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Chapter

Introductory Chapter: Unraveling the Complexities of DNA Replication, Epigenetics, and Gene Therapy Applications

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1. Introduction

"DNA Replication: *Epigenetic Mechanism and Gene Therapy Applications*" is primarily conceived to present the latest trends of the DNA replication research field via presenting the basic fundamentals, involving molecular mechanisms of initiation and termination, functional cross-talk and/or association to the genome, DNA recombination and repair, analytical methods, recent applications and discussing the future directions of the area; a highly-exciting and -timely book to the interested reader, designed to provide a dedicated focus on the clinical contexts and translational processes. Hence, this book aims to bring together and synthesize contributions from active and prominent researchers, with the chapters ranging from the basic molecular biology of the cell, cell growth and division, DNA isolation, duplication, and DNA damage response, to epigenetics, DNA methylation, and gene silencing, to a review of the most recent and advanced (modern) analytical and computational tools, to the relevance of all this to human disease, in general, drug delivery and genomic imprinting in specific and last but not least, a dedicated section to dentistry and oral health, highlighting the revolutionary potential and use of DNA replication, via an applied demonstration in a range of clinical treatments, procedures, and interventions, such as dental pulp inflammation repair, the fate of the dental cells, periodontal therapy, augmented dental implantology, and whole tooth bioengineering. Henceforth, "DNA Replication: *Epigenetic Mechanism and Gene Therapy Applications*" is a comprehensive guide to the latest research and advancements in the field of genetics. The book provides an in-depth exploration of DNA replication, epigenetics, and gene therapy applications, offering a detailed understanding of the mechanisms that govern our genetic makeup and how they can be manipulated to improve human health. The book is a suitable resource for students, researchers, and medical professionals looking to expand their knowledge of genetics and explore new avenues for therapeutic interventions. With contributions from leading experts, it is a must-read for anyone interested in understanding the fundamental processes that underlie our genetic code, in a simplified manner. For instance, the introductory chapter aims to define DNA and RNA methylation, the mechanisms of histone modification, and presents a variety of epigenetic modifications which can lead to anti-tumor drug resistance. It also explores how targeting epigenetic modifiers can reverse drug resistance.

The fields of DNA replication, epigenetics, and gene therapy are all complex and rapidly evolving, with many exciting developments and challenges ahead [1, 2]. Briefly, DNA replication involves the intricate molecular mechanisms that ensure accurate duplication of genetic material, and hence, understanding these processes is crucial for various fields, including genetics, cancer research, and drug discovery [3, 4]. Epigenetics, on the other hand, focuses on the regulation of gene expression through modifications of DNA and its associated proteins and has emerged as a vital area of research with implications for aging, development, and disease [3–7]. Last but not least, gene therapy aims to correct or modify genetic material in order to treat or prevent disease, has the potential to revolutionize medicine [8, 9], and nonetheless also poses significant challenges related to gene delivery, bio-efficacy, and -safety [10–12]. It can be projected that in the future, the integration of these sub-fields will likely continue to drive the advances in personalized medicine, with gene therapy and epigenetic modifications being used to target specific diseases and individual patients [10, 12]. Additionally, ongoing research in DNA replication and repair mechanisms will likely lead to the development of new therapies and drugs for cancer and other genetic diseases [13–15]. Henceforth, it is worth mentioning that while there are still many unknowns and complexities to be navigated, the potential benefits of this research and innovation area are immense and will undoubtedly play a critical role in shaping the future of medicine and biotechnology [1, 3, 6, 10].

2. DNA replication

DNA replication is the process by which a cell makes an identical copy of its DNA [1, 2]. It is a crucial process that occurs in all living organisms and is necessary for cellular division, mitosis, and the transmission of genetic information from one generation to the next [2, 4]. Basically, during DNA replication, the double-stranded DNA molecule is unwound and separated into two strands. Each strand then serves as a template for the synthesis of a new complementary strand by the attachment of nucleotides in a specific order, according to the base-pairing rules (A-T, C-G). The end result is two identical DNA molecules, each consisting of one original and one new strand [1–6]. DNA replication (**Figure 1**) is a complex and highly regulated process crucial for the accurate transmission of genetic information from one generation of cells to the next. It involves the duplication of the entire DNA molecule in a semi-conservative manner, meaning that each new DNA molecule holds one original strand and one newly synthesized strand [1, 4, 6].

