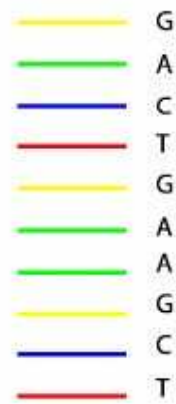


DNA sequencing

Determining the order of bases in a section of DNA is called DNA sequencing. It involves making many copies of the base sequence that match the code of a single strand of DNA. These copies vary in length by one nucleotide; the shortest is only one nucleotide long, and the longest matches the whole entire DNA strand.

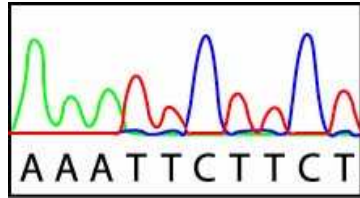
An example follows.

- The double helix of the DNA segment is separated into single strands.
- The single-stranded DNA is mixed with the four nucleotides to construct matching strands.
- Some of each nucleotides are slightly different from the normal nucleotides and are also fluorescent — each type of base is a different colour.
- Many matching strands are constructed. Construction of any one strand stops when a fluorescent nucleotide is added to the sequence.
- The mixture is separated on an electrophoresis gel to produce a sequence of coloured bands.
- The sequence of coloured bands indicates the order of the nucleotides in the constructed strands.



The sequence in the constructed strands can be converted to the sequence of nucleotides in the original DNA strand by using the matching base-pair rules (adenine with thymine and cytosine with guanine).

DNA sequencing results may be viewed on a computer screen as a set of coloured peaks. These peaks also indicate the order of nucleotides in the constructed copies, and the coloured graph is interpreted in the same way as the coloured bands in the electrophoresis results.



In this activity, you will work out the base sequence of two tested DNA segments using each of the sequencing methods.




The following animation can be helpful in understanding DNA sequencing:
www.dnai.org/b/index.html, go to Techniques, then Sorting and sequencing and Cycle sequencing.

You will need:

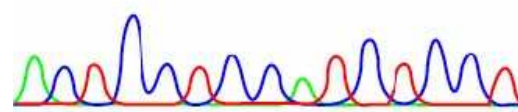

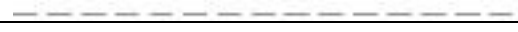
- A colour copy of this worksheet (or black and white copy and a set of 4 coloured pens to construct a coloured version).

What to do:

1. Use the spaces provided in the exercises below to work out the sequence of bases in the tested segment of DNA (yellow = G, green = A, blue = C, red = T).

The pattern of bands	Base sequence in the constructed strands	Base sequence in the tested DNA (use base-pairing rules)
		

2. Compare your answers with someone else in the class. If your answers disagree, work together to correct your answers.

Computer results	
Base sequence in the constructed strands	
Base sequence in the tested DNA (use base-pairing rules)	

For further investigation

Draw your own electrophoresis gel pattern of coloured bands or a computer graph of coloured peaks and ask a partner to work out the sequence of bases in the strand of DNA tested.

Find out why DNA is sequenced and what it can be used for.

Biotechnology Online School Resource