Published in final edited form as:

J Am Dent Assoc. 2009 September ; 140(0 1): 17S–24S.

Science is the fuel for the engine of technology and clinical practice

Malcolm L. Snead, D.D.S., Ph.D. and **Harold C. Slavkin, D.D.S.**

The University of Southern California, School of Dentistry, The Center for Craniofacial Molecular Biology, 2250 Alcazar St., Los Angeles, CA 90033

Abstract

The biological, chemical, behavioral and physical sciences provide the fuel for the engine of innovation and discovery that continuously improve the quality of the human condition. Dramatic breakthroughs in computer power derived from the digital revolution, provides extraordinary computational capacity for diagnostic imaging, bioinformatics (the science of information), and numerous aspects of how we practice dentistry in the 21st century. Moreover, the biological revolution with the elucidation of the structure of DNA, celebrated its 56th anniversary in May ¹, provides the body of knowledge proving itself to be catalytic for discoveries that improve patient care---diagnostics, treatments and therapeutics, and new biomaterials. Humanity's most basic and recognizable characteristics are now understood through the elucidation of our genome and proteome ^{2, 3} the genes and the proteins they encode. We are now beginning to utilize "personalized" genetic predisposition and so much more. These and so many more "ways of knowing" continue to fuel advances in the scientific disciplines that support our dental profession and form the basis for new diagnostic tests, affording new therapies and procedures that improve the quality of life for our patients. And we are today on the thresh hold of stem cell biology applied to regenerative medicine and dentistry. We have entered an era when engineering biological solutions to the loss of tissues and organs as a consequence of disease are now realities.

Keywords

Discovery; molecular biology; chair side application

INTRODUCTION

Science is the fuel for the engine of technology and clinical practice. How do we see and formulate diagnosis and prognosis? What are the ways of treating the diseases and disorders that challenge the human condition? Is one outcome better than another? The answers to these questions come from our sustained investment in science and fuel our educational system. Bright minds exposed to questions like these have and will continue to create new technology that serves to improve patient care.

Correspondence to: Malcolm L. Snead; Harold C. Slavkin.

Conflicts: The authors identify no commercial interests to the content and no financial conflicts of interests.

We think of the “Scientific Revolution” of the sixteenth and seventeenth centuries as the intellectual and technological movement that shaped the modern world, yet the adventure continues now as we live in another time of a scientific revolution, one characterized by great speed and enormous accomplishment in the chemical, physical and biological realms of inquiry---from discovery to application. The 20th century heralds the human accomplishment of understanding and applying the structure and function of deoxyribonucleic acid (DNA) to the principles of cell and molecular biology and to better understand the microbial, as well as human ecosystems and their interdependence ⁴. The scientific disciplines in the 20th century have been most recently shaped by the merging of biology, genetics, engineering, computational sciences with the key ingredient of well trained clinicians who make observations and seek solutions--a mixture that has resulted in making remarkable strides in our understanding of the mechanisms of disease at the molecular level. Now, and into the future, dentistry will rely on science for creation of new diagnostic tests and therapies to catalyze improvements in the care of our patients ⁵⁻¹⁰. Optimizing care for our patients must be the unifying goal of our professional practices.

In this all-too-brief review, we highlight a few select examples of discoveries, drawn from the last 50-60 years to celebrate the 150th Year Anniversary of the American Dental Association. The reader should appreciate that this review is but a small “sampler” of the incredible scientific advances that have shaped what we know, how we think and how we practice clinical dentistry in the dawn of the 21st century.

