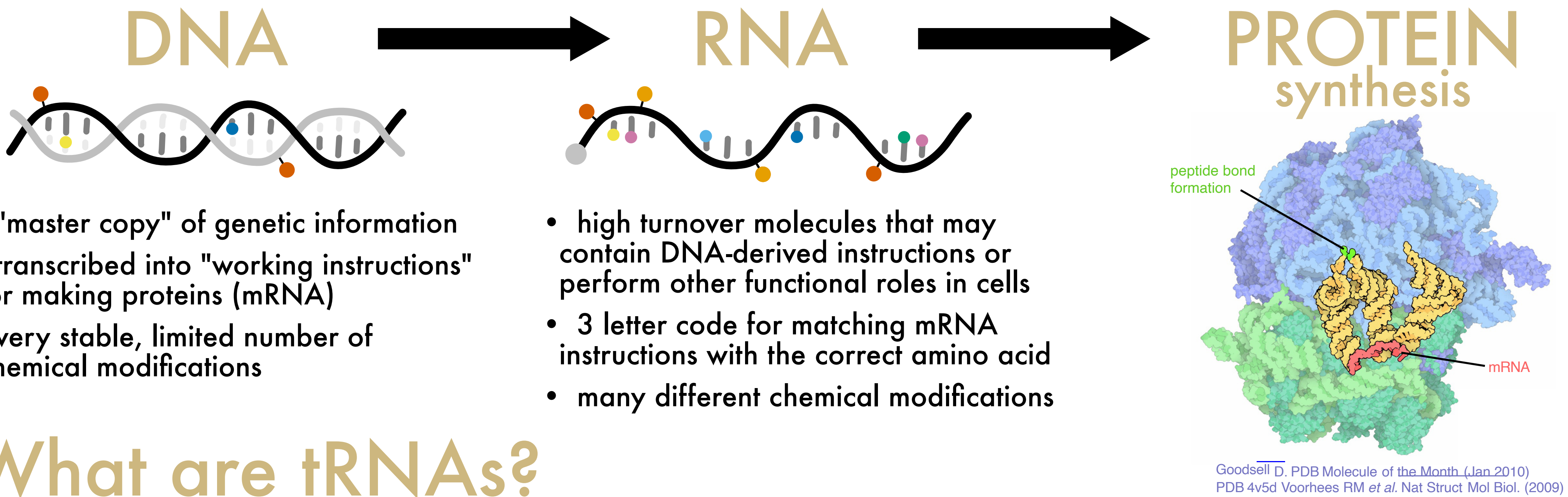
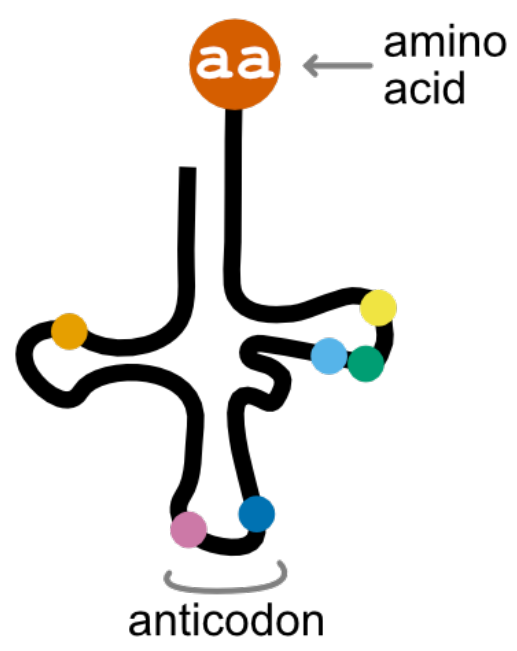


Sequencing RNA in New Ways:
*Engineering the Future of Nanopore Direct RNA
Sequencing at the RNA Bioscience Initiative*
Laura K. White, PhD



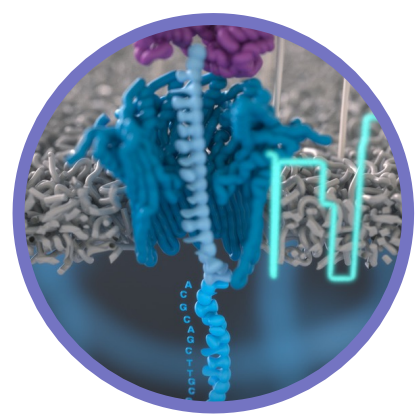
What are tRNAs?



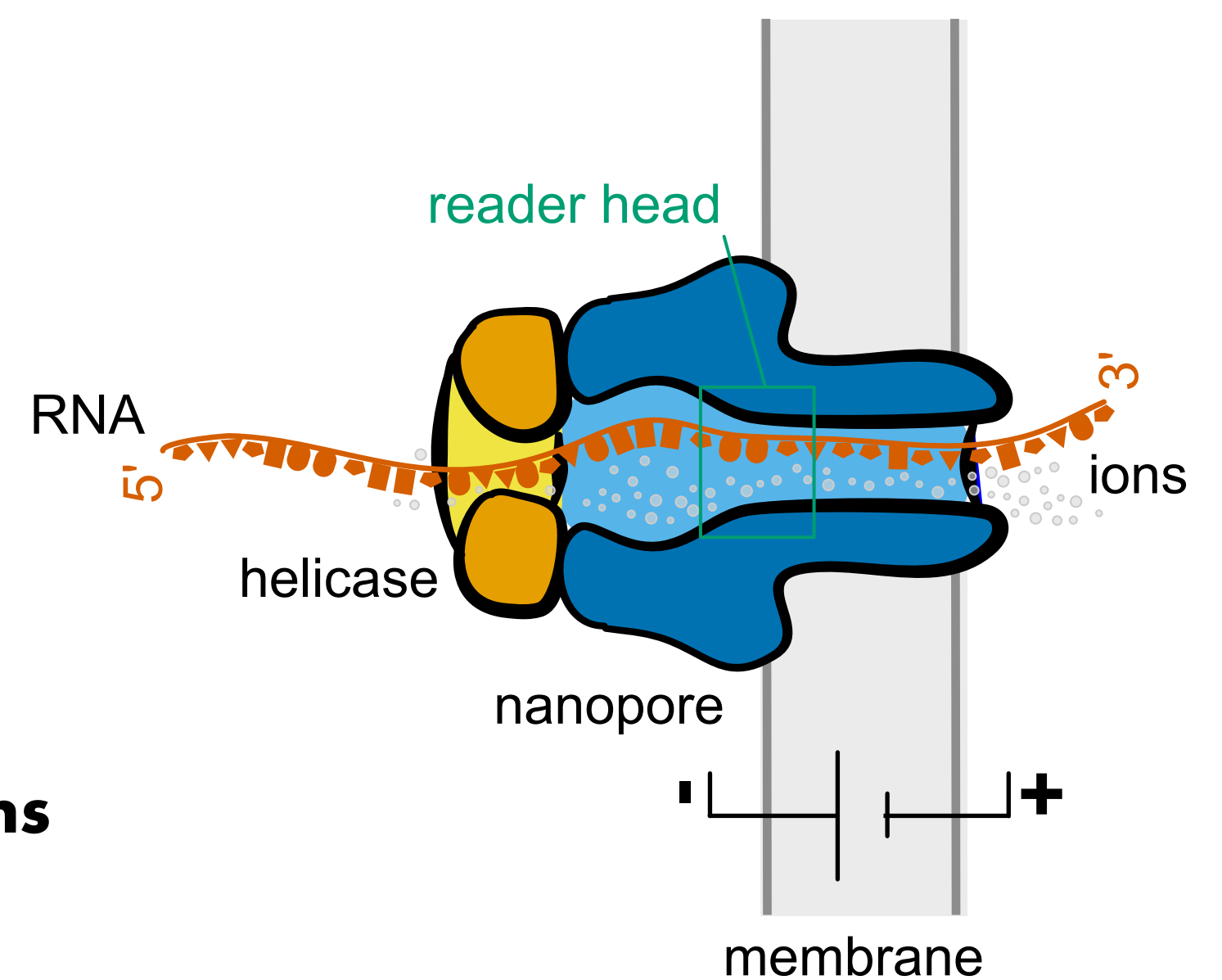
- Essential **adapter molecules** for making proteins
- At the bottom, the tRNA anticodon enables **specific pairing** with a corresponding codon sequence in mRNA
- At the top, tRNAs are charged with **amino acids** (the building blocks of proteins) that match their anticodon
- **Chemical modifications** impart stability & function

In the image above, the yellow shapes are individual tRNAs whose anticodons are paired up with matching codons in mRNA to make sure the right amino acids are added to the growing protein

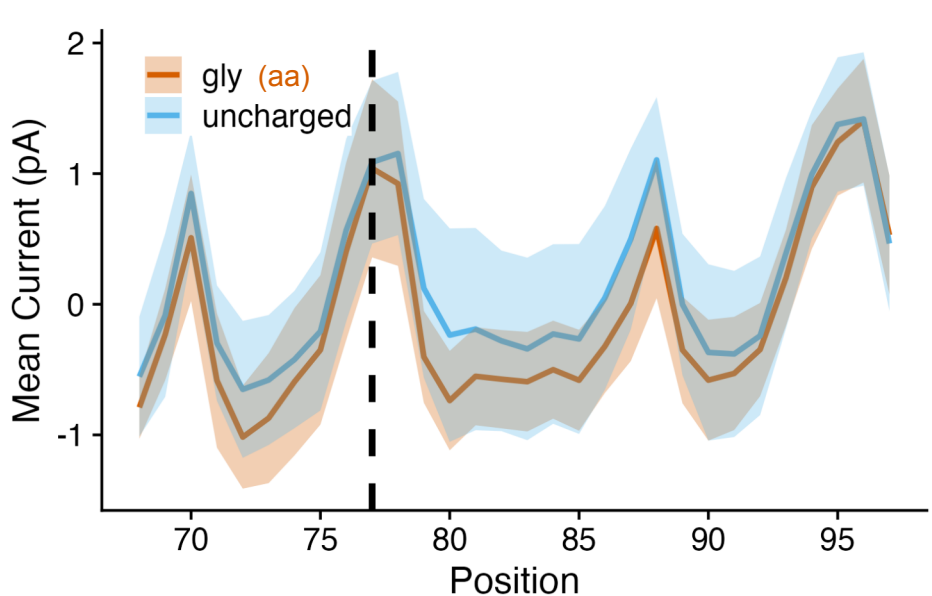
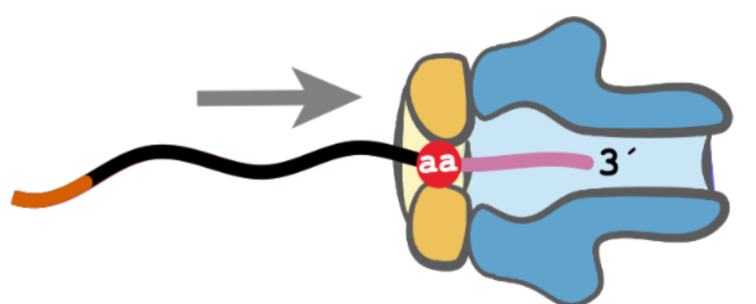
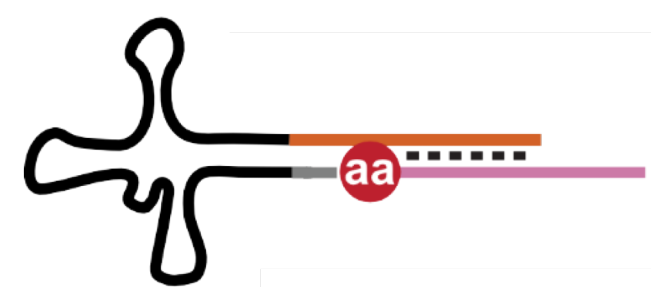
What is nanopore sequencing?



- Compact, commercially available emerging technology for sequencing **RNA or DNA** with chemical modifications intact
- The only way to **directly** sequence RNA molecules
- RNA is pulled through **biological nanopores** embedded in ionically charged membranes
- Different RNA nucleotides (A,C,G,U) produce distinct changes in the flow of **ionic current** through the nanopores
- Additional distortions in ion flow due to **chemical modifications** can be identified using **machine learning** algorithms



We developed a new approach to nanopore sequence tRNAs with their amino acids intact.



1. Proprietary **chemical ligation** to amino acids
2. Attach additional **sequencing adapters**
3. **Sequence** on nanopore RNA flow cells
4. Classify tRNAs as aminoacylated or "uncharged" using custom trained **machine learning** models

We want to use this to:

- ✓ study how tRNA aminoacylation is disrupted in **childhood** and **neurological diseases**
- ✓ understand relationships between **tRNA modifications** and amino acids
- ✓ develop machine learning models to identify amino acids from nanopore signal (new ways to **sequence proteins** using nanopores?)

Want to know more?

Read all about it here! →

White, Radakovic et al. Nanopore sequencing of intact aminoacylated tRNAs. BioRxiv preprint. Nov. 2024



RNA Bioscience Initiative

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