



# Dynamic multilayer DNA data storage

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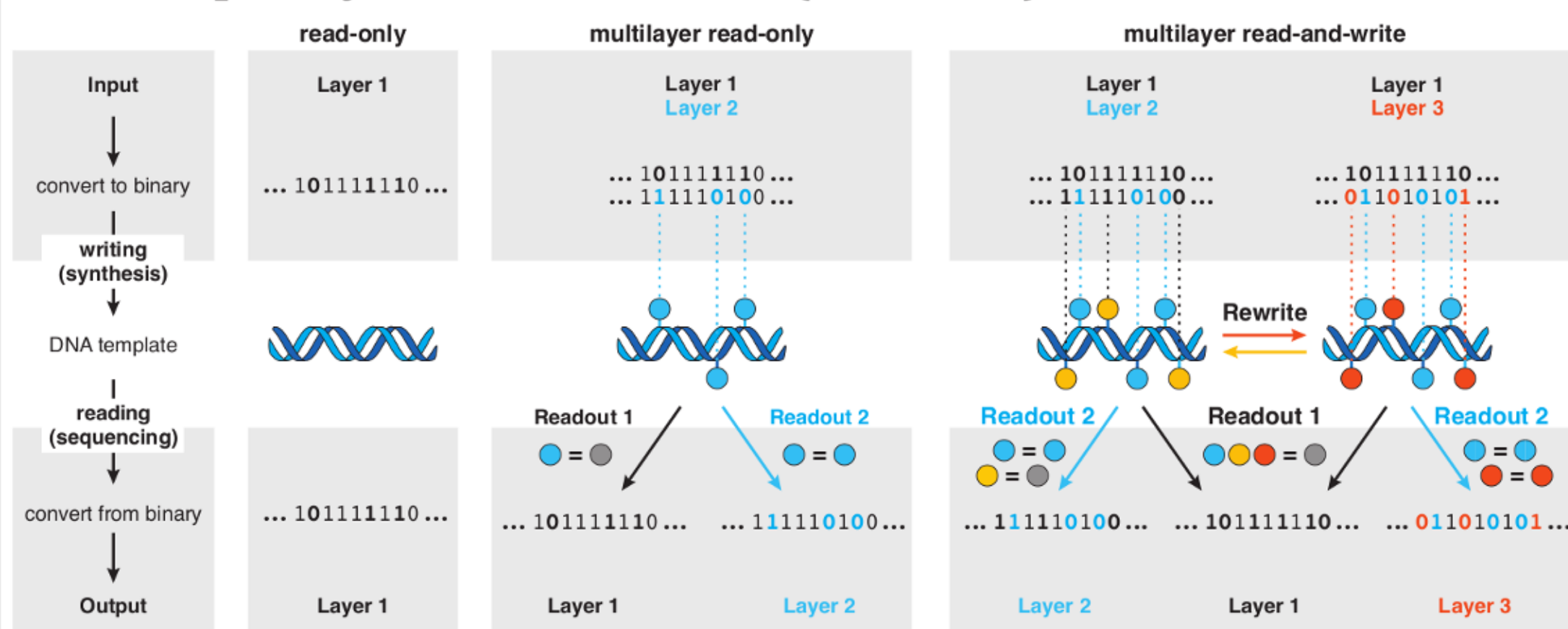
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## An introduction to DNA data storage

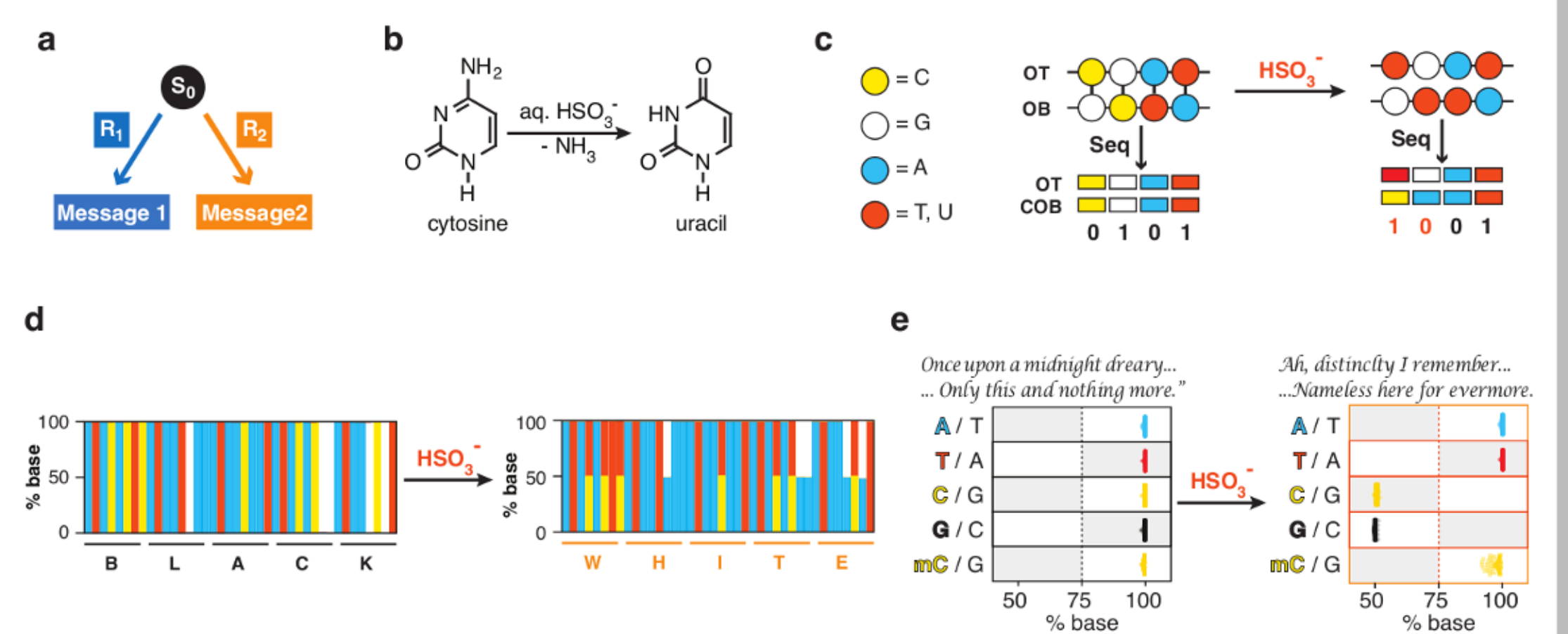
- Biopolymers are an attractive alternative to traditional magnetic and optical storage media. [1]
- DNA combines high data storage density with a proven track record as lasting information bearer in biology.
- An arbitrary binary input can be allotted to a library of synthetic oligonucleotides.
- Chemical transformation can, in principle, enable **encoding of multiple layers** of information (Scheme 1).



Scheme 1: Blueprint for encoding multiple layers of information in DNA.

## Multilayer encoding Part I - two messages

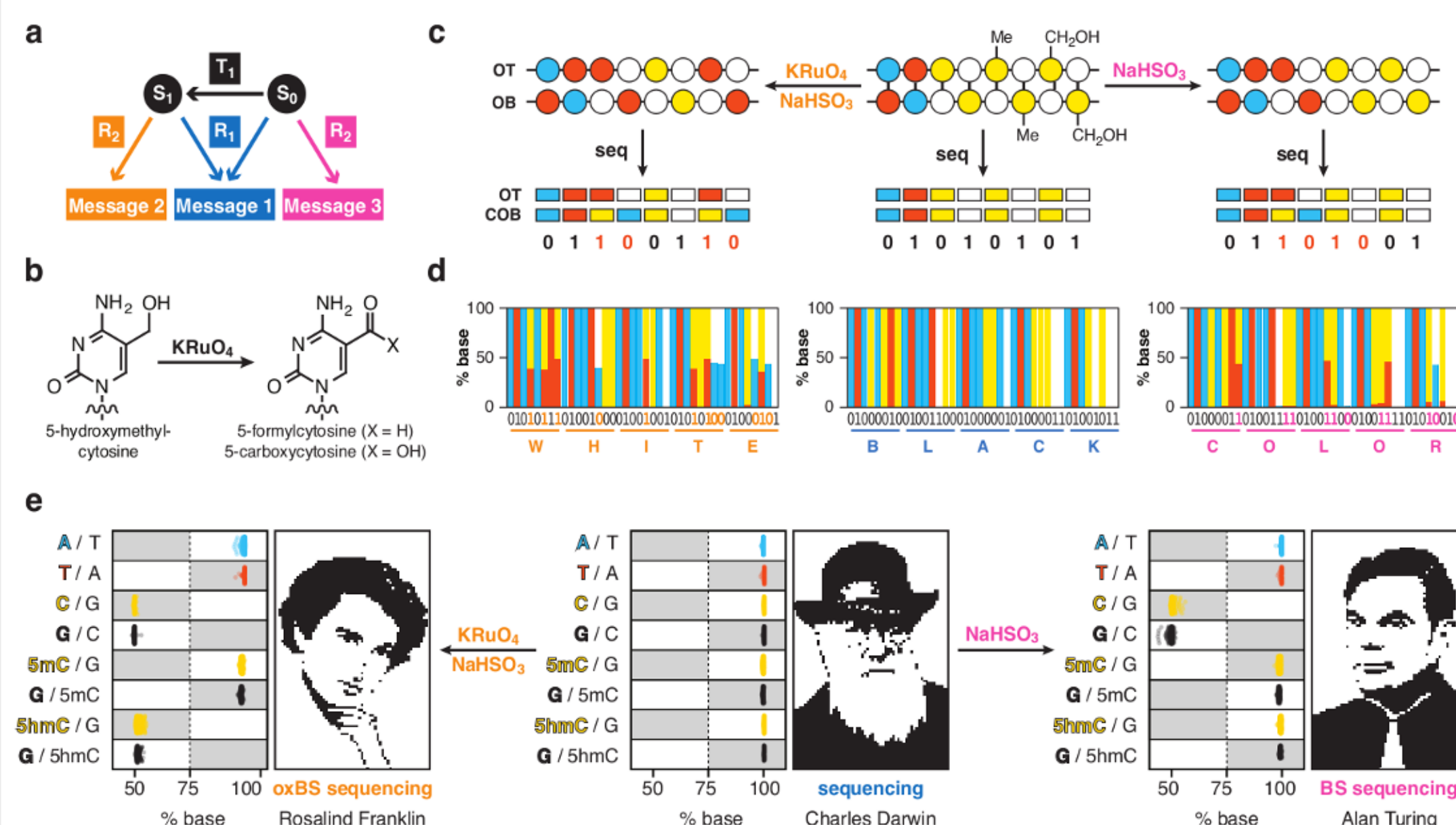
- We exploit bisulfite induced cytosine to uracil conversion for the simultaneous **encoding of two messages** in DNA (Scheme 2a & 2b).
- Transformation of cytosine to uracil enables the interconversion of binary digits (Scheme 2c).
- As a result two messages can be recovered from a single template (Scheme 2d). The approach is **general** and **scalable** (Scheme 2e).



Scheme 2: Simultaneous encoding of two layers of binary information in DNA.

## Multilayer encoding Part II - three messages

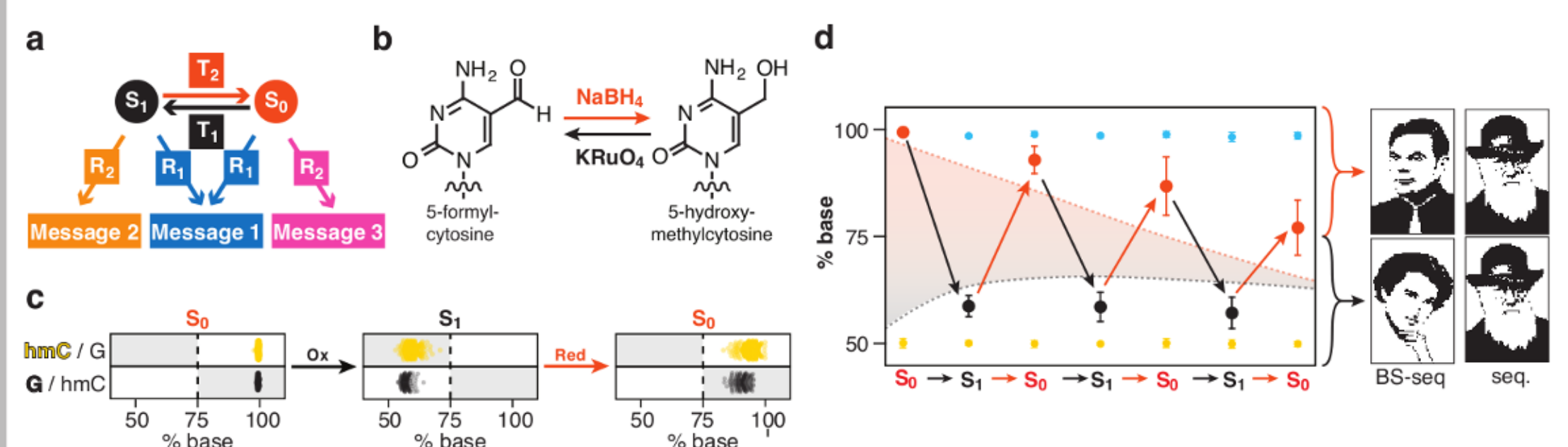
- Addition of 5-methylcytosine and 5-hydroxymethylcytosine [2] to the canonical nucleobases allows for the simultaneous encoding of **three layers of information** (Scheme 3a - 3d).
- Portraits of Charles Darwin, Rosalind Franklin, and Alan Turing were recovered from a single oligonucleotide library (Scheme 3e).



Scheme 3: Simultaneous encoding of three layers of binary information in DNA.

## Multilayer encoding Part III - reversibility

- Reversible encoding of three layers of information is possible when the selective oxidation 5-hydroxymethylcytosine to 5-formylcytosine is incorporated into a redox cycle (Scheme 4a - 4c).
- The encoded information could be rewritten **3 times** (Scheme 4d).



Scheme 4: Reversible encoding of three layers of information in DNA.

## Conclusion

- Selective chemical transformations turn DNA into a dynamic data storage medium.
- When several reactions are combined, up to three layers of information can be reversibly encoded in DNA.