The Linkage of Dentistry and Genetics

The engine of science has clearly contributed to significant advances by producing the human genome and proteome, the information that contains all the genes and their encoded proteins that make us human. This has resulted in a remarkable precision of diagnostic tests and rapid improvements in patient treatment. One of the most extraordinary scientific discoveries of the 20th century was the discovery of the structure and possible functions of deoxyribonucleic acid (DNA). Significantly, it was a dentist named Norman Simmons who first isolated pure DNA in 1952, which was used by Rosalind Franklin to create the first x-ray crystallography images from DNA. These images led James Watson, Francis Crick and Maurice Wilkins to predict the structure of DNA in 1953 ¹. In Wilkins' acceptance speech for the Nobel Prize in 1962, he credited Norman Simmons for "having refined techniques of isolating DNA and thereby helping a great many workers including ourselves" ¹¹. Norman Simmons received his Bachelor of Science degree (1935) from the City College of New York and a D.M.D. (1939) and Ph.D. (1950) from Harvard. His Ph.D. thesis was entitled “Investigation of Submaxillary Mucoid and the Defenses of the Mouth”¹². Simmons was actually nominated for the Nobel Prize in 1972 for Physiology and Medicine for his fundamental studies of changes in light absorption associated with conformational changes within proteins and polypeptides, the so-called “Cotton Effects” (named after Aime Cotton), that he used to explore the structure of viral particles ¹³. Thereafter his fundamental scientific work in nuclear medicine and oral biology at the University of California at Los Angeles, studies of the isolation of Tobacco Mosaic Virus DNA and RNA, serve as the foundation that subsequently led to numerous nucleic acid and polypeptide biomarkers for disease diagnostics.

Earning a college degree at Columbia University and his dental degree at Washington University, the University of Minnesota School of Dentistry thereafter became the home base for Robert Gorlin for the last half of the 20th century. From this academic perch, Gorlin became the “encyclopedia” of craniofacial birth defects; he deftly integrated clinical observation, phenotypic traits, and the specific gene or genes involved. One of his contributions was the ability to discriminate between syndromic versus non-syndromic birth defects. His memory of craniofacial anomalies was almost as extraordinary as his clinical prowess and he was consulted by dentists and physicians for his diagnostic expertise. Indeed, his diagnostic gifts became internationally known through his lectures, chapters, books and peer-reviewed papers ¹⁴. From the esoteric to the mainstream, Gorlin became trusted as an expert in formulating a diagnosis for craniofacial-oral-dental birth defects. The critical key to his success was his gifted ability to see, to know, and to integrate an array of seemingly disparate information: he saw the "system" of the body, when others saw only derangements of its parts. Gorlin, a dentist, became one of the leading geneticist in the world and the recipient of numerous awards, including the Award for Excellence from the American Society for Human Genetics ¹⁵.

By the end of the 20th century, in no small measure due to the remarkable sensitivity and specificity derived from molecular biology for applications to clinical dentistry, we have begun to identify the specific role of genes various oral diseases. For example, we have learned that autosomal recessive Papillon-Lefevre syndrome, which is characterized by periodontal disease and palmoplantar keratosis and is mainly diagnosed by dentists, is caused by mutation in the cathepsin-C gene ^{16, 17}. Another scientific discovery was the isolation, characterization and clinical application of the major gene for enamel formation, amelogenin. Collaboration of an interdisciplinary team from Baylor School of Medicine and the University of Southern California enabled the first dental gene to be cloned ¹⁸. Isolation of this gene to the X- and Y-chromosomes ¹⁹ provided a forensic tool to discriminate the corporal remains of male versus female, and provided the basis for advancing our understanding of the Mendelian inheritance of enamel birth defects ²⁰. Other major advances in dental genetics include discovery that a gene on chromosome 4 generates three different gene products: dentin phosphoprotein, dentin sialoprotein and dentin glycoprotein ^{21, 22}. The molecular cloning and placement on a human chromosome or mapping of the gene for the second most abundant enamel-forming protein – ameloblastin was accomplished ²³ and the human genome and proteome have been linked at the level of teeth, extending our understanding of normal and abnormal formation of the dentine and enamel bioceramic tissues ²⁴.

Another use of genetic science includes somatic cell gene therapy to treat human disease. At the National Institute for Dental and Craniofacial Research (NIDCR), efforts are currently underway to move genes from the laboratory to chair-side to treat salivary gland diseases ²⁵. It may also be possible to transfer genes to readily accessible salivary glands and use them as “bio-factories” or as a source for replacing deficiencies of proteins that cause other diseases. As dentists, we appreciate that the mouth is readily accessible and that its tissues may provide a relatively easy route to introduce genes to prevent or treat a variety of oral and other diseases. The future holds great promise that many more diseases will have their

genetic basis identified so that specific prevention and treatment may be provided to our patients through gene transfer therapy.

