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CRISPR: A review on hype versus reality

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Abstract- CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats) is a genome editing technology. It is a general notion that if we decode genome, we can decode the key of life. The hope and hype associated with CRISPR carry the same notion. The CRISPR-Cas system, derived from a bacterial adaptive immune mechanism, has emerged as a groundbreaking tool for genome editing. Its precision, efficiency, and adaptability have transformed basic research, biotechnology, and hold immense promise for treating genetic diseases. This review article investigates the fundamental principles of CRISPR-Cas systems, delves into their diverse applications, discusses present challenges, and explores the ethical considerations surrounding this revolutionary technology. CRISPR-Cas systems, once an obscure mechanism of bacterial immunity, have transformed our ability to manipulate the genome with unprecedented precision. This review delves into the evolutionary origins of CRISPR, explores the key technological breakthroughs fueling its applications, and discusses the profound implications for scientific research, biotechnology, and the treatment of genetic diseases. While ethical considerations and technical challenges remain, CRISPR stands poised to rewrite the biological landscape and fundamentally shift our approach to health and disease.

Key words: CRISPR, epigenetic, genome, phenotype, Cas9

INTRODUCTION

CRISPR, an acronym for Clustered Regularly Interspaced Short Palindromic Repeats, refers to sequences found within prokaryotic genomes. These sequences, along with Cas (CRISPR-associated) proteins, form a system with extraordinary targeting capabilities. Initially recognised as a defence against invading viruses, the CRISPR-Cas system can be programmed to target virtually any DNA sequence, revolutionising gene editing through deletions, insertions, and base modifications.[1,2] The most widely used system, CRISPR-Cas9, uses a guide RNA (gRNA) to direct the Cas9 endonuclease to a specific DNA site, inducing a double-stranded break. Cellular repair mechanisms, such as non-homologous end joining (NHEJ) or homology-directed repair (HDR), can then be harnessed to introduce desired genetic alterations.[3] Jennifer Doudna (University of California, Berkeley) and Emmanuelle Charpentier (then at the Max Planck Unit for the Science of Pathogens, Berlin) were awarded the 2020 Nobel Prize in Chemistry for their pioneering work in developing CRISPR-Cas9 as a powerful genetic engineering tool.[4,5] CRISPR-Cas9 technology is rapidly evolving, offering great promise for revolutionising medicine, agriculture, and many other fields. CRISPR-Cas9 is a transformative tool changing how we approach medicine, agriculture, and basic biological research.[6,7] Its incredible potential

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demands ongoing responsible discussion and careful ethical guidance as applications reach into even more complex areas. It's crucial that research moves forward guided by ethical considerations and international agreements. The future of CRISPR promises a dramatic transformation of numerous areas of our lives. However, its widespread and responsible use will depend on the scientific community, policymakers, and the public working together to address the technical and ethical challenges it brings.

MATERIALS & METHODS

Several research papers have been consulted.

DISCUSSION

CRISPR-Cas9 is a revolutionary genome editing tool derived from a bacterial defense system. Here's what makes it so exciting:

What is it?

- CRISPR stands for "Clustered Regularly Interspaced Short Palindromic Repeats" - peculiar patterns found in bacterial DNA.
- Cas9 is an enzyme that acts like molecular scissors, able to cut DNA at specific locations.
- It works like a guided missile: A segment of RNA guides Cas9 to a matching DNA sequence, where Cas9 makes the cut.[8,9]

How does it work?

- Design: Scientists create guide RNA that matches the DNA sequence they want to edit.
- Targeting: The guide RNA leads Cas9 to the target location on the DNA strand.
- Cutting: Cas9 snips the DNA at the desired spot.
- Editing: The cell's natural repair mechanisms can then be used to:
- Inactivate a faulty gene.
- Insert a new gene.
- Correct a genetic error.[10,11]

Why is CRISPR a big deal?

- Precision: Unlike older genetic engineering tools, CRISPR allows for incredibly precise changes to DNA.
- Ease of use: CRISPR is relatively simple and affordable compared to previous methods.
- Versatility: It works in various organisms, from bacteria to plants to humans.[12,13]

Applications of CRISPR

MEDICINE

- **Sickle Cell Disease and Beta Thalassemia:** Clinical trials are underway to use CRISPR to modify blood stem cells, with the goal of alleviating symptoms in patients suffering from these inherited blood disorders.
- **Cancer:** Researchers are engineering immune cells using CRISPR to better recognise and destroy specific tumour cells, leading to personalised cancer treatments.
- **HIV:** CRISPR holds the potential to remove HIV DNA that has integrated into patients' immune cells, offering hope for a functional cure.
- **Genetic Blindness:** Researchers are working towards treating inherited forms of blindness by directly editing the affected genes within the eye.

AGRICULTURE

- **Disease-Resistant Crops:** Scientists are using CRISPR to modify crops, making them resistant to devastating diseases. Examples include fungi-resistant bananas and virus-resistant potatoes.
- **Enhanced Nutrition:** CRISPR can be used to improve the nutritional profile of crops, such as increasing vitamin content in rice or improving oil quality in soybeans.
- **Drought Tolerance:** Editing genes allows researchers to develop plants that withstand challenging conditions like drought or extreme temperatures.

OTHER APPLICATIONS

- **Fighting Against Malaria:** CRISPR is used to create gene drive systems in mosquitoes. These engineered mosquitoes have reduced fertility or an inability to transmit the malaria parasite, potentially curbing the spread of the disease.
- **Organ Transplantation:** Gene editing of pigs with CRISPR aims to create organs that are less likely to be rejected when transplanted into humans, addressing the critical organ shortage problem.
- **Industrial Biotechnology:** Scientists are using CRISPR-edited bacteria and yeasts to produce specialised chemicals, fuels, and even new products for various industries.[14,15]

Challenges and Limitations of CRISPR

- **Off-target Effects:** While the specificity of CRISPR has dramatically improved, unintended edits at non-target locations within the genome remain a concern. Ongoing research focuses on refining CRISPR-Cas systems for increased accuracy.
- **Delivery:** Successfully delivering CRISPR-Cas components into target cells in living organisms poses a significant challenge, particularly for in vivo therapeutic applications. Viral and non-viral delivery methods are actively being explored and refined.
- **Immune Response:** Introduction of CRISPR-Cas components, sometimes derived from bacteria, has the potential to trigger immune reactions that impact delivery and sustained action.[16,17]

Ethical Considerations

- CRISPR raises both excitement and ethical concerns. Questions arise about human germ line editing (heritable genetic changes), potential unintended consequences, access to treatments, and responsible use of the technology. Open dialogue between scientists, policymakers, and the public is crucial to guide the ethical use of CRISPR-Cas.

Some others ethical concerns:

- **Designer Babies:** Altering human embryos creates the fear of altering the human gene pool with unpredictable consequences.
- **Unintended Consequences:** Off-target effects could introduce unforeseen mutations.
- **Social Inequality:** Access to CRISPR therapies could be limited to the wealthy, exacerbating existing disparities.[18,19]

CONCLUSION

CRISPR-Cas systems present a watershed moment in the realm of genetic manipulation. While challenges remain, continuous development and innovation propel this technology forward to treat diseases, improve lives, and reshape our understanding of the fundamental workings of biology. Beyond Cas9, newer CRISPR-associated enzymes and innovative delivery methods promise continued expansion of the CRISPR toolkit. This underscores the urgent need for an interdisciplinary and conscientious approach to navigate the incredible potential and implications of CRISPR technology. Organism does not completely depend on genome, epigenomics plays a key role in development, growth and phenotype. It is similar to C-value paradox. Let's understand it by an example, we know that genes make neurons (proteins) that makes brain, then brain do all function not the genes. During evolution a complex web created between environments and phenotypes. CRISPR can manipulate genome but there is no surety that it is expressed as phenotype. Let's assume that if you want to make a tailless monkey with CRISPR editing, there will always be an uncertainty of its success. It is just a matter of chance that epigenetics favour it or not. Genome to phenotype conversion is more complex than we understand. We need more time, knowledge and research to decode the key of life.

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