# STORING THE DECLARATION OF HUMAN RIGHTS ON ONE DNA MOLECULE


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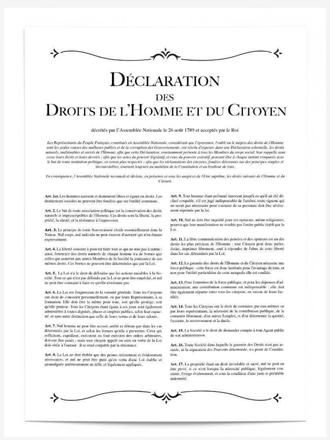
## ABSTRACT

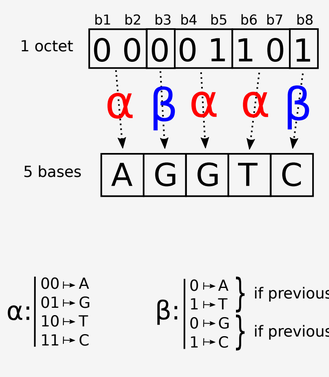
Today, the **community consensus** to store information on DNA is to use **short single strand DNA (ssDNA) molecules**. This approach has some limitations: encoding constraint, DNA stability, recovering DNA, sequencing technology, *etc*. To overcome them, we chose to store information on **long double-strand DNA (dsDNA) molecules**.

**Our demonstration consists of storing the first articles of the declaration of human rights (4,2ko text document) on a single DNA molecule.**


| Declaration of Human Right storage features | Standard approach | dnarXiv approach |
|---|---|---|
| DNA structure | Single-strand (ssDNA) | Double-strand (dsDNA) |
| DNA strand length | 100-200 nucleotides (nt) | 25,000 base pairs (bp) |
| DNA molecules number | 210 - 420 molecules | 1 molecule |
| Information density | 0,81 bits/nt | 1,36 bits/nt |
| Error correction method | Yes | No |
| Sequencing technology | Illumina: short read | Nanopore: long read |

## ENCODING



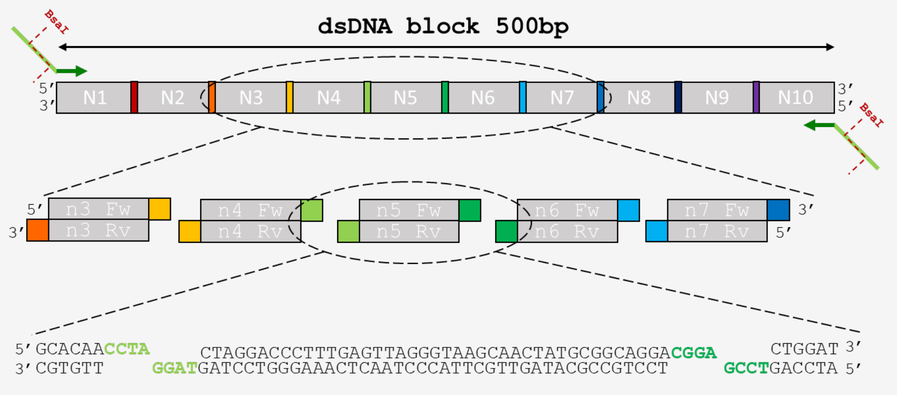



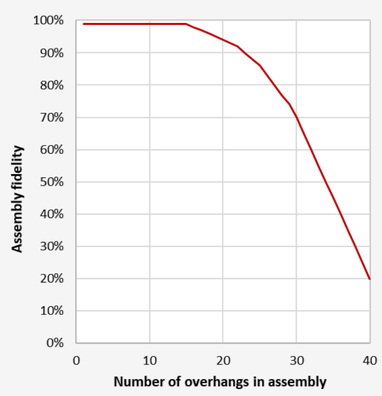

- Input : **binary sequence**
- Code respecting **biological constraints**
- Output : **dna sequence**