Protein Discovery, Wound Healing, Tissue Repair and Stem Cells – Examples of Science Driving Clinical Practice

We have come a long way since osteoblasts first appeared in primitive bony fish (the ostracoderms); during this evolution, a bony armor formed around the head leaving cavities for the organs of sight, smell, and hearing and, of course, the brain. Eventually, some twenty-two different bones evolved to articulate and form the craniofacial-oral-dental complex. Bone has an essential role in supporting the teeth during mastication but age, disease, trauma, and birth defects all serve to remove bone. Today, our clinical challenge is to devise a strategy to regenerate bone eliminated by birth defects, injury or disease. In the 1960s, it was observed that histiocytes were transformed to osteocytes by "auto-induction", the process in which explanted bone induced new bone formation, often with hematopoietic marrow^{26, 27}. Others showed the capacity of the urinary bladder to induce new bone formation when it came into contact with abdominal muscle cells²⁸ and the biochemical sequences in the transformation of normal fibroblasts into bone cells were described²⁹. The extractable protein that induced new bone was termed "bone morphogenetic protein" (BMP)³⁰ and recombinant DNA technology was used to the service of humankind by identifying a complementary DNA (cDNA) clone for one of the BMPs, allowing this new technology to produce therapeutic amounts of the protein³¹. The commercial availability of BMP protein helped us understand how BMPs works so we can harness its healing powers. Overall, this research allowed this otherwise scarce protein to be manufactured in the lab for use at chairside and bedside^{32, 33}. Efforts to examine the ability of dentine to induce bone led dental researchers to identify a small molecular weight protein isolated from dentine that induced a change in naive cells by inducing them to form cartilage and bone. Rather than a newly discovered BMP, the isolated protein was instead a small, amelogenin protein that was previously thought to participate only in forming the enamel matrix^{34–36}.

The recovery of enamel matrix proteins, mainly amelogenin protein, led to the production of a commercial gel of enamel matrix proteins that can be used to induce progenitor cells to regenerate bone and cementum in the treatment of human periodontal disease³⁷. Enamel matrix proteins and BMPs are now clinically used to recruit and direct resident stem cells to regenerate lost tissues such as periodontal ligament, acellular cementum and alveolar bone. Other dental practitioner /scientists remain engaged in the study of enamel matrix proteins, such as amelogenin and ameloblastin, hoping to discover how these proteins alter cell fate and direct differentiation to form bone and cementum, while also attenuating the inflammatory response^{38, 39}.

Dental researchers working at the NIDCR recovered stem cells from human deciduous teeth, a site that was not previously known to contain such cells.⁴⁰ In a large collaborative effort that marks the intense research needed for progress in this field, stem cells were used in pigs to engineer a functioning cell-mediated root replacement, complete with a periodontal ligament⁴¹. Periodontal ligament stem cells⁴², as well as other sources of stem cells have also been shown to modulate the immune response, offering hope for a new therapeutic tool

for patients suffering from autoimmune diseases such as lupus erythematosus⁴³. Other dental scientists are investigating the use of implant-supported distraction osteogenesis that will prove useful for bone regeneration in craniofacial reconstruction⁴⁴. It has been shown that a unique population of cells with stem-like qualities, known as the neural crest, respond to TGF signals and participate in forming the bones of the head and face, as well as contributing to the sutures that permit growth of the skull^{45 46}. Collectively, this research provides novel insight into the molecular mechanisms that may cause craniofacial anomalies and offers great promise for treatment to improve the quality of life of affected patients.⁴⁷