3. Epigenetics

Epigenetics is the study of changes in gene expression that occur without any alteration in the DNA sequence itself [7]. These changes can be heritable and reversible and are caused by modifications to the DNA molecule and its associated proteins, which regulate gene expression. Examples of epigenetic modifications include DNA methylation, histone modification, and non-coding RNA molecules. Epigenetic changes can influence gene expression by either activating or silencing genes, and they play a crucial role in various biological processes such as development, aging, and disease. Likewise, epigenetics is a rapidly growing field that has the potential to revolutionize our understanding of gene regulation with effects on many areas of bio-medicine, such as cancer, neurological disorders, personalized-medicine and -dentistry.

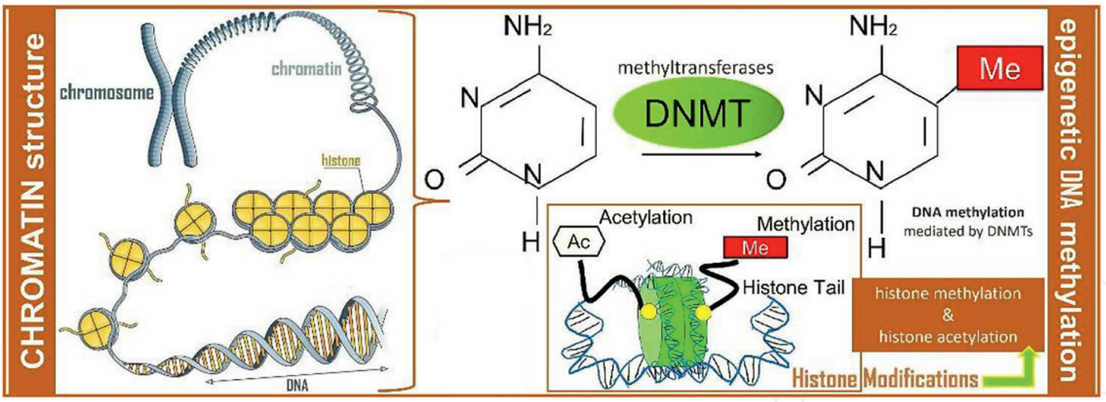

Figure 1.
DNA methylation and replication and some of the involved enzymes and proteins. DNA methylation is a biochemical process that involves the addition of a methyl group (-CH3) to the DNA molecule. It is a fundamental mechanism of epigenetic regulation, which plays a crucial role in gene expression and cellular differentiation. DNA methylation is a dynamic process and primarily occurs at cytosine residues within CpG dinucleotides, where a cytosine is followed by a guanine in the DNA sequence. It involves: DNA methyltransferases (DNMTs): DNA methylation is catalyzed by a group of enzymes called DNA methyltransferases. The two main DNMTs involved in establishing and maintaining DNA methylation patterns are DNMT3A and DNMT3B, responsible for de novo methylation, and DNMT1, which maintains existing methylation patterns during DNA replication; CpG Islands: CpG islands are regions of DNA that contain a high frequency of CpG dinucleotides. These regions are often found near the promoter regions of genes and are involved in the regulation of gene expression. CpG islands can be either unmethylated, associated with active gene expression, or methylated, associated with gene silencing; methylation patterns: DNA methylation patterns can vary across different cell types, tissues, and developmental stages. They are heritable during cell division and can be influenced by environmental factors and cellular signaling pathways. Methylation patterns can undergo dynamic changes throughout life, playing a crucial role in cellular processes such as embryonic development, X-chromosome inactivation, genomic imprinting, and gene regulation; methylation and gene regulation: DNA methylation typically represses gene expression by interfering with the binding of transcription factors and other regulatory proteins to the DNA sequence. Methylated CpG sites can recruit proteins that modify chromatin structure, leading to a condensed chromatin state (heterochromatin) and preventing gene activation. However, DNA methylation can also be associated with gene activation in certain contexts, such as gene bodies; and DNA demethylation: In addition to DNA methylation, DNA demethylation processes exist to remove or alter DNA methylation patterns. Passive DNA demethylation occurs during DNA replication when newly synthesized DNA strands lack methyl groups, leading to a gradual loss of DNA methylation over successive cell divisions. Active DNA demethylation involves enzymatic processes that actively remove or modify methyl groups, including 10–11 translocation (TET) enzymes. Please note that DNA methylation can be influenced by various factors, including environmental cues, hormonal signals, and disease states. The precise patterns of DNA methylation are crucial for normal development and cellular function, and aberrant DNA methylation can be associated with various diseases, including cancer, neurological disorders, and imprinting disorders. On the other hand, the DNA replication process is tightly regulated to ensure accuracy and efficiency. Various checkpoints and control mechanisms exist to monitor the integrity of the DNA and ensure that any errors or damage are corrected before the replicated DNA is passed on to the next generation of cells. During DNA replication, several enzymes and proteins work together to coordinate and carry out the process. Some of the key players involved are helicase: This enzyme is responsible for unwinding the double helix structure of the DNA molecule, separating the two strands and creating a replication fork; DNA polymerase: These enzymes are responsible for synthesizing new DNA strands by adding complementary nucleotides to the existing template strands. There are several types of DNA polymerases involved in different stages of replication, including the main replicative polymerases (DNA polymerase alpha, delta, and epsilon) and the DNA repair polymerases; primase: Primase is an enzyme that synthesizes short RNA primers on the DNA template. These primers provide a starting point for DNA polymerase to initiate DNA synthesis; single-Strand binding proteins (SSBs): These proteins bind to the separated DNA strands and stabilize them, preventing them from re-annealing or being degraded by nucleases; DNA ligase: DNA ligase is responsible for sealing the gaps between the newly synthesized DNA fragments, known as Okazaki fragments, on the lagging strand; topoisomerases: These enzymes relieve the tension and strain that builds up ahead of the replication fork by cutting and rejoining the DNA strands; and DNA proofreading and repair proteins: These proteins monitor the replication process and check for errors. If any errors are detected, they can remove the mismatched nucleotides and replace them with the correct ones. Remember that the precise patterns of DNA methylation are crucial for normal development and cellular function, and aberrant DNA methylation can be associated with various diseases, including cancer, neurological disorders, amongst others.