α: 00 ↦ A, 01 ↦ G, 10 ↦ T, 11 ↦ C

β: 0 ↦ A, 1 ↦ T if previous base ∈ {G,C}; 0 ↦ G, 1 ↦ C if previous base ∈ {A,T}

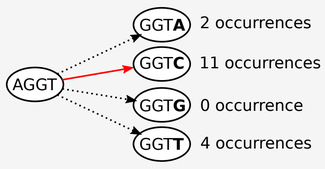
## SEQUENCE DESIGN



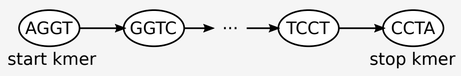



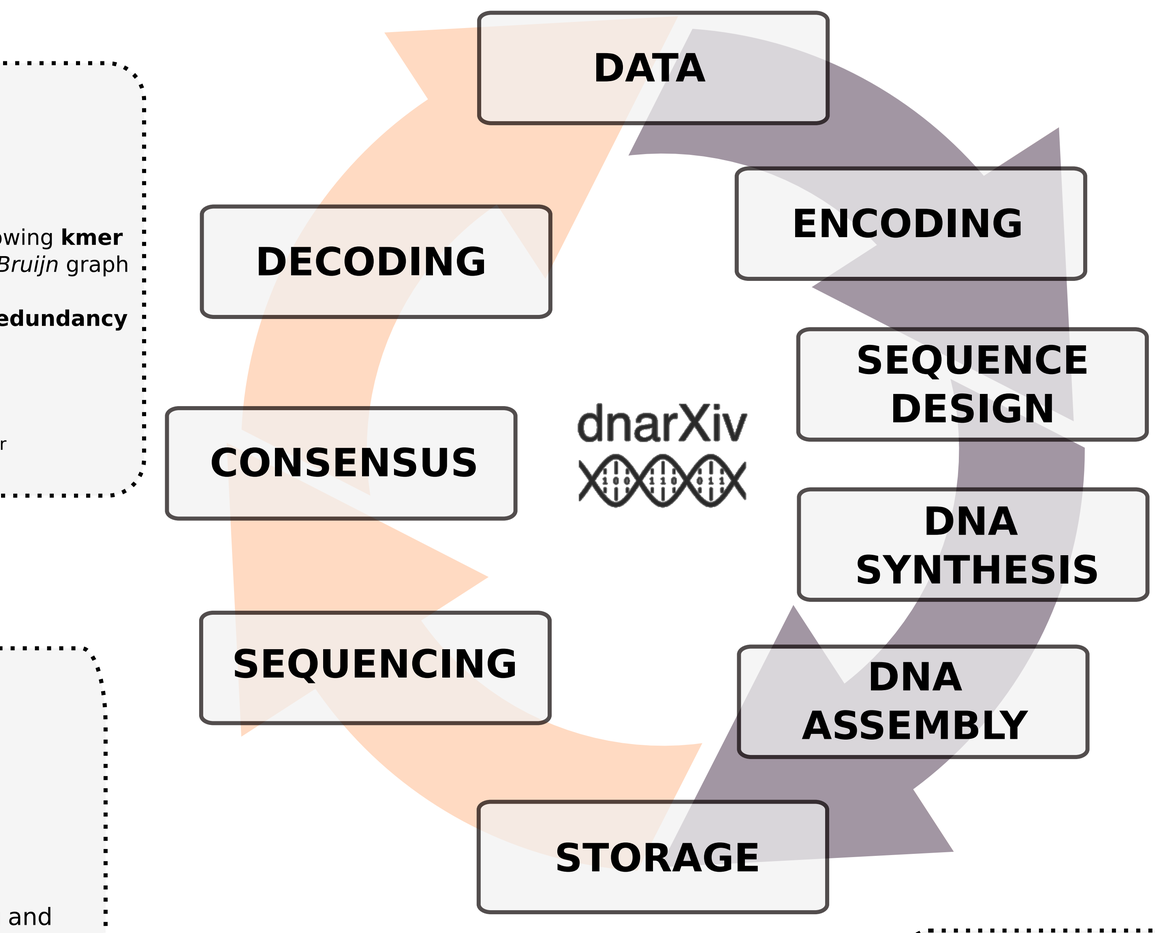

**Build 500bp dsDNA block with ssDNA oligonucleotides:**
- **84,8%** "**Payload**": encoded data.
- **7,2%** "**Overhangs**": 9 specific 3' overhangs of 4nt.
- **8,0%** "**PCR primers**": block amplification + add specific 3' overhangs through BsaI restriction sites.