Diagnostic Imaging

We stand on the shoulders of previous science that has fueled the creative advances in technology of the 20th century. Roentgen accidentally demonstrated that human bones could be imaged and used for dental or medical diagnostics – resulting in the first opportunity to see inside the body without a surgical wound⁴⁸. The first radiograph of the jaw was accomplished just several weeks after Roentgen's discovery by dentist Otto Walkhoff. A remarkable dentist, Edmund Kells, engaged radiography, as well as fitting his dental operatory with electric equipment, compressed air and suction, items, while improved, are still in use today^{49 50, 51}. Following World War II, it was dentist Robert Ledley, a graduate of New York University who worked at the Dental Labs in Washington, D.C., the precursor to the National Institute of Dental Research under the direction of H. Trendly Dean, who revolutionized how we know what we see^{52, 53}. Ledley pioneered computerized tomography (CT) scanning in the early 1950s. He took the scientific discovery of x-rays to a new whole new level of understanding. His inventions, hardware and algorithms for software introduced a 3-dimensional approach by which x-rays were transmitted through varying tissue densities to capture two- and three-dimensional images of all parts of the human anatomy. This remarkable achievement was the precursor of modern imaging diagnosis in both dentistry and medicine.

Wound Healing, Tissue Repair, Atherosclerosis

Remarkable scientific advances have been made in tissue repair, wound healing and tissue regeneration due in no small part to the genius of dentist Russell Ross, who moved on to a distinguished career in pathology, serving for many years as the Chair of Pathology at the University of Washington, School of Medicine. Armed with, at that time, the “new” transmission electron microscopy (TEM), a novel animal model of parabiotic mice, radiology, and an exquisite knowledge of the early advances in wound immunology and pathology, he synthesized the essence of wound healing in his many publications---defining the timing, cytology, physiology, immunology, and connective tissue biochemistry of wound healing during the 1960s–1980s^{54, 55}. His brilliant strategy of using parabiotic mice enabled his team to trace cell origins and cell fate during various stages of wound healing. Simply stated, his team provided the foundation for our modern understanding of wound healing.

During the early 1970s, Ross and his team proposed that localized injury to the lining of the arterial wall was responsible for the unusual accumulation of smooth muscle cells within the

wall of the artery and thereby reduced the lumen of the vessel ⁵⁶. Further, Ross was a catalyst and mobilized interdisciplinary sciences around the problem of atherosclerosis. His team discovered a new growth factor called "platelet-derived growth factor" (PDGF) and further found the cell surface receptor accumulated on smooth muscle cells ^{57, 58}. Curiously, these accumulated smooth muscle cells contained elaborate secretory vesicles that were filled with several types of collagen, fibronectin, metalloproteinases, proteoglycans and fatty acids that assembled into an abundant extracellular matrix associated with atherosclerosis. Ross and his collaborators concluded that atherosclerosis was an inflammatory disease ^{59, 60}. These contributions are examples of how scientific advances in improving the human condition were derived from the passion and creativity of the minds who began their careers in dentistry.

Tissue Destructive Enzymes

Tissue destruction was another focus of significant attention for dental scientists and their efforts to understand tissue loss associated with periodontal disease led to significant improvements in our understanding of enzymatic degradation of collagen, revealing that collagenase was present in the tissues of the human host ⁶¹. These studies had far reaching consequences for other investigators working to understand the destructive process brought about by inflammation. Here, investigators identified a new class of metal containing enzymes, the metalloproteinases, along with their endogenous inhibitory counterparts, formed a "ying" and a "yang" for homeostasis. These studies have impacted the biomedical sciences in many areas, helping to understand such phenomenon as cancer cell metastasis, angiogenesis, as well as the degradation of enamel matrix proteins during enamel biomineralization ⁶²⁻⁶⁴. Studies on the metabolism of the extracellular matrix led to the formulation of an artificial basement membrane, an event that allowed cells to be maintained in a three-dimensional architecture that resembled a native tissue ⁶⁵.

Saliva as a Diagnostic Fluid

As we look to the future, saliva is emerging as an exciting diagnostic tool for future dentists and physicians. For example, dental scientists at UCLA are investigating saliva to aid in the diagnosis of oral cancer ⁶⁶. Many dentists have provided the foundation for using saliva as a diagnostic fluid. Dating back to at least 1960, Irwin Mandel of Columbia University in New York recognized the potential of saliva as an "informative body fluid" ^{67, 68}. His passion to understand saliva was infectious and attracted many talented dental scientists to this field of inquiry. His contributions to science opened opportunities in many areas of biomedical scientific research and clinical practice to diagnose or monitor the progression of disease or treatment using salivary biomarkers. Mandel asserted that saliva, like blood and urine, reflected informative clues about health and disease. His basic work in saliva sampling and analysis provided the framework for the contemporary saliva studies of many investigators. In the mid-1960s, dental scientists found that viral particles can be secreted through the salivary glands, connecting general health with the oral cavity ⁶⁹. Later, others identified the molecular basis for the beneficial effects of saliva on the oral cavity by identifying antimicrobial properties of various salivary proteins ⁷⁰ and provided the foundation for enamel re-mineralization to control caries by identifying salivary proteins that modulate the

maintenance of salivary calcium and phosphorous^{71, 72}. Others have been instrumental in characterizing the salivary proteome (e.g., all of the proteins produced by the salivary gland) and thereby laid a foundation for the use of saliva as a diagnostic fluid since it contains not only salivary proteins, but also proteins from other organs that can be used as surrogate markers for a variety of disease states^{73, 74}.

While working at the State University of New York at Buffalo, Michael Levine spawned several important discoveries regarding salivary proteins and served as the mentor for the training of a platoon of combined DDS and PhD degreed individuals⁷⁵. Levine and colleagues identified the importance of salivary proteins as part of the framework for bacterial adherence to the teeth by bacterial proteins interacting with specific domains within salivary proteins, forming a molecular "Velcro" for adhesion⁷⁶. Others, who began their career at Buffalo include dentist-scientists who went on to contribute to our understanding of streptococci in the aggregation of human platelets and virulence factors associated with bacterial endocarditis^{77, 78}.

The remarkable capacity of the oral and aerodigestive tract cavity to defend itself was identified with studies on the role of mucosal immunity to participate with the adaptive and innate immune response in providing defense against invading pathogens and biofilms⁷⁹. Investigators have shown that immunity could be achieved through a mucosal route of administration, including ingestion that produced a protective response of IgA in salivary secretions in the absence of a detectable IgG response in serum⁸⁰. These findings led others to explore the possibility and the urgent need to produce a vaccine to the most common infection of mankind by streptococci, an organism associated with dental caries^{81, 82}.

Summary

"Dental science" in the 20th century evolved from the crucible of William J. Gies, a biochemist at Columbia University in New York, who convinced the Carnegie Foundation to support an analysis of dental science and education in America, aligned with their previous support of Abraham Flexner's report of the analyses of medicine⁵. The Gies Report was published in 1926 and heralded a new age in American dentistry that would have a foundation in the biological, chemical and physical sciences as found in major universities⁶. During the 20th century, leaders of the American Dental Association helped enable the establishment of a "dental institute" within the National Institutes of Health in Bethesda in 1948. The interdisciplinary work of so-called "dental research" blossomed and became the beacon of dental science for the entire world. The first major scientific achievement of the fledgling dental institute was the use of fluoridation to prevent caries, made possible by H. Trendley Dean (first director of the NIDR)⁸³. Thereafter, the Institute served as the nest and fuel for the fundamental basic science of oral microbiology and immunology, human craniofacial-oral-dental genetics, salivary glands and saliva, connective tissue biochemistry, bone biology, craniofacial biology, microbial genomics and proteomics, oral neoplasia, biobehavioral, pain research and international outreach. The Institute also provides the essential mission of providing scientific training for oral health professionals who continue to grow what is thought and what is taught. To more accurately its evolving research mission, the institute was renamed in 1998 to become the NIDCR. Over the years,

it has bloomed and nurtured many scientists and clinicians to improve the health of Americans. The Surgeon General's Report on America's oral health in 2000 marked the new millennium by emphasizing that good general health must include good oral health ⁸⁴, a mission we celebrate on the 150th anniversary of the American Dental Association.

Acknowledgements

The authors thank their colleagues for the many stimulating conversations and manuscripts that form the foundation of this brief review. We apologize for the numerous exclusions required by space considerations. We note the passion that investigators bring to their work, that they share with their colleagues and that they inoculate into their students. Their traits assure us now that the next Century will be filled with discover and innovation that will improve the care of our patients.

References

1. Watson JD, Crick FH. Molecular structure of nucleic acids, a structure for deoxyribose nucleic acid. *Nature*. 1953; 171(4356):737–738. [PubMed: 13054692]
2. Venter JC, Adams MD, Sutton GG, Kerlavage AR, Smith HO, Hunkapiller M. Shotgun sequencing of the human genome. *Science*. 1998; 280(5369):1540–1542. [PubMed: 9644018]
3. Venter JC, Adams MD, Myers EW, Li PW, Mural RJ, Sutton GG, et al. The sequence of the human genome. *Science*. 2001; 291(5507):1304–1351. [PubMed: 11181995]
4. Chambers DA. Forty years of DNA. *Ann N Y Acad Sci*. 1995; 758:1–11. [PubMed: 7625686]
5. Flexner, A. Medical Education in the United States and Canada: A Report to the Carnegie Foundation for the Advancement of Teaching. Boston: The Merymount Press; 1910.
6. Gies, WJ. Dental Education in the U.S. and Canada: A Report to the Carnegie Foundation for the Advancement of Teaching. New York: Carnegie Foundation for the Advancement of Teaching; 1926.
7. Fields, MJ. Dental Education at the Crossroads, Challenges and Change. Washington, D.C.: National Academy Press; 1995.
8. Harris, RR. Dental Science in a New Age: A History of the National Institute of Dental Research. Rockville, Maryland: Montrose Press; 1989.
9. Slavkin HC. Recombinant DNA technology and Oral Medicine. Paper presented at: DNA the Double Helix: Perspectives and Prospectives at Forty Years. 1995
10. Chambers, DA. DNA The Double Helix: Perspective and Prospective at Forty Years. Chambers, DA., editor. 1995. p. 1-472.
11. Wilkens, MHF. Nobel Prize Award Ceremony. Stockholm, Sweden: Nobel Foundation; 1962. The molecular configuration of nucleic acids.
12. Simmons NS. Studies on the defense mechanisms of the mucous membranes with particular reference to the oral cavity. *Oral Surg Oral Med Oral Pathol*. 1952; 5(5):513–526. [PubMed: 14929529]
13. Simmons NS, Blout ER. The structure of Tobacco Mosaic Virus and its components. *Biophysical Journal*. 1960; 1(1):55–62. [PubMed: 19431302]
14. Cohen MM, Gorlin Robert J Jr. 1923–2006: a remembrance. *Am J Med Genet A*. 2006; 140(23): 2516–2520. [PubMed: 17089425]
15. Gorlin RJ. 2004 ASHG Award for Excellence in Human Genetics Education. And the band played on. *Am J Hum Genet*. 2005; 76(2):216–218. [PubMed: 15714687]
16. Toomes C, James J, Wood AJ, Wu CL, McCormick D, Lench N, et al. Loss-of-function mutations in the cathepsin C gene result in periodontal disease and palmoplantar keratosis. *Nat Genet*. 1999; 23(4):421–424. [PubMed: 10581027]
17. Hart TC, Hart PS, Bowden DW, Michalec MD, Callison SA, Walker SJ, et al. Mutations of the cathepsin C gene are responsible for Papillon-Lefevre syndrome. *J Med Genet*. 1999; 36(12):881–887. [PubMed: 10593994]

18. Snead ML, Zeichner-David M, Chandra T, Robson KJ, Woo SL, Slavkin HC. Construction and identification of mouse amelogenin cDNA clones. *Proc Natl Acad Sci U S A*. 1983; 80(23):7254–7258. [PubMed: 6139813]
19. Lau EC, Mohandas TK, Shapiro LJ, Slavkin HC, Snead ML. Human and mouse amelogenin gene loci are on the sex chromosomes. *Genomics*. 1989; 4(2):162–168. [PubMed: 2737677]
20. Hart S, Hart T, Gibson C, Wright JT. Mutational analysis of X-linked amelogenesis imperfecta in multiple families. *Arch Oral Biol*. 2000; 45(1):79–86. [PubMed: 10669095]
21. MacDougall M, Simmons D, Luan X, Nydegger J, Feng J, Gu TT. Dentin phosphoprotein and dentin sialoprotein are cleavage products expressed from a single transcript coded by a gene on human chromosome 4. Dentin phosphoprotein DNA sequence determination. *J Biol Chem*. 1997; 272(2):835–842. [PubMed: 8995371]
22. Yamakoshi Y, Hu JC, Fukae M, Zhang H, Simmer JP. Dentin glycoprotein: the protein in the middle of the dentin sialophosphoprotein chimera. *J Biol Chem*. 2005; 280(17):17472–17479. [PubMed: 15728577]
23. Krebsbach PH, Lee SK, Matsuki Y, Kozak CA, Yamada KM, Yamada Y. Full-length sequence, localization, and chromosomal mapping of ameloblastin. A novel tooth-specific gene. *J Biol Chem*. 1996; 271(8):4431–4435. [PubMed: 8626794]
24. Hu JC, Yamakoshi Y, Yamakoshi F, Krebsbach PH, Simmer JP. Proteomics and genetics of dental enamel. *Cells Tissues Organs*. 2005; 181(3–4):219–231. [PubMed: 16612087]
25. Cotrim AP, Mineshiba F, Sugito T, Samuni Y, Baum BJ. Salivary gland gene therapy. *Dent Clin North Am*. 2006; 50(2):157–173. vii. [PubMed: 16530055]
26. Urist MR. Bone: formation by autoinduction. *Science*. 1965; 150(698):893–899. [PubMed: 5319761]
27. Reddi AH, Huggins CB. Formation of bone marrow in fibroblast-transformation ossicles. *Proc Natl Acad Sci U S A*. 1975; 72(6):2212–2216. [PubMed: 1056025]
28. Huggins CB. The formation of bone under the influence of epithelium of the urinary tract. *Clin Orthop Relat Res*. 1968; 59:7–19. [PubMed: 4874884]
29. Reddi AH, Huggins C. Biochemical sequences in the transformation of normal fibroblasts in adolescent rats. *Proc Natl Acad Sci U S A*. 1972; 69(6):1601–1605. [PubMed: 4504376]
30. Urist MR, Strates BS. Bone morphogenetic protein. *J Dent Res*. 1971; 50(6):1392–1406. [PubMed: 4943222]
31. Wozney JM, Rosen V, Celeste AJ, Mitsock LM, Whitters MJ, Kriz RW, et al. Novel regulators of bone formation: molecular clones and activities. *Science*. 1988; 242(4885):1528–1534. [PubMed: 3201241]
32. Franceschi RT. Biological approaches to bone regeneration by gene therapy. *J Dent Res*. 2005; 84(12):1093–1103. [PubMed: 16304438]
33. Nussenbaum B, Krebsbach PH. The role of gene therapy for craniofacial and dental tissue engineering. *Adv Drug Deliv Rev*. 2006; 58(4):577–591. [PubMed: 16766080]
34. Veis A, Sires B, Clohisey J, Sabsay B, Amar S. Rat incisor dentine contains a factor which alters the phenotypic expression and stimulates chondrogenesis in fibroblast-like cells in vitro. *Biomaterials*. 1990; 11:35–37. [PubMed: 2397256]
35. Amar S, Sires B, Sabsay B, Clohisey J, Veis A. The isolation and partial characterization of a rat incisor dentin matrix polypeptide with in vitro chondrogenic activity. *J Biol Chem*. 1991; 266(13):8609–8618. [PubMed: 1902476]
36. Nebgen DR, Inoue H, Sabsay B, Wei K, Ho CS, Veis A. Identification of the chondrogenic-inducing activity from bovine dentin (bCIA) as a low-molecular-mass amelogenin polypeptide. *J Dent Res*. 1999; 78(9):1484–1494. [PubMed: 10512382]
37. Hammarstrom L. Enamel matrix, cementum development and regeneration. *J Clin Periodontol*. 1997; 24(9 Pt 2):658–668. [PubMed: 9310870]
38. Hoang AM, Klebe RJ, Steffensen B, Ryu OH, Simmer JP, Cochran DL. Amelogenin is a cell adhesion protein. *J Dent Res*. 2002; 81(7):497–500. [PubMed: 12161464]
39. Gundersen RY, Ruud TE, