8.3 CRISPR-Cas examples and applications

Slide 1.

The use of CRISPR-Cas in molecular biology goes beyond genome engineering. Therefore, in this presentation I will give you an overview of the already used and potential future applications of this exciting technology.

Slide 2.

The genome engineering potential of CRISPR-Cas has its applications in various domains, from basic biology to biotechnology and medicine.

One of the most well-known applications is the development of new laboratory models to get more insights into human diseases and to apply in the discovery of new drugs. Both modified animals and new cell lines can be efficiently created to mimic the specific phenotypes.

In biotechnology, manipulation of genetic building blocks and metabolic pathways can result in the construction of useful biological systems. For example, improving ethanol production in algae or corn can be an attractive source for renewable energy. Furthermore, manipulating specific biological circuits can result in the generation of useful synthetic materials. Additionally, precise genetic engineering of important crops could make them tastier, more nutritious, pathogen-resistant, or more robust to environmental conditions.

CRISPR-Cas has also its applications in medicine. For example, the technology can be used to develop new antiviral or antibacterial strategies. Another widely discussed application is the direct correction of genetic defects in human genomes in order to address the root cause of genetic diseases, like cystic fibrosis. Nowadays, scientists have already edited the genomes of human embryos. However, this raises some serious ethical concerns.

Slide 3.

Besides genome engineering, CRISPR-Cas is also used for other purposes. One of the earliest applications is strain typing. This technique is based on the high degree of polymorphism in the CRISPR locus, that is the result of the incorporation of new spacers from invading nucleic acids over time.

Here you can see an example of molecular subtyping of *Salmonella* isolates. Previous studies reported the presence of two CRISPR loci in *Salmonella*. By comparing the spacer content of these two loci in five subserotypes of *Salmonella*, all marked with another color, similar spacers and cluster were found, showing that these subtypes share a common ancestor. This result corresponded to the result of another strain typing experiment shown in figure A, confirming the successful use of CRISPR-Cas.

Slide 4.

We can find another interesting application of CRISPR-Cas in the dairy industry. For many years, cheese and yogurt makers have been relying on CRISPR to produce starter cultures that can evade bacteriophage infections, resulting in higher yields and less food waste. For this, spacers that specifically match fragments in the infecting phages are introduced in the starter cells, generating more robust bacterial cultures.

Slides 5.

Recently, the CRISPR-Cas technology has been successfully used to eliminate HIV infection from a 'humanized' model. In this model, mice were transplanted with human immune cells and infected with the virus. The modified Cas9 nuclease together with the specially designed sgRNA were then able to excise the HIV DNA from the genome, eliminating further infection. The next stage would be to repeat the study in primates, an eventually in human patients.

Slide 6.

CRISPR-Cas is also widely used to create new cellular and animal models. One groundbreaking experiment was the engineering of monkey embryos. Monkeys serve as an important model organism for studying human diseases and developing therapeutic strategies. Chinese scientists successfully achieved precise gene targeting in cynomolgus monkeys by co-injecting Cas9 mRNA and sgRNA into one-cell-stage embryos. In this experiment, no off-target mutations were detected, showing the efficiency and reliability of this technique.

Slide 7.

So now the question is: can we use the CRISPR-Cas technology to generate so-called 'designer babies'? The answer is: yes we can. However, we are not there yet and will probably never be, as most of the scientific community is against this because of ethical concerns. In 2015, Chinese scientists performed a first test on human zygotes, using non-viable zygotes containing an extra set of chromosomes. Nonetheless, this led to an international embargo on the use of CRISPR in human embryos. Ignoring this embargo, extra experiments in 2016 were performed. However, no successful results were obtained. The latest study of the Chinese scientists in 2017 was the first performed on viable human embryos. Although this study was very small, the result suggests that CRISPR works much better in normal embryos compared to non-viable zygotes.

Other groups from around the world have also begun editing the genomes of normal human embryos. Recently, scientists in the United states announced that they altered the first human embryos using the CRISPR-Cas technology. Therefore, we can expect that there will be done much more research in the upcoming years on the use of CRISPR in normal human embryos. This might eventually result in the use of CRISPR technology to cure and eradicate deadly diseases.