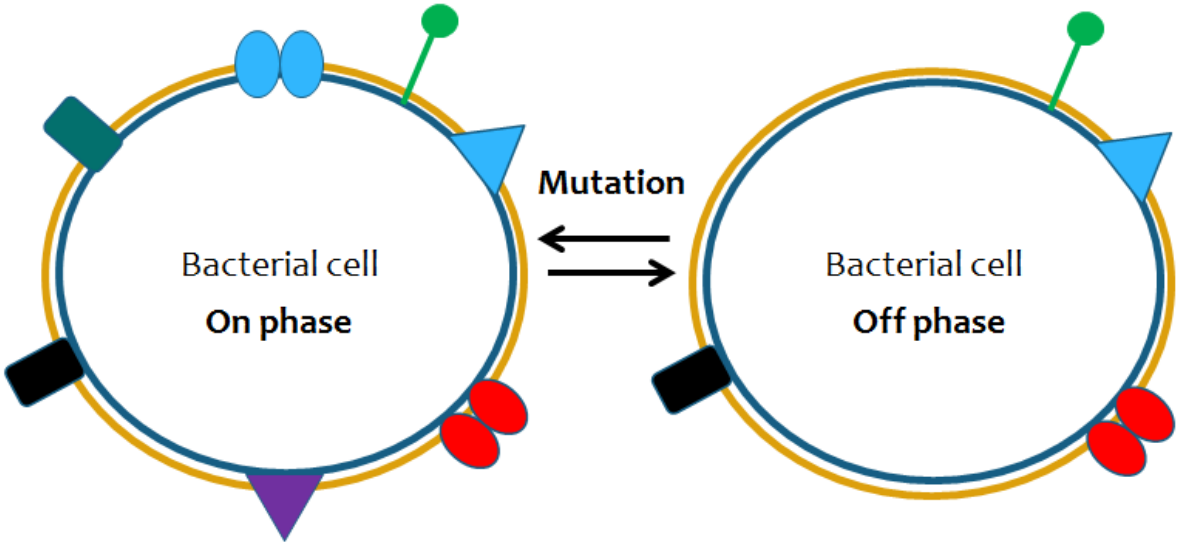
Background: phase variation (optional)

Genetic mutation is the key driver of evolution by natural selection. Some mutations may be deleterious to the harbouring organism, while others may confer a benefit. Mutations are therefore essential for a population to adapt to rapidly changing and hostile environments.

Pathogenic bacteria are subject to many pressures and hostility when invading the human body. Mechanisms to rapidly develop diversity within populations of such bacteria are therefore advantageous. One mechanism conserved throughout many bacterial taxa is known as **phase variation.** Phase variation is the switching of gene expression from an **on** phase to an **off** phase interchangeably leading to a population derived from a single cell, all with different proteomic compositions (see diagram below).

Phase variable switches occur in response to mutations in **hypermutatable,** DNA sequences in either the promoters of phase variable genes or the genes themselves. This mechanism is known as **slipped strand mispairing (SSM).**

Note: there are other mechanisms of phase variation but this is commonly accepted as the most common.



Slipped strand mispairing

Campylobacter jejuni is a Gram-negative bacterium commonly implicated in foodborne gastroenteritis and an excellent model organism for studying phase variation. *C. jejuni* genomes contain ~30 poly-G-nucleotide tandem repeats (poly-G tracts) that are far more prone to insertion/deletion mutations through SSM than other sequences. In SSM, the replication process goes awry and the strands briefly separate and then, when they re-anneal, form a "kink" in either the template strand or the new copy. When the kink forms in the new strand, an extra base is added, whereas when the kink forms in the original strand, one less base is added to the new strand causing a deletion.

These events result in **frame shift** mutations that either, cause a non-sense protein to by synthesized, or a stop codon to be introduced. Both of these are the **off** phase. Insertions and deletions through SSM can just as easily occur in the opposite direction, switching protein expression back into the **on** phase. In the example below (which is actually sequence from C. jejuni strain cj0031/2), deleting a base from the original 10G tract allows translation to continue and the full length, functional gene to be produced.

Off

On

The rate of phase variable mutation varies between strains, and is governed by many factors. Deciphering factors that affect the mutation rate of phase variable genes is an active area of research in Dr Bayliss' group.

The activity on the next page will help you to consider phase variation in a population context and the role that phase variation can play in the infection process.

Activity: simulating phase variation

Accompanying this activity you have been provided with a simulator programme. This simulator will model phase variation in the meningococcus.

When you start the simulator you will begin with a single bacterial cell. This cell has **three** phase variable genes each encoding a virulence determinant.

The cells will begin to divide, and some of them will mutate at a given rate. The cells will continue to divide until there are too many on the screen to continue growing.

In the simulator, each gene is represented by a number (I.E gene 1 is represented by the first number, gene 2 the second number and so on).

If a gene is switched on, its corresponding number will be 1; if it is switched off the number will be 0 (For example, if expression of all 3 genes is switched on, the number will be 111). Running totals of each genotype are provided next to their assigned colour.

Gene 1 encodes Opa

Gene 2 encodes HpuA

Gene 3 encodes HmbR

Genes in the simulator.

Functions of these genes have been previously reported.

To cause disease, there must be at least 30 cells with all three genes switched on (the 111 genotype).

You should use the simulator, alongside searches of the literature to complete the activity provided.

**Note: to get in and out of the simulator you may need to use the 'Win + D' (PC) or 'F11' (Mac) keyboard shortcuts.*