4. Gene therapy

Gene therapy can be defined as a *biomedical* approach that involves the introduction, removal, and/or alteration of genetic material within the cells of an individual to either treat or prevent disease [8]. Herein, this genetic material can be in the form of DNA, RNA, or proteins and can be delivered to cells using various methods such as viral vectors, liposomes, or nanoparticles (and gene editing tools). The aim of gene therapy is to correct or replace faulty genes, modify gene expression, or introduce new *functional* genes to cells, with the goal of curing or alleviating genetic disorders, as well as some non-genetic diseases such as cancer and infectious diseases [9, 10]. The process typically involves: [1] *Vector Selection* where a suitable vector, often a modified virus or a plasmid, is chosen to deliver the therapeutic gene into the target cells. The vector acts as a vehicle to transport the desired genetic material; [2] *Gene Insertion* where the therapeutic gene or the modified gene is inserted into the chosen vector, which is then introduced into the target cells. Various techniques, such as viral-mediated gene transfer or non-viral methods like electroporation or lipofection, can be used for gene delivery; [3] *Cellular Uptake* where the target cells, depending on the specific strategy, take up the vector carrying the therapeutic gene. Once inside the cells, the vector releases the therapeutic gene; and [4] *Transcription and Translation*: where the introduced therapeutic gene is transcribed into messenger RNA (mRNA) by the cellular machinery, including enzymes involved in DNA replication. The mRNA is then translated into a functional protein. Remember that while gene therapy is not directly related to the process of DNA replication, it does rely on the understanding of DNA replication and the cellular machinery involved in DNA synthesis. Henceforth, gene therapy is an exciting and rapidly evolving field that holds great promise for the treatment of a wide range of diseases [11, 13]. Yet, as with any new technology, it poses some challenges related to the safety, specificity, efficacy, and delivery of genetic material to target cells (gene therapy can lead to activating oncogenes which can cause cancer and the use of viral vectors can also trigger an immune response in the body leading to inflammation amongst other adverse effects). **Note**: The successful integration and expression of the therapeutic gene rely on the cellular processes involved in DNA replication, including the availability of DNA polymerases, DNA repair mechanisms, and other proteins that facilitate DNA synthesis and gene expression [4, 7, 8, 10].

5. Perspective

Today, DNA replication, epigenetics, and gene therapy are at the forefront of cutting-edge research and innovation with many recent advances and promising developments [12, 14, 15]. In laboratories, researchers are working to unravel the complexities of DNA replication, with a particular focus on the mechanisms of initiation and termination, functional cross-talk with the genome, and DNA recombination and repair, for example. Further, new analytical methods, such as single-cell sequencing and CRISPR-based gene editing, are also being developed to enhance our understanding of DNA replication and its role in human health and disease. In the sub-field of epigenetics, recent studies have revealed the potential of epigenetic modifications, such as DNA methylation, histone modification, and non-coding RNA molecules, to be used as therapeutic targets for a range of diseases [15]. For example, recent research has shown that targeting epigenetic modifiers can reverse drug

resistance in certain cancers, while other studies have demonstrated the potential of epigenetic therapies for neurological disorders such as Alzheimer's disease. Gene therapy, meanwhile, is also advancing rapidly from bench-top to bed-side, with a growing number of clinical trials and approved treatments. Recent successes include the development of gene therapies for genetic disorders such as spinal muscular atrophy and inherited retinal disease, as well as for non-genetic diseases such as cancer and infectious diseases. Patents are being filed for new gene therapies, such as those utilizing CRISPR-Cas9 gene editing technology, with an increasing interest in the use of gene therapy for personalizing health care.

In the field of oral health care, dentistry, and cranio-maxillo-facial surgery, in particular, there are many potential applications of DNA replication, epigenetics, and gene therapy [8–11]. Here are a few examples (**Figure 2**): *Dental pulp inflammation repair*: Gene therapy has been investigated as a potential treatment for dental pulp inflammation, which can lead to tooth loss. Researchers have explored the use of gene therapy to deliver anti-inflammatory cytokines to the inflamed pulp tissue, which could promote healing and prevent further damage to the tooth. *Fate of dental-derived mesenchymal stem/stromal cells*: DNA replication and epigenetic modifications play a critical role in the development and differentiation of dental cells. Understanding these processes could help researchers to develop new therapies for conditions such as tooth loss and gum/periodontal disease. *Periodontal therapy*: Epigenetic modifications, such as DNA methylation and histone modifications, have been shown to play a role

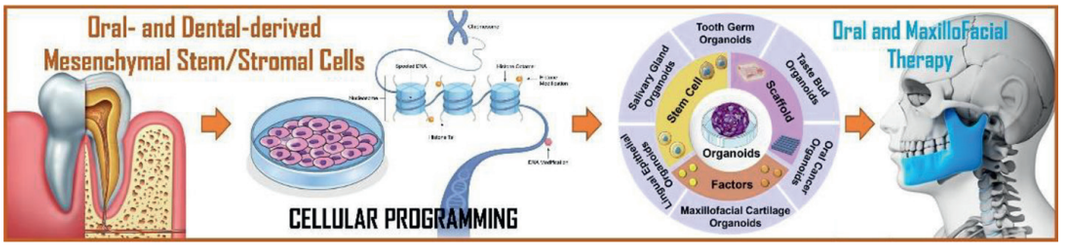

Figure 2.
DNA replication, epigenetics, and gene therapy in Oral, dental, and Cranio-Maxillo-facial applications. While applications do hold promise, please note that they are still under active research and development and innovation (R&D&I). The translation of concepts into clinical practice requires further investigation, ensuring safety, efficacy, and long-term outcomes. Nonetheless, advancements in DNA replication, epigenetics, and gene therapy have the potential to significantly impact Oral health care and cranio-maxillo-facial surgery in the future. Some examples include genetic disorders: Gene therapy holds promise for treating genetic disorders that affect oral health, such as orofacial clefts (e.g., cleft lip and palate) and genetic syndromes associated with dental abnormalities. By introducing functional genes or correcting genetic mutations, gene therapy could potentially address the underlying genetic causes of these conditions; periodontal disease: Epigenetic modifications, including DNA methylation and histone modifications, have been implicated in the development and progression of periodontal disease. Understanding the epigenetic mechanisms involved could lead to the identification of novel therapeutic targets for preventing or treating periodontal disease; salivary diagnostics: Saliva contains DNA from oral tissues, and the analysis of saliva-derived DNA can provide valuable diagnostic information. DNA replication techniques, such as polymerase chain reaction (PCR), are commonly used in salivary diagnostics to detect and quantify specific pathogens or genetic markers associated with oral diseases; tissue engineering and regenerative medicine: DNA replication plays a crucial role in cell proliferation and tissue regeneration. In dentistry and cranio-maxillo-facial surgery, tissue engineering approaches aim to regenerate damaged or lost oral and facial tissues, such as bone, cartilage, and teeth. Gene therapy can be used to enhance tissue regeneration by introducing genes that promote cell growth, angiogenesis, or extracellular matrix synthesis; pain management: Epigenetic modifications can influence pain perception and sensitivity. Understanding the epigenetic mechanisms involved in chronic orofacial pain conditions could lead to the development of targeted therapies for pain management in dentistry; and oral cancer: Epigenetic alterations, including DNA methylation and histone modifications, play a critical role in the development and progression of oral cancer. Epigenetic therapies, such as DNA demethylating agents and histone deacetylase inhibitors, are being investigated as potential therapy options for oral cancer (and others).

in the development and progression of periodontal disease. Researchers are exploring the potential of epigenetic therapies to target these modifications and prevent or treat periodontal disease. *Augmented dental implantology*: Gene therapy has been investigated as a potential approach to enhance the integration of dental titanium implants with the surrounding hard/soft tissue. Researchers have explored the use of gene therapy to deliver growth factors and other molecules to promote osteogenesis and growth of other tissues around the implant. *Whole tooth bioengineering*: Researchers are exploring the use of gene therapy and tissue engineering techniques to develop fully functional teeth that can be implanted in patients who have lost their natural teeth. This could revolutionize oral and dental care, particularly for patients who cannot opt for traditional dental implants due to insufficient bone (quantity/quality).

6. Closing remarks

In conclusion, the integration of DNA replication, epigenetics, and gene therapy has the potential to revolutionize medicine and improve patient outcomes [3, 14, 15]. As researchers continue to make breakthroughs, the translation of these discoveries from bench-top to bed−/chair-side will be crucial for delivering effective treatments to our patients. Indeed, the possible applications of DNA replication, epigenetics, and gene therapy in medicine, oncology, oral and dental health care amongst other related fields are vast, and with ongoing research, development and innovation, state-of-the-art literature, and cutting-edge technologies driving the field forward, it is likely to yield many exciting new discoveries and applications in the years to come.

Conflict of interest

The author declares no conflict of interest.

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References

[1] Ekundayo B, Bleichert F. Origins of DNA replication. PLoS Genetics. 2019;**15**(9):e1008320

[2] Nordman J, Orr-Weaver TL. Regulation of DNA replication during development. Development. 2012;**139**(3):455-464

[3] Peixoto P, Cartron PF, Serandour AA, Hervouet E. From 1957 to nowadays: A brief history of epigenetics. International Journal of Molecular Sciences. 2020;**21**(20):7571

[4] Song H-Y, Shen R, Mahasin H, Guo Y-N, Wang D-G. DNA replication: Mechanisms and therapeutic interventions for diseases. MedComm. 2023;**4**:e210

[5] Delgado-Morales R, Agis-Balboa RC, Esteller M, Berdasco M. Epigenetic mechanisms during ageing and neurogenesis as novel therapeutic avenues in human brain disorders. Clinical Epigenetics. 2017;**9**:67

[6] Muñoz S, Méndez J. DNA replication stress: From molecular mechanisms to human disease. Chromosoma. 2017;**126**(1):1-15

[7] Ganesan A, Arimondo PB, Rots MG, Jeronimo C, Berdasco M. The timeline of epigenetic drug discovery: From reality to dreams. Clinical Epigenetics. 2019;**11**(1):174

[8] Scheller EL, Villa-Diaz LG, Krebsbach PH. Gene therapy: Implications for craniofacial regeneration. The Journal of Craniofacial Surgery. 2012;**23**(1):333-337

[9] Gupta K, Singh S, Garg KN. Gene therapy in dentistry: Tool of genetic engineering. Revisited. Archives of Oral Biology. 2015;**60**(3):439-446

[10] Siddique N, Raza H, Ahmed S, Khurshid Z, Zafar MS. Gene therapy: A paradigm shift in dentistry. Genes (Basel). 2016;**7**(11):98

[11] Fliefel R, Kühnisch J, Ehrenfeld M, Otto S. Gene therapy for bone defects in Oral and maxillofacial surgery: A systematic review and meta-analysis of animal studies. Stem Cells and Development. 2017;**26**(4):215-230

[12] Wilhelm T, Said M, Naim V. DNA replication stress and chromosomal instability: Dangerous liaisons. Genes (Basel). 2020;**11**(6):642

[13] Larsson L. Current concepts of epigenetics and its role in periodontitis. Current Oral Health Reports. 2017;**4**(4):286-293

[14] Zeng B, Liu G, Huang J. DNA methylation and histone modification in dental-derived mesenchymal stem cells. Stem Cell Reviews and Reports. 2022;**18**(8):2797-2816

[15] Zhang H, Fu H, Fang H, Deng Q, Huang H, Hou D, et al. Epigenetic regulation of methylation in determining the fate of dental mesenchymal stem cells. Stem Cells International. 2022:5015856. DOI: 10.1155/2022/5015856. PMID: 36187229; PMCID: PMC9522499