

On the trail of viruses: understanding virus research

Info sheet: primer design

What is the structure of DNA/RNA?

DNA is the carrier of genetic information. This means that it is the blueprint of an organism. All important information is encoded in the DNA.^[1]

DNA consists of individual building blocks called nucleotides (Figure 1Ⓐ). A nucleotide consists of a phosphate acid, a sugar and a base. There are four different bases: Thymine, cytosine, adenine and guanine (Figure 1Ⓑ), whereby in RNA the base thymine is replaced by uracil. The sugar is the eponym of DNA or RNA. In DNA, the sugar is a deoxyribose, in RNA a ribose.

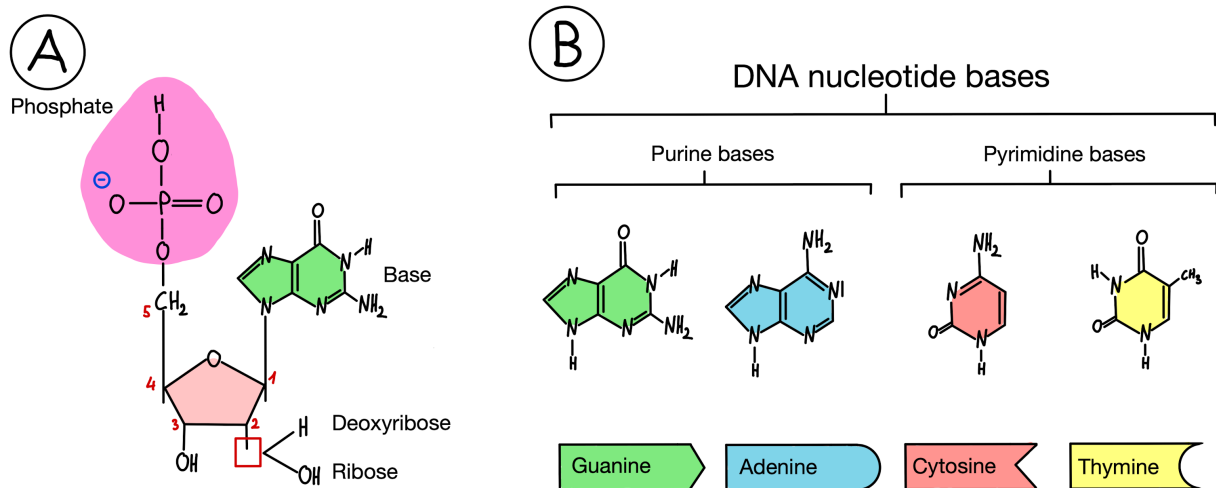


Figure 1: Structure of the nucleotides Ⓐ and DNA nucleotide bases Ⓑ
Image courtesy of the author

Each DNA molecule consists of two DNA strands that run antiparallel, i.e. in opposite directions to each other (Figure 2). The DNA strands are numbered at their ends. They have a 3' and a 5' end. The numbering refers to the carbon atoms in the deoxyribose or ribose (see Figure 1Ⓐ).

The two DNA strands are linked to each other via the bases. An adenine base (A) always pairs with thymine (T) and vice versa, and a guanine base (G) with cytosine (C) and vice versa. Hydrogen bonds are formed between the bases: two hydrogen bonds per AT pair and three hydrogen bonds per GC pair (Figure 2). A GC bond is therefore more stable than an AT bond, as more hydrogen bonds are formed. This is also important for the primers later on.^[1,2]

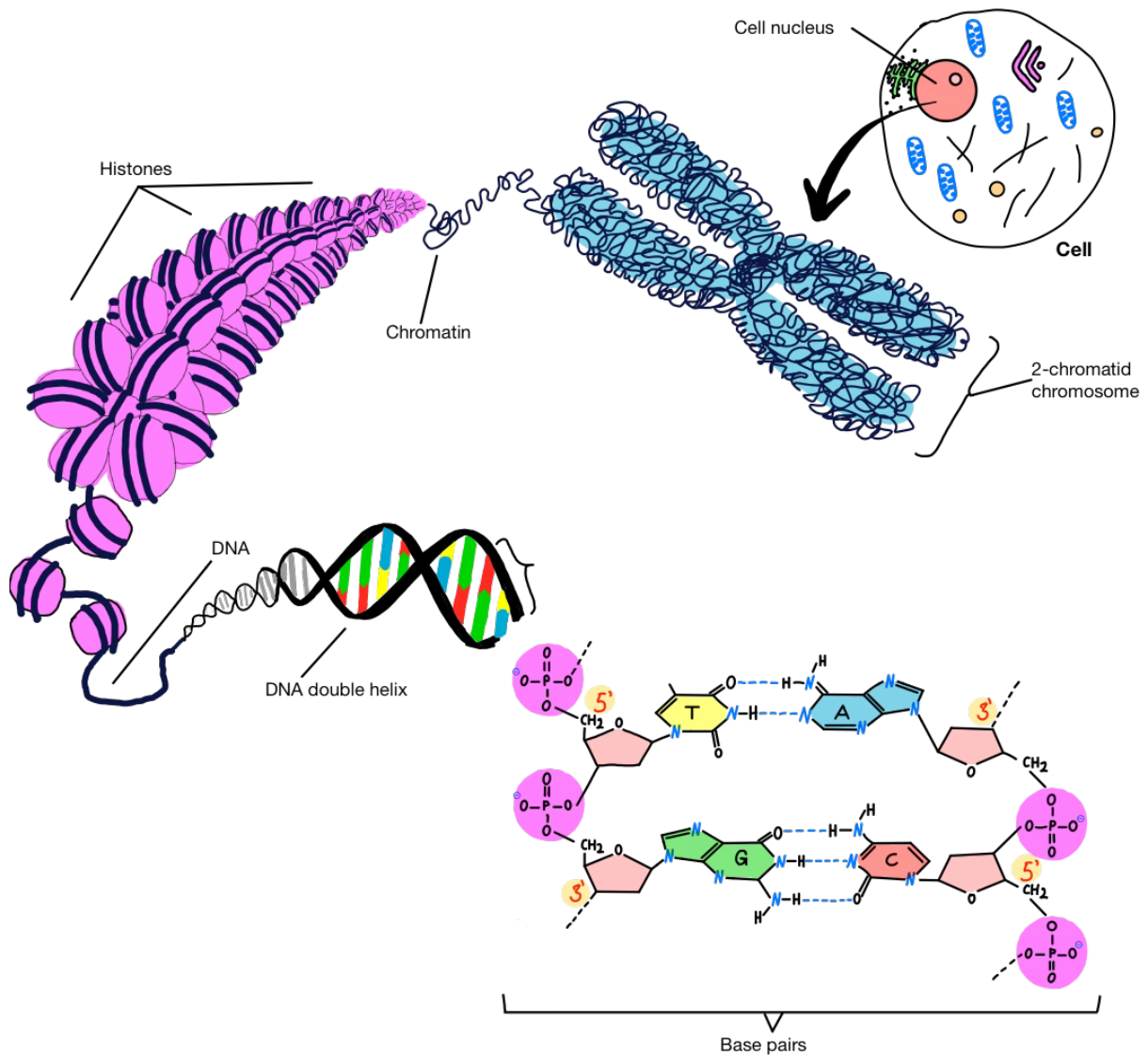


Figure 2: DNA structure and base pairing
 Image courtesy of the author

What is a primer?

A primer is an artificial single strand of DNA, that matches a small part of the two DNA strands. It usually consists of around 15 to 30 nucleotides. It is required during DNA replication and is used in molecular biology and in medicine for PCR and sequencing.^[3-6]

What is the function of a primer?

The primer marks the start for the enzyme polymerase, which synthesises DNA strands. Without such a starting point, the DNA polymerase could not recognise the DNA strand and would therefore not replicate it.

In order to produce a primer, the DNA sequence to be analysed must be known. The primers are produced artificially using computer programs based on the DNA sequence to be analysed. For this purpose, the number of nucleotides a primer should consist of and the prevailing conditions during the PCR are determined. The programme then suggests several primers at best.^[3,4,7,8]

Acknowledgements

This resource has been sponsored by the Joachim Herz Stiftung and produced by the European XFEL.



References

- [1] Watson JD, Crick FHC (1953) [Molecular structure of nucleic acids: A structure for deoxyribose nucleic acid](#). *Nature* **171**: 737–738. doi: 10.1038/171737a0
- [2] Chargaff E (1950) [Chemical specificity of nucleic acids and mechanism of their enzymatic degradation](#). *Experientia* **6**: 201–209. doi: 10.1007/BF02173653
- [3] Mullis KB, Faloona FA (1987) [Specific synthesis of DNA in vitro via a polymerase-catalyzed chain reaction](#). *Methods in Enzymology* **155**: 335–350. doi: 10.1016/0076-6879(87)55023-6
- [4] Saiki RK et al. (1985) [Enzymatic amplification of beta-globin genomic sequences and restriction site analysis for diagnosis of sickle cell anemia](#). *Science* **230**: 1350–1354. doi: 10.1126/science.2999980
- [5] Saiki RK et al. (1988) [Primer-directed enzymatic amplification of DNA with a thermostable DNA polymerase](#). *Science* **239**: 487–491. doi: 10.1126/science.2448875
- [6] Sanger F, Nicklen S, Coulson AR (1977) [DNA sequencing with chain-terminating inhibitors](#). *PNAS* **74**: 5463–5467. doi: 10.1073/pnas.74.12.5463
- [7] Dieffenbach CW, Lowe TM, Dveksler GS (1993) [General concepts for PCR primer design](#). *PCR Methods and Applications* **3**: 30–37. doi: 10.1101/gr.3.3.s30
- [8] Jorapur S, Srivastava A, Kulkarni S (2023) [Evaluating the usefulness of a large language model as a wholesome tool for de novo polymerase chain reaction \(PCR\) primer design](#). *Cureus* **15**: e48336. doi: 10.7759/cureus.48336