## CONSENSUS

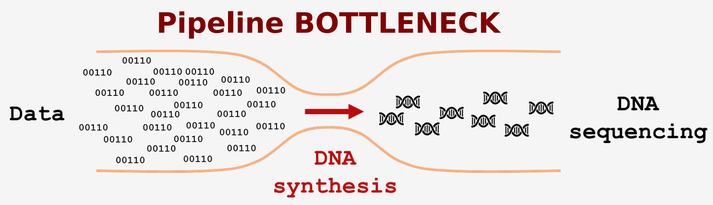



- **Kmer-counting**
- Path building by following **kmer occurrences** in a *de Bruijn* graph
- Error correction by **redundancy**



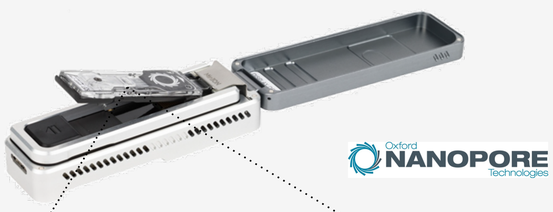

## DNA SYNTHESIS

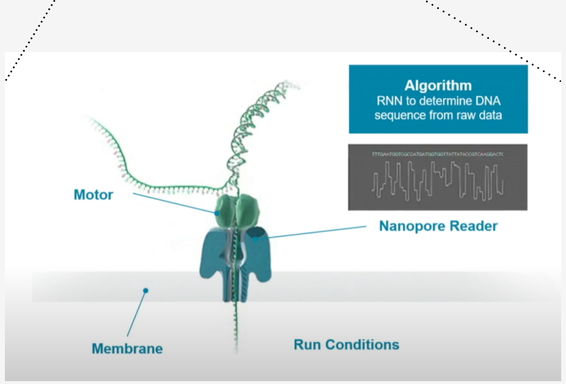



- **Chemical synthesis** by phosphoramidite method
- **High cost**: 0,123€/base
- Oligonucleotide length: **350nt**
- **Error rate: negligible**
- Synthesis speed: **100 to 200 seconds/nucleotide**
- In development: **enzymatic synthesis**

## DNA SEQUENCING

- Read length: **up to 1Mb**
- **Acccuracy: 99%**
- **Real time** sequencing
- **Portable**
- **Similar cost to Illumina sequencing**
- **In progress**: real-time selective sequencing and detection of chemical modification




**Nanopore reader**: raw signal (fast5)
**Basecalling**: converting files in DNA sequence (fastq)

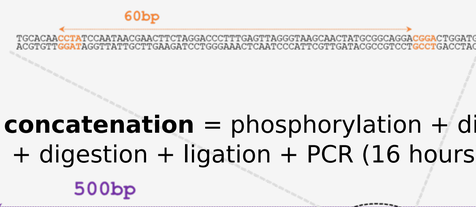
## STORAGE

***In vitro***: DNA molecules are simply stored at **room temperature** in a **buffer** (Tris-EDTA pH8).

| *In vitro* DNA storage | | |
|---|---|---|
| Features | Short ssDNA | Long dsDNA |
| Storage density* | + | ++ |
| Random access capacity° | + | +++ |
| Duplication capacity** | - | + |
| Storage stability | + | ++ |
| Security & stenography | + | ++ |

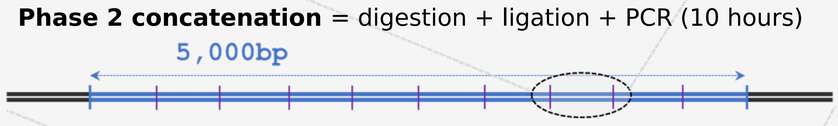
* Storage density: DNA molecules/mm$^3$ .
° Random access: access to a specific file without decoding the entire storage.
** Duplication: create complete copy of DNA storage.

## DNA ASSEMBLY

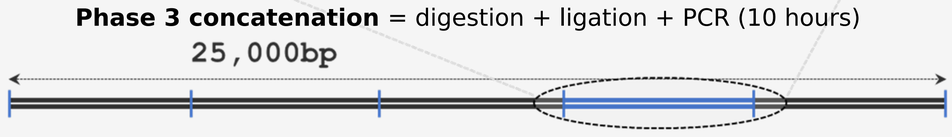



**Phase 1 concatenation** = phosphorylation + dimerization + digestion + ligation + PCR (16 hours)


**Phase 2 concatenation** = digestion + ligation + PCR (10 hours)




**Phase 3 concatenation** = digestion + ligation + PCR (10 hours)




Declaration of the Rights of Man and the Citizen of 1789