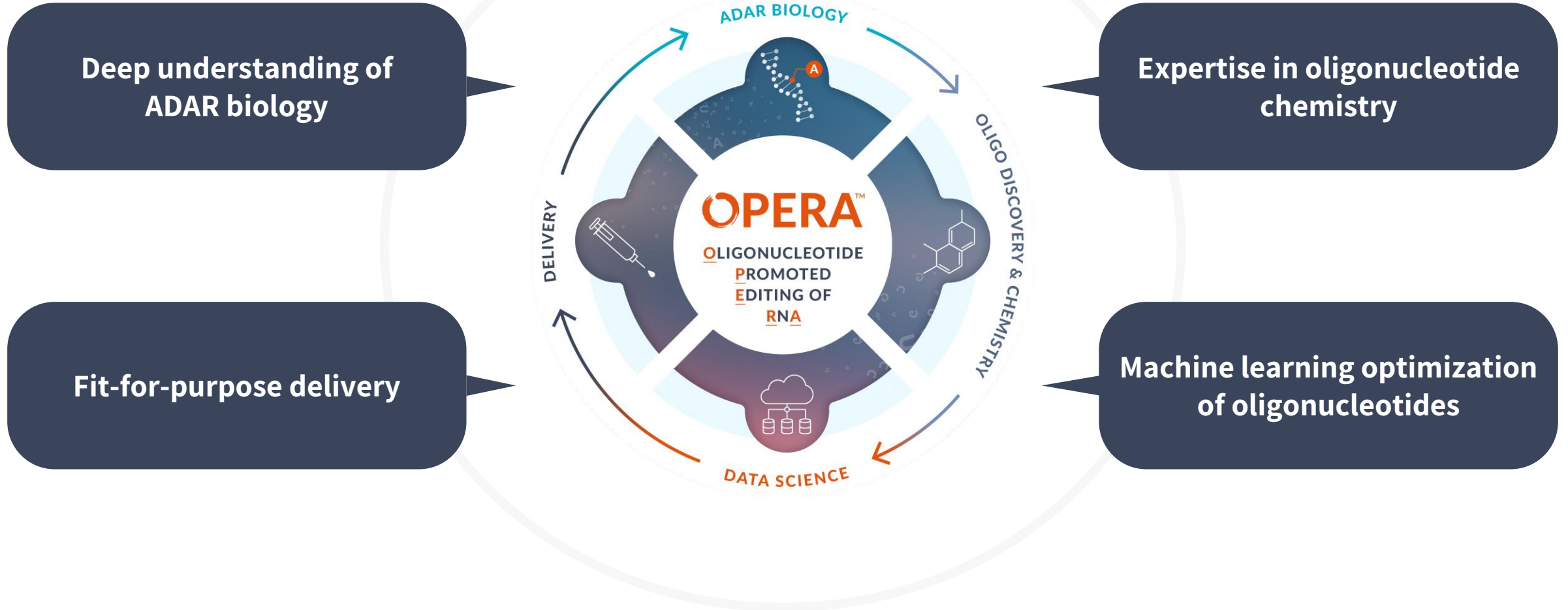
# Overview of Topics

1. **Korro Bio Methodology**
2. Machine Learning for Chemically Modified Oligonucleotide Design
3. Methods for Increasing Use-Cases for ML Oligonucleotide Design
  - New Chemical Modifications
  - New Targets

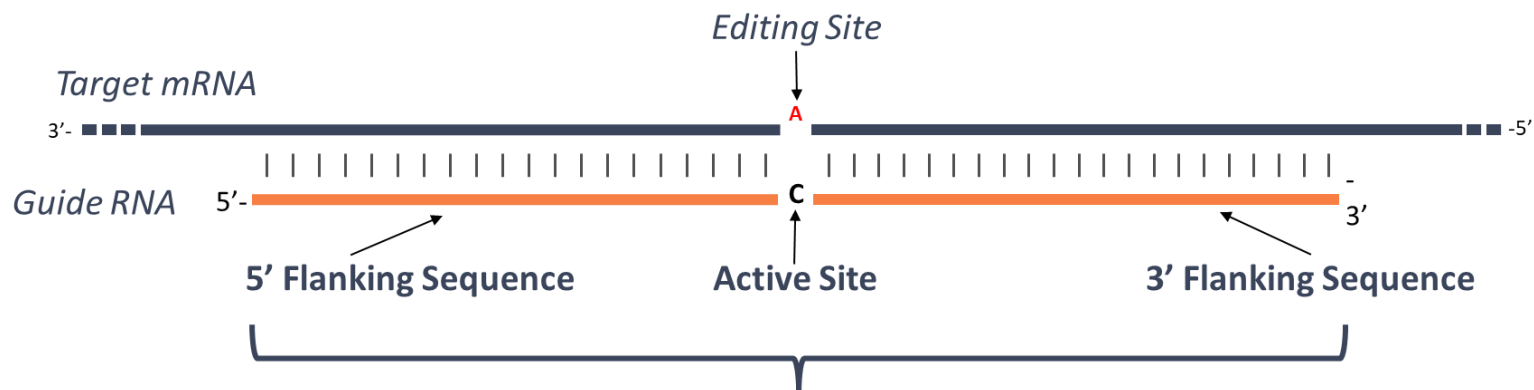
# RNA Editing: Transiently Effecting an A-to-I Edit on RNA Using an Oligonucleotide



# OPERA: Our Differentiated Approach for RNA Editing



# Chemical Modifications are Employed to Drive Activity and Stability

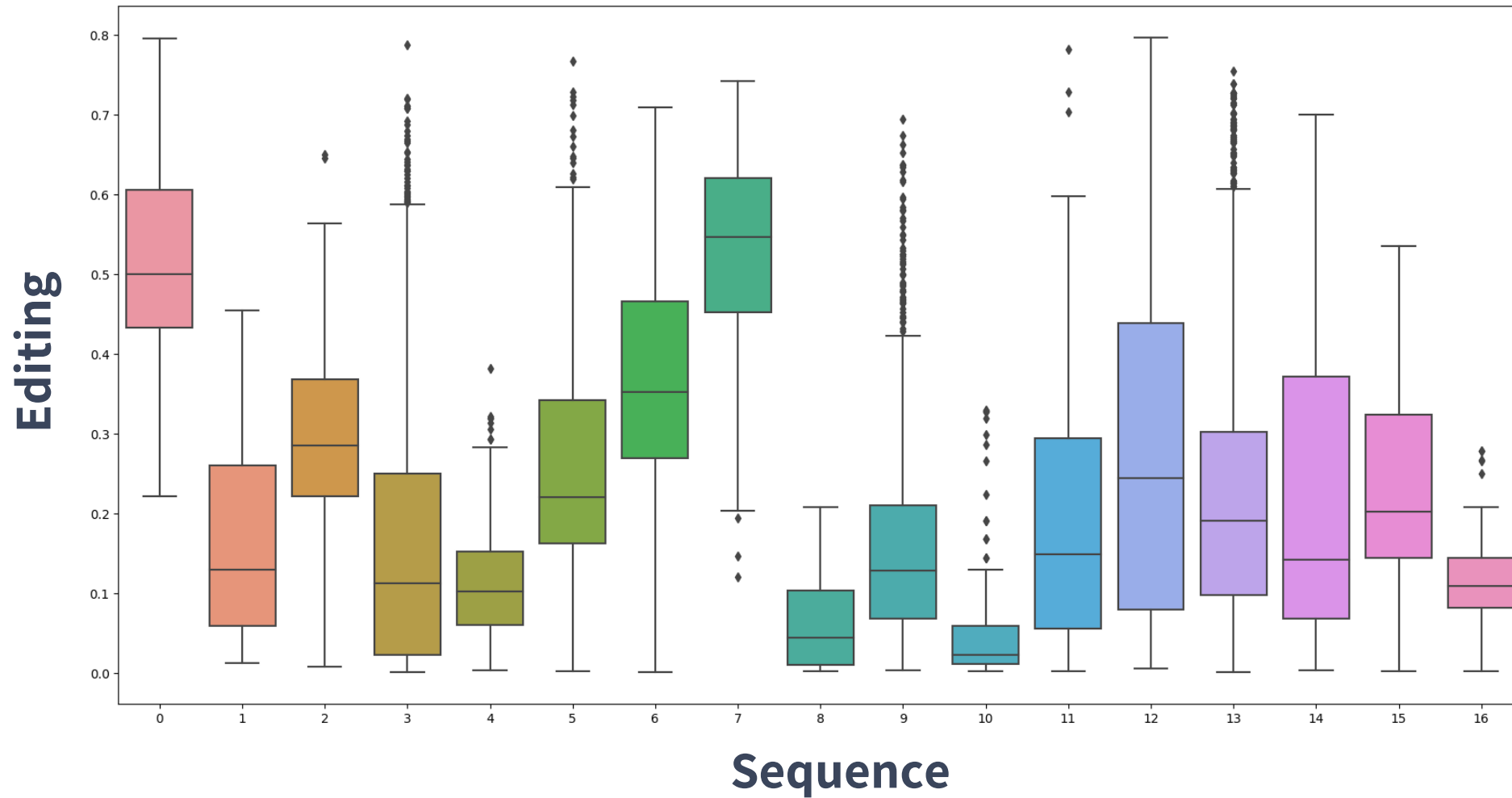


Examples of chemical modifications in approved products

*Korro leverages established and novel chemical modifications to improve pharmacology of ADAR guide oligonucleotides*

- (N) DNA
- (N) RNA
- (N) PMO
- (N) 2'-O-methoxyethyl
- (N) 2'-O-methyl
- (N) 2'-Fluoro
- Y 5-Methyl pyrimidine
- Phosphorothioate
- Phosphodiester

# Chemical Modification Pattern Significantly Impacts Editing of Target mRNA

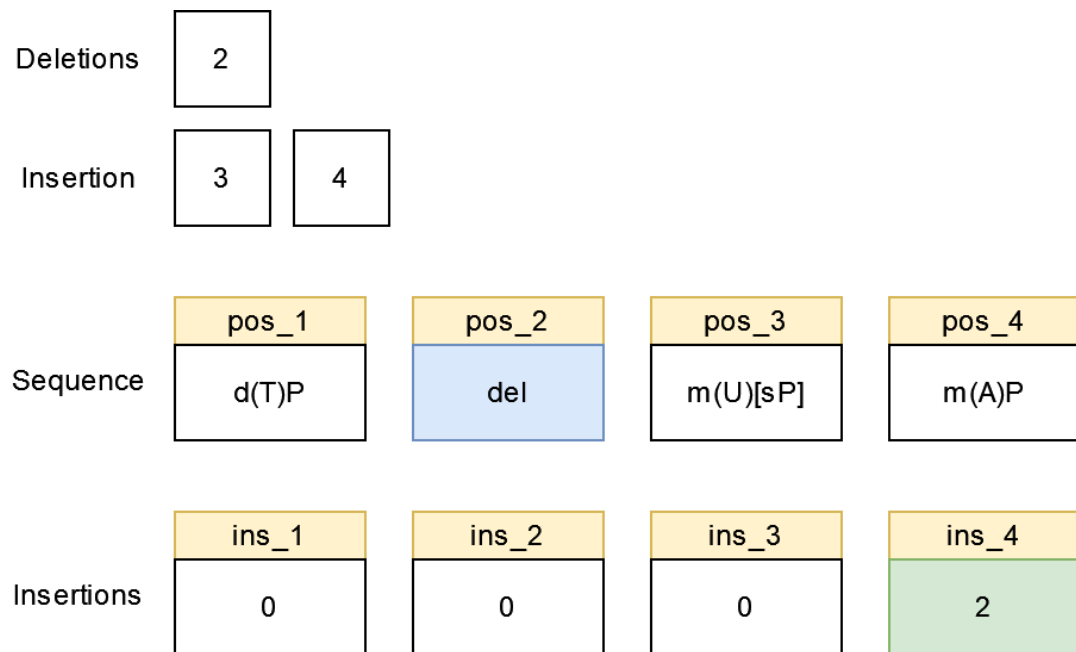


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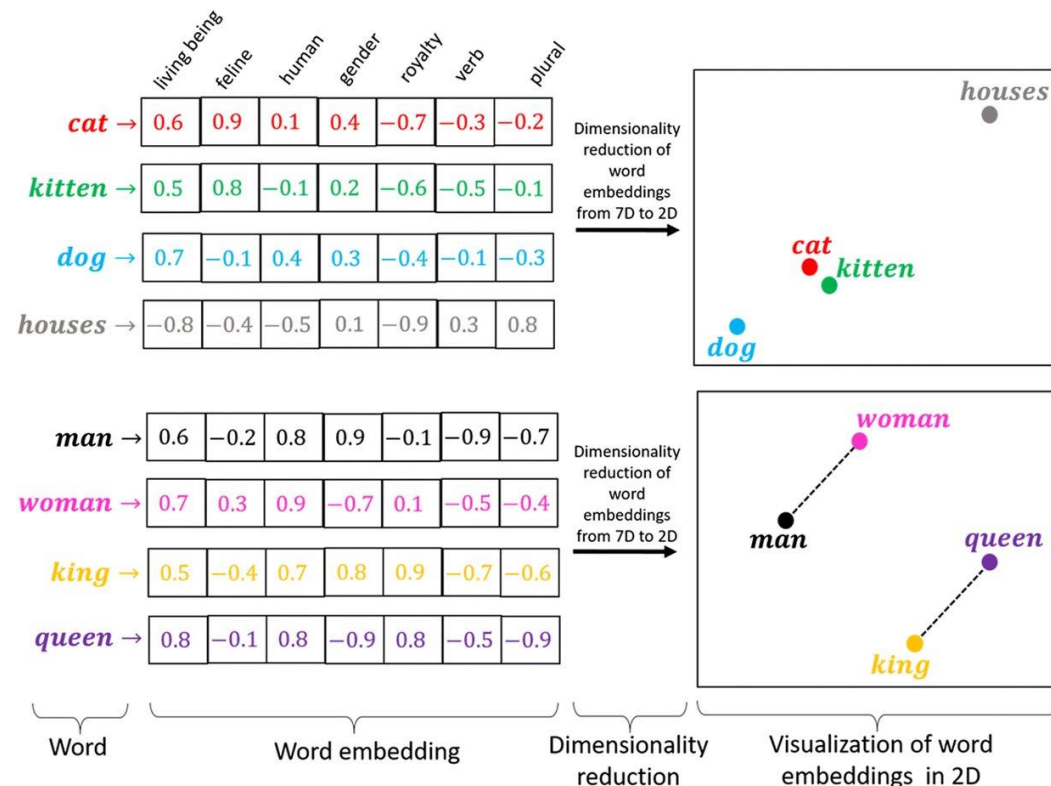
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# Sequence-Level Featurization

## Structural Features



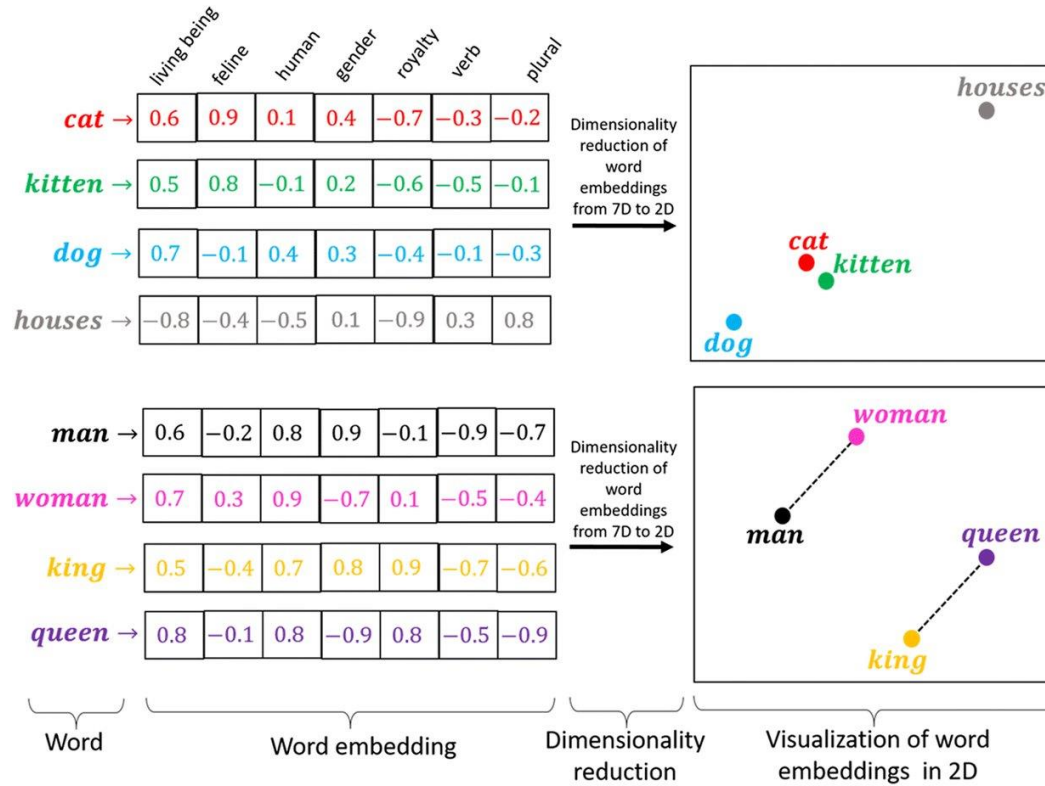
## Entity Embeddings



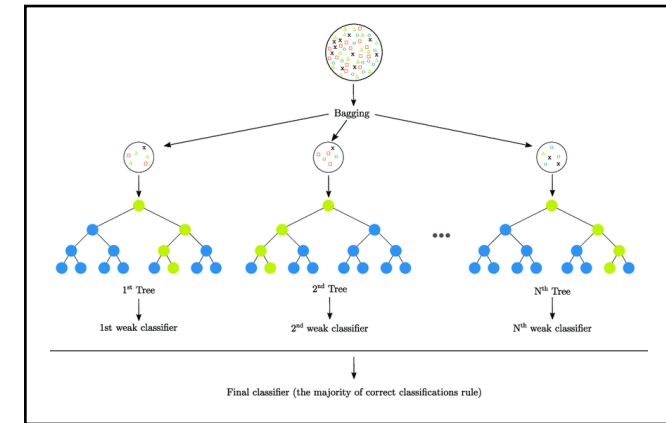


# Sequence-Level Featurization

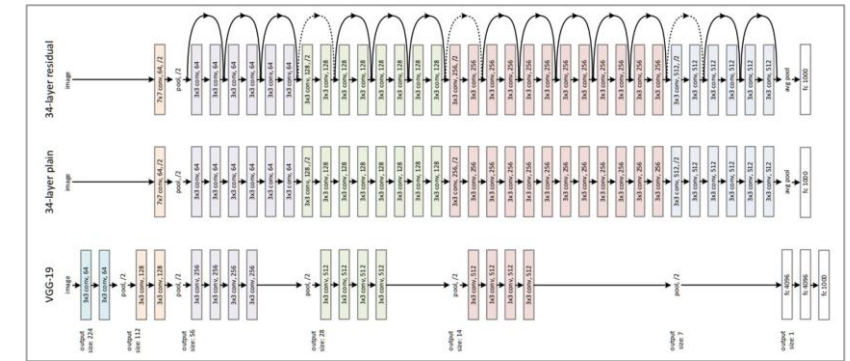
## Entity Embeddings



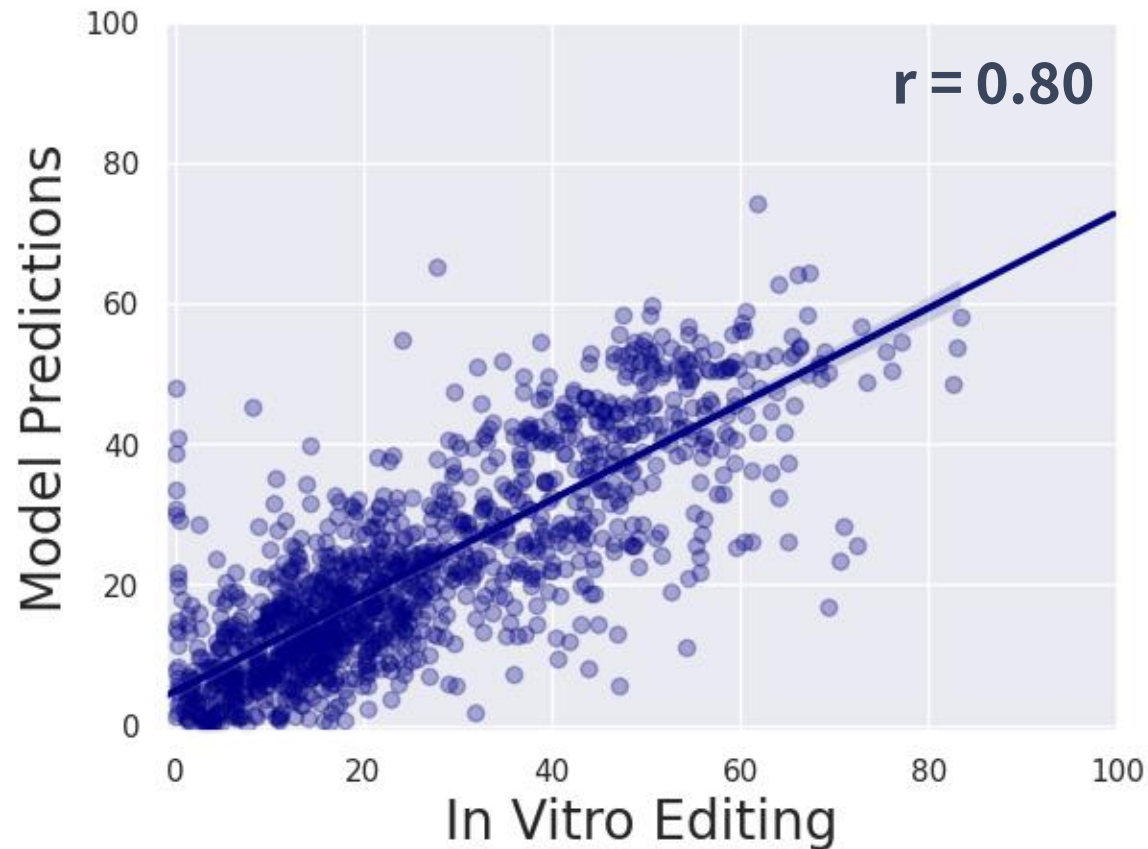
## Tree-based Models



## Convolutional Neural Network



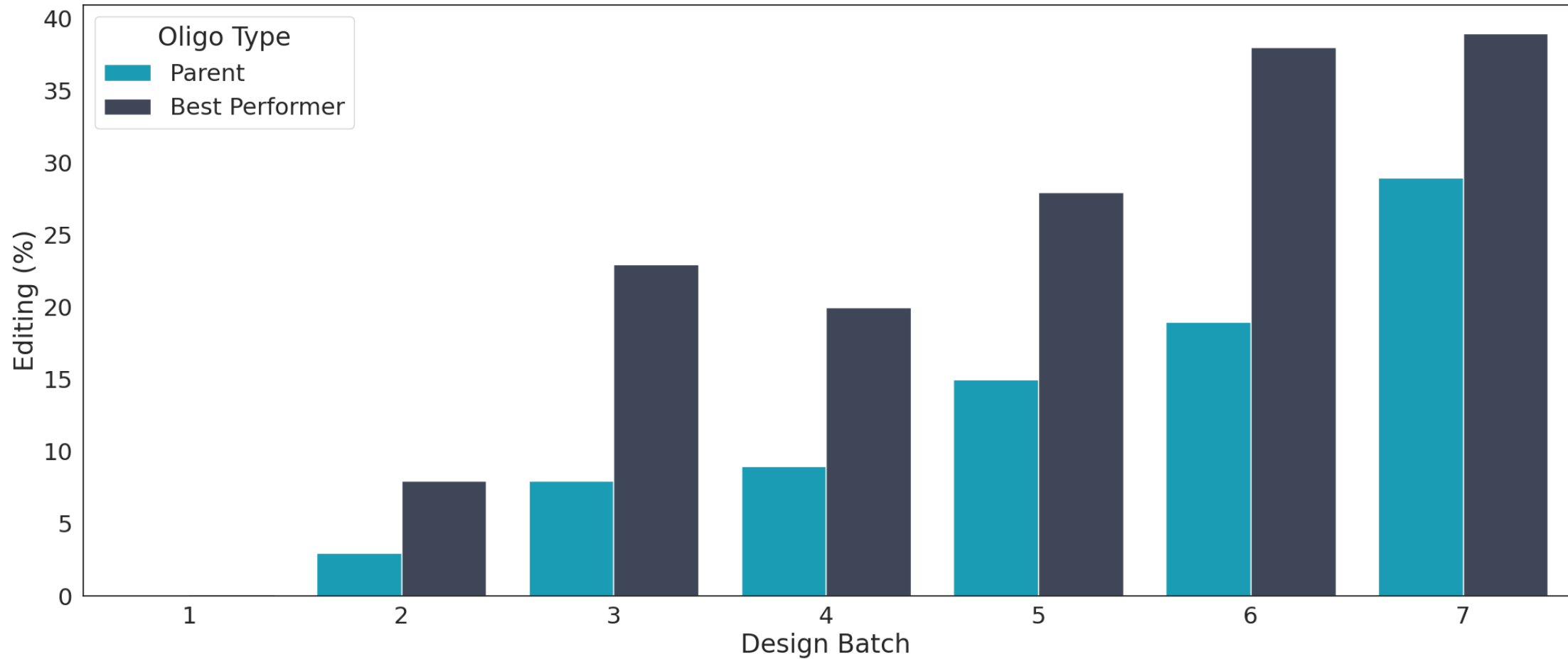
# Our Model Learns the Effect of Chemical Modification Patterns on *In Vitro* Editing



- Model tested on 20% of in vitro data for a single target
- Understanding of relationship between sequence, chemical modifications and in vitro editing
- Predictions within **7%** of in vitro editing



# Machine Learning Boosted mRNA Editing Through Iterative Design Batches



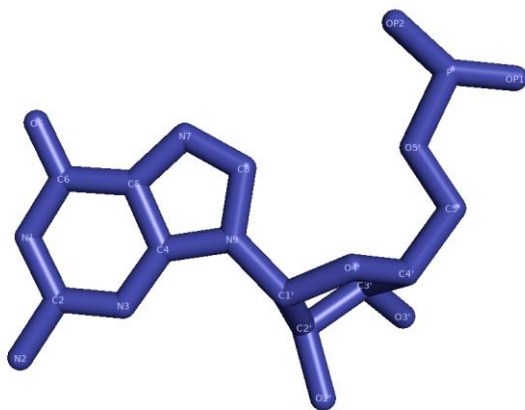
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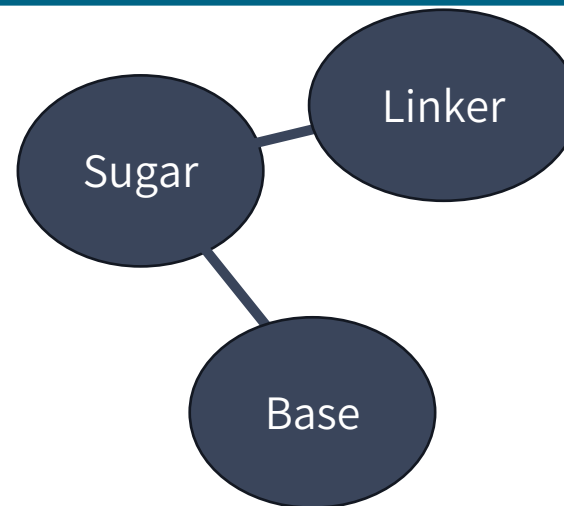
# Biological Feature Extraction Can Help Us Increase Use-cases for Machine Learning

AUGCACAGG

*Sequence*

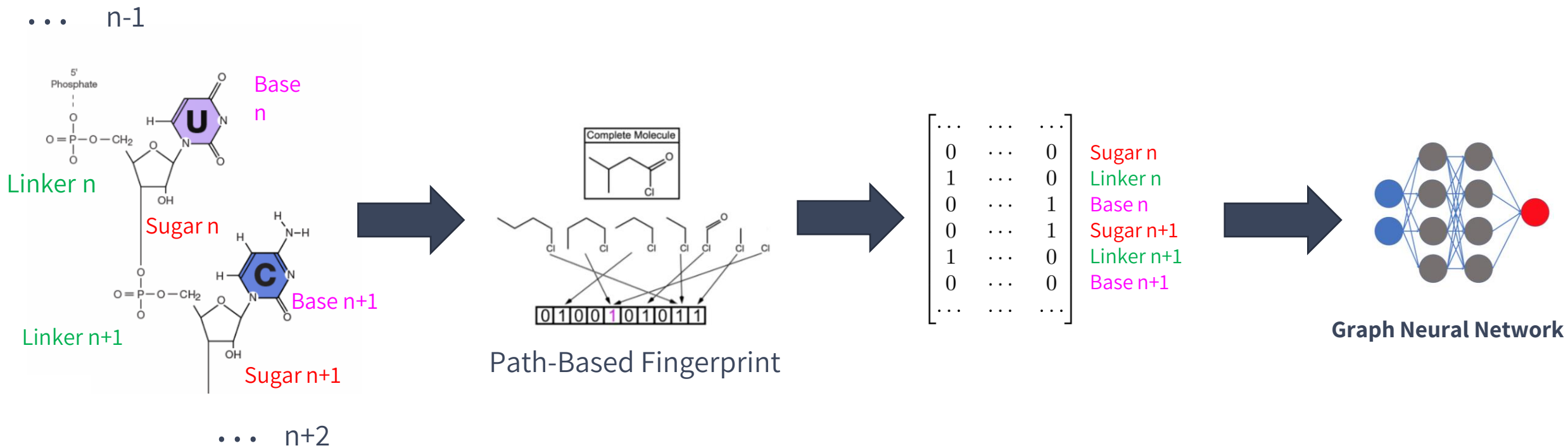


*Atoms*



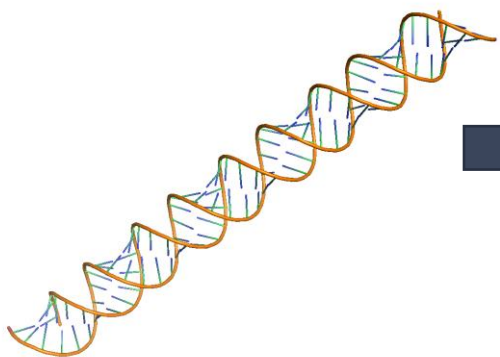
*Monomers*

# Monomer-Level Featurization

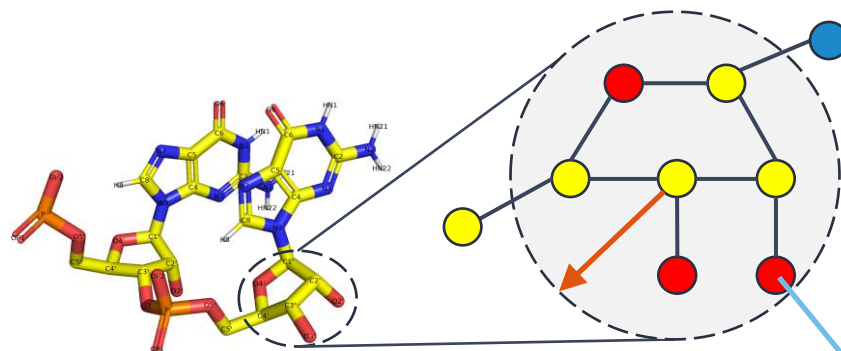


# Atom-Level Featurization

Starting Structure



Input Representation

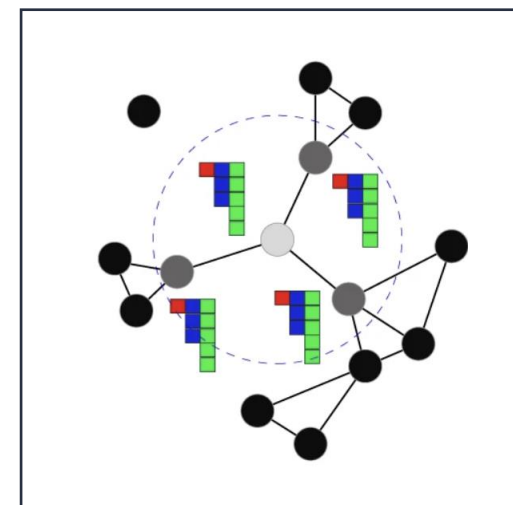


Node attributes  $X$

- Atomic Symbol?
- Atom Degree?
- Implicit Valence?
- Hybridization?
- Number of Hydrogens?
- Stereochemistry?

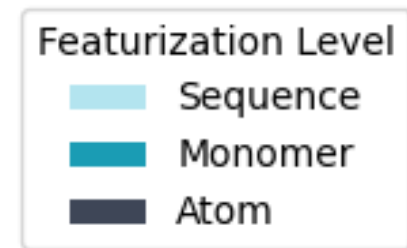
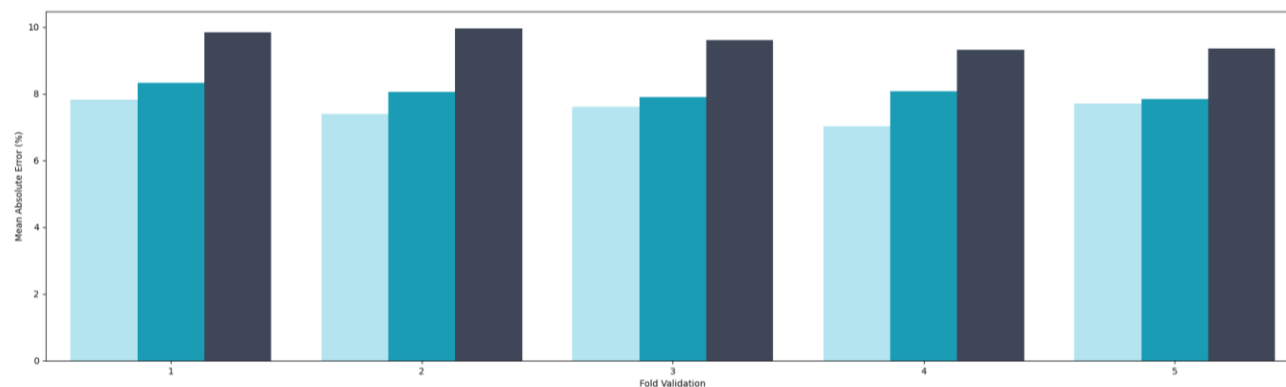
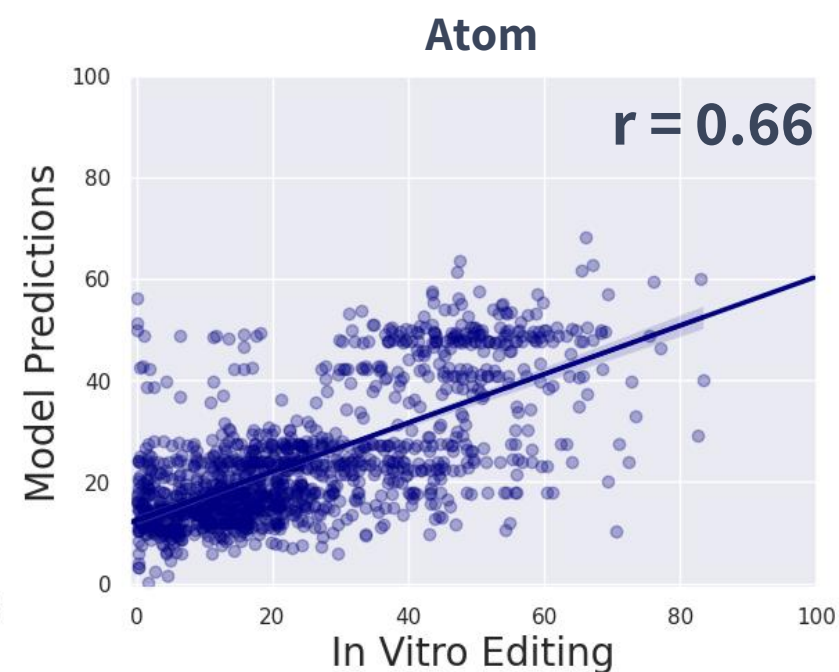
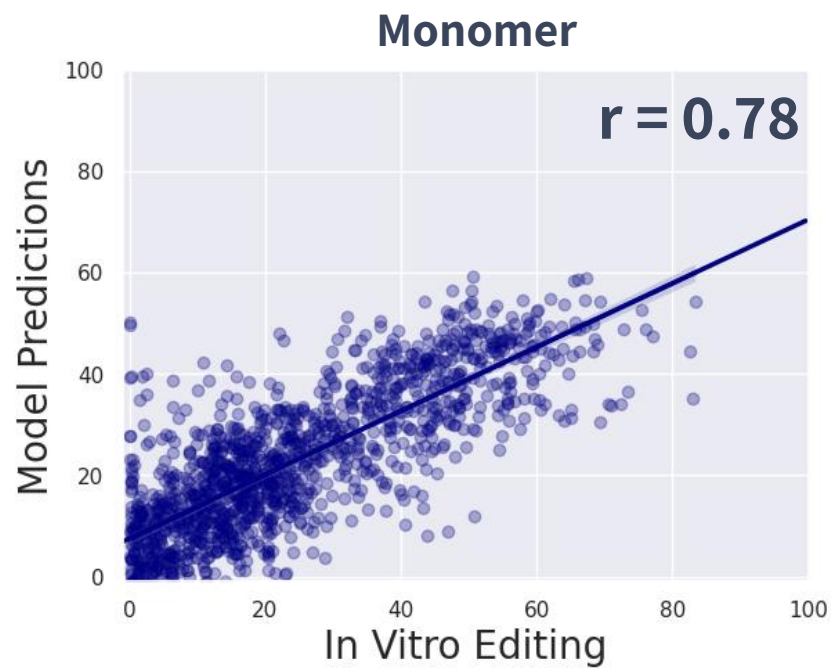
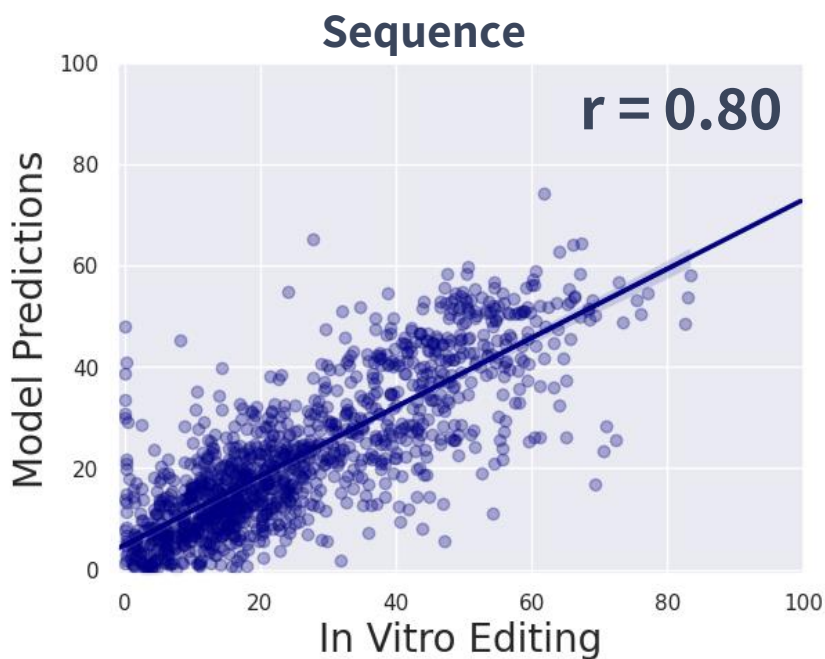


Graph Neural Network





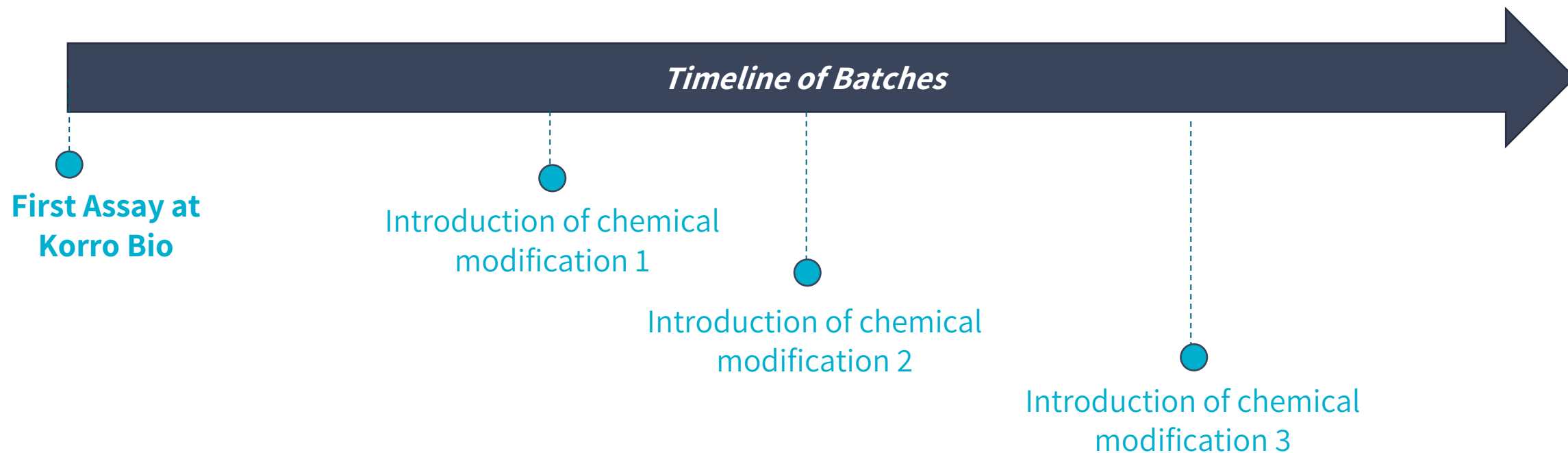
# Increasing Chemical Featurization Trends in a ~1% Increase in Overall Error of Model



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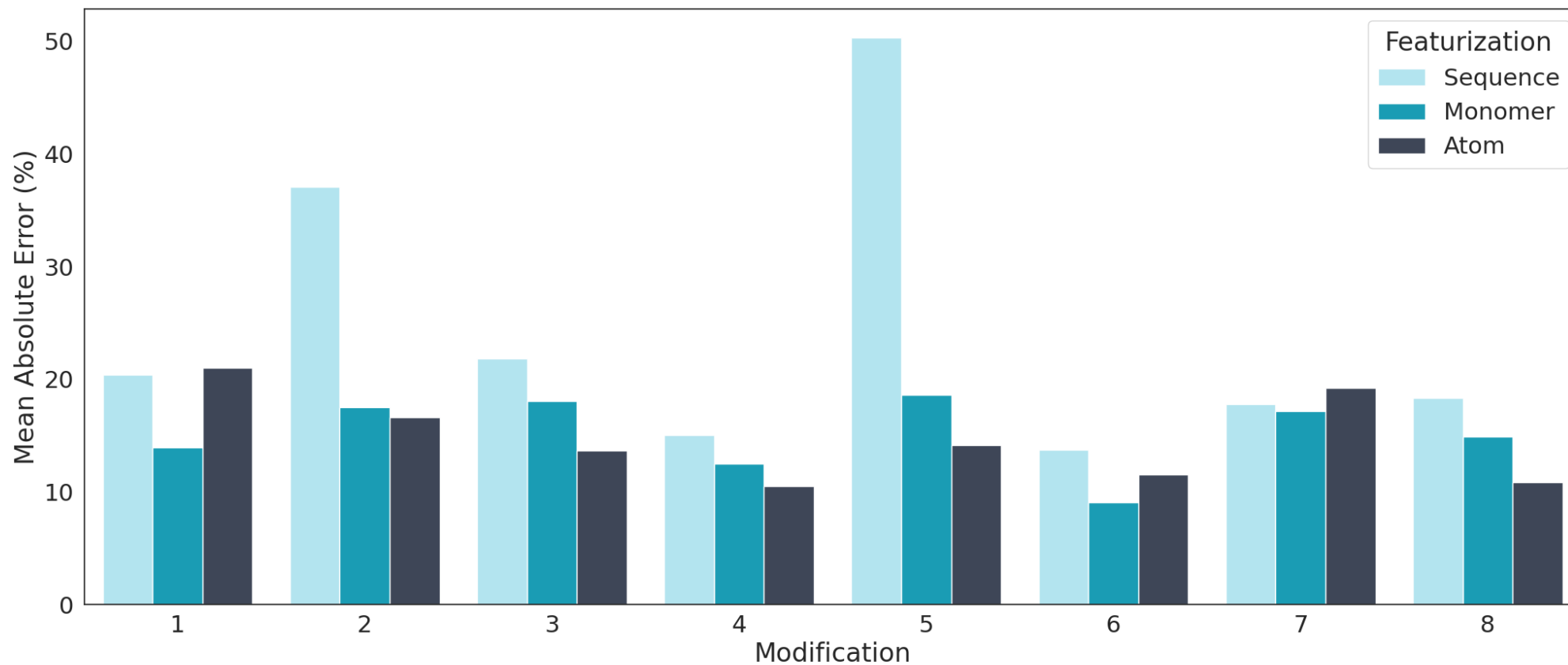
# Testing The Model's Ability to Incorporate New Modifications at Scale



## Assessing Chemical Generalization:

- Models are trained on all data **BEFORE** the introduction of a new modification
- Models are tested on the data from the batches that introduced the modification

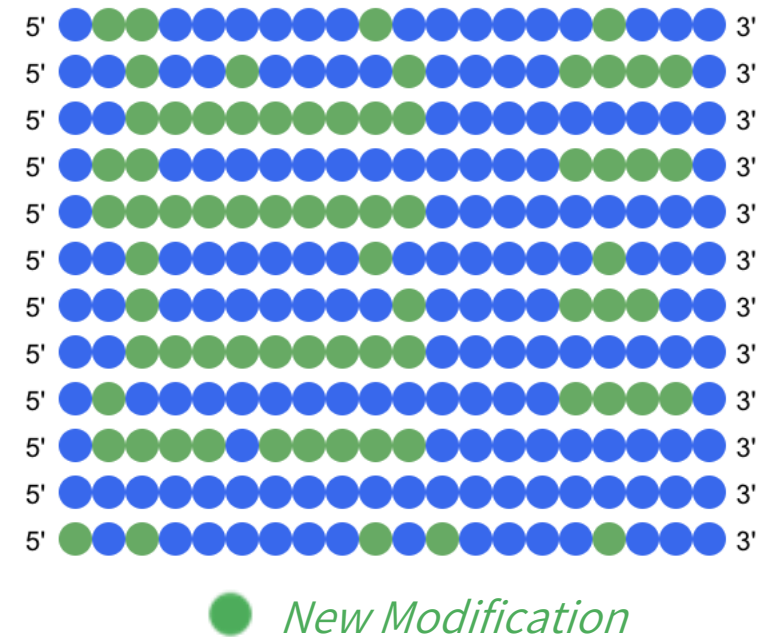
# Chemical Features Improve Overall Error Across 8 Unseen Modifications in Oligonucleotides



# Chemical Modifications are Often Tested in “Titration” Experiments



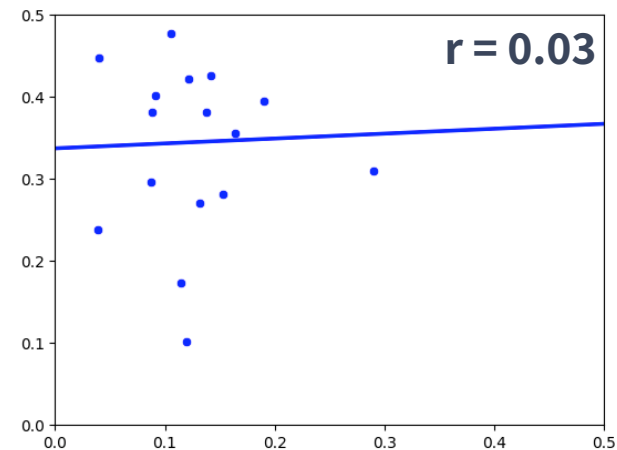
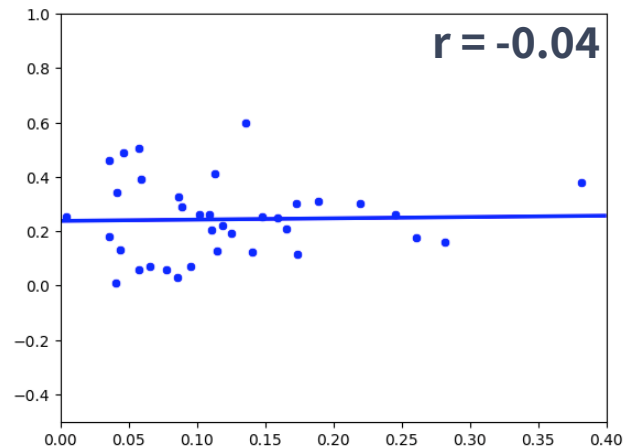
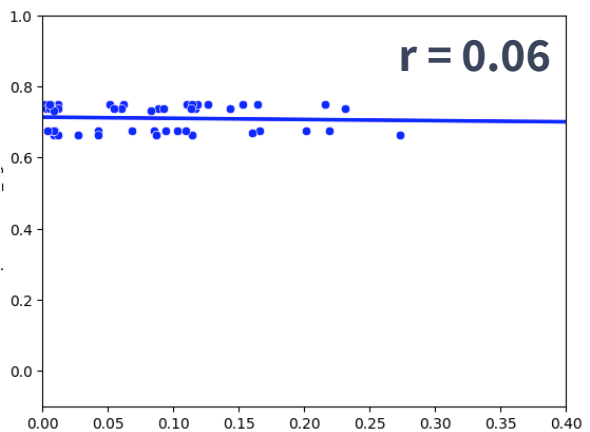
## New Chemical Modification Titration



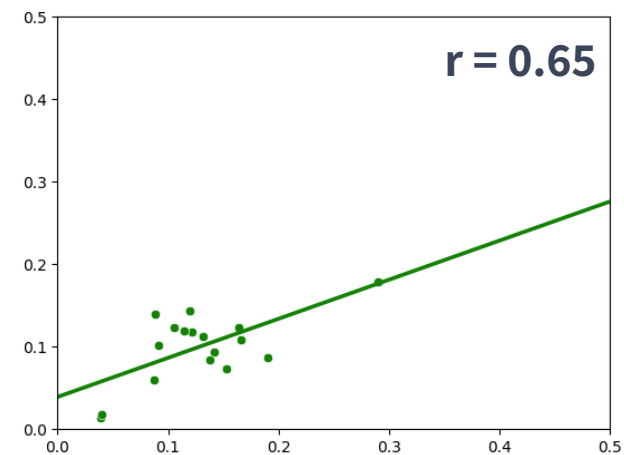
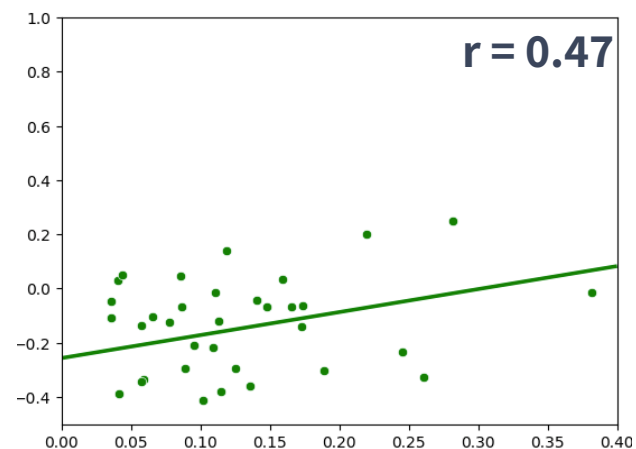
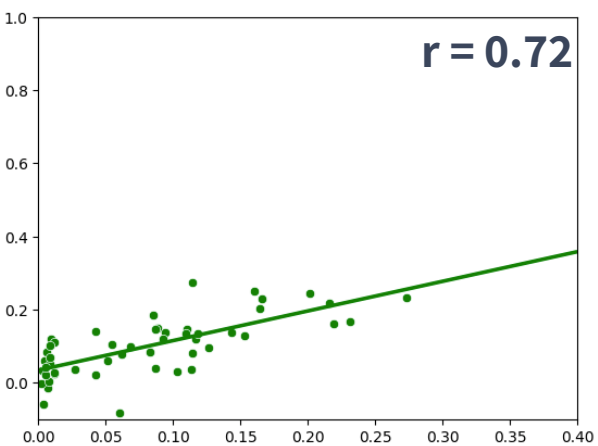
# Chemically Generalized GNN Can Help Us with New Modification Titrations

Model Prediction

Sequence



GNN



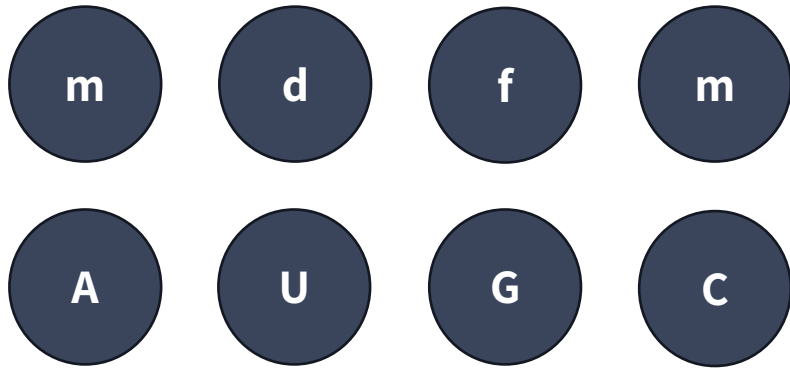
Editing

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# To Use Our Data on New Targets, We Needed to Take the Opposite Approach

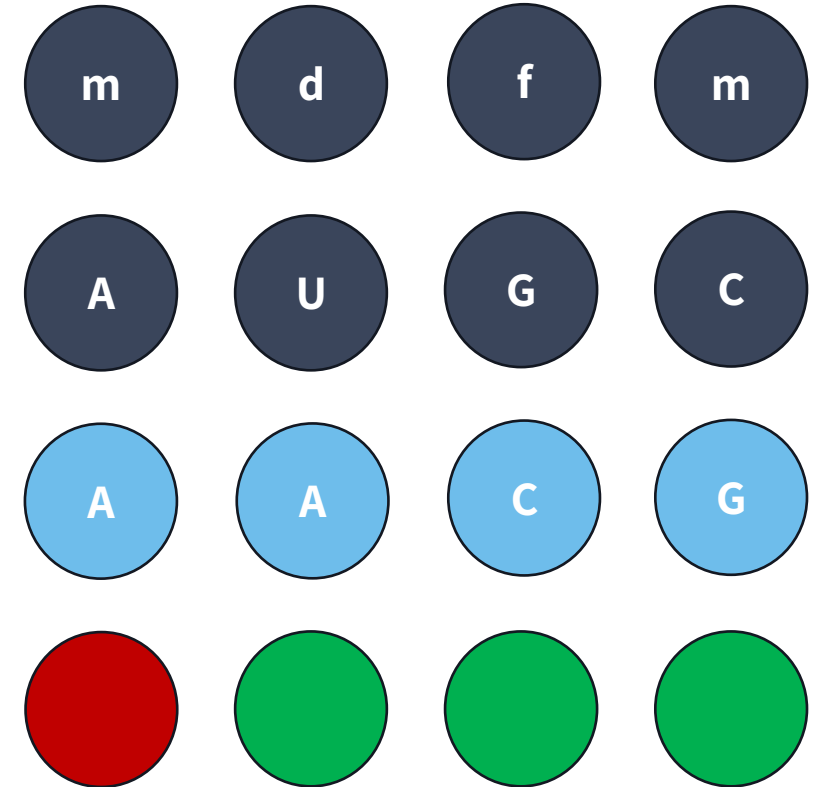
Sequence-Based Featurization



Insert Relationship to Target



Target-Agnostic Featurization

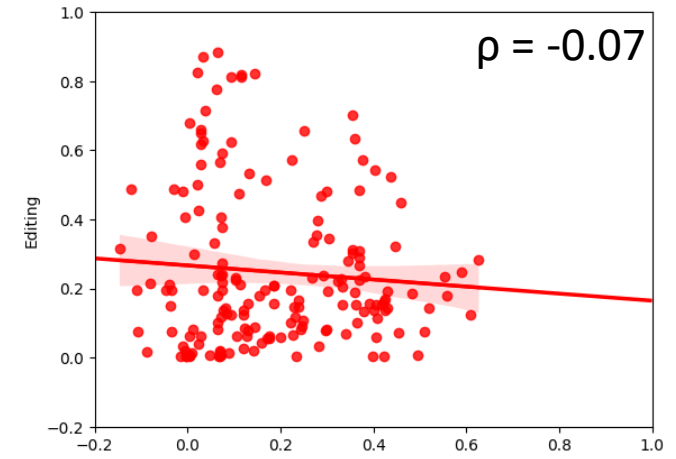
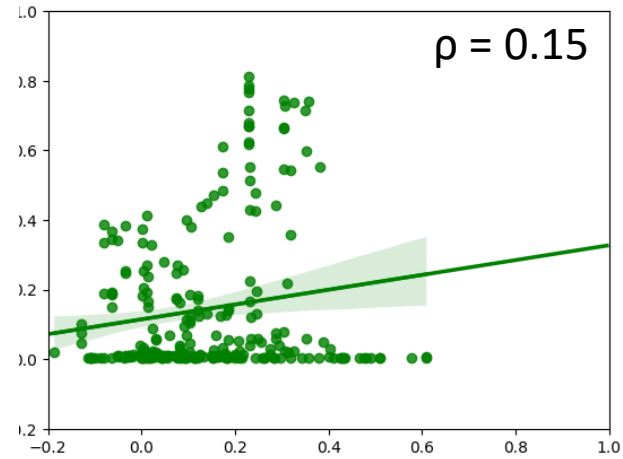
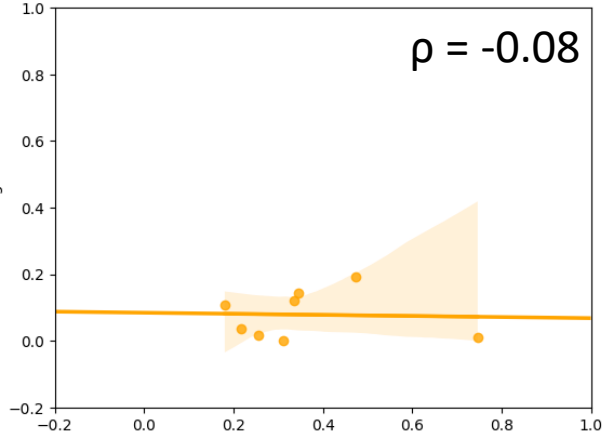




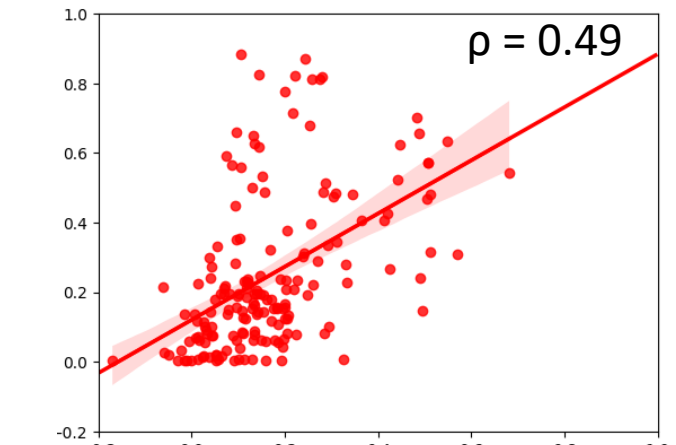
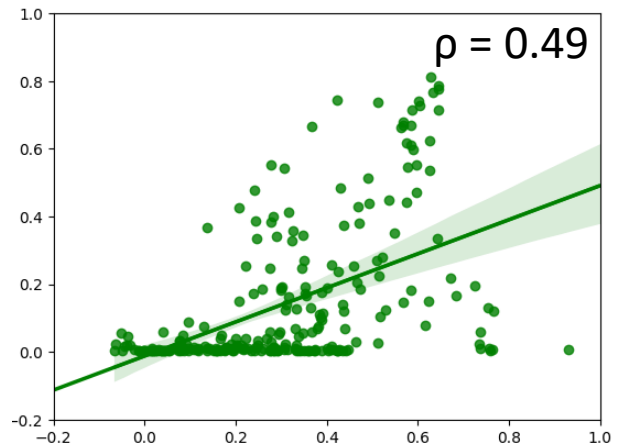
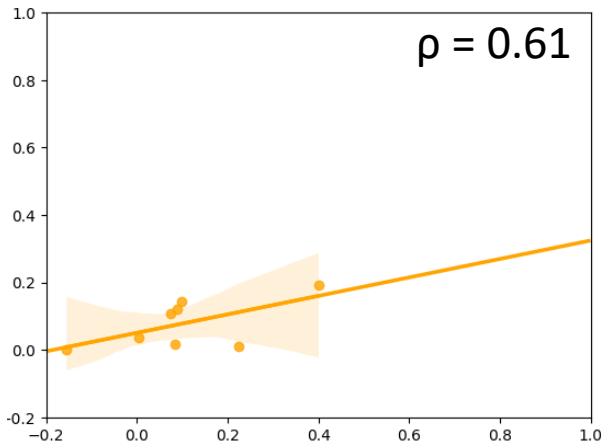
# Oligo-Target Interaction Features Lead to Better Models for New Targets and Cell Lines

Model Predictions

Sequence



Target Agnostic



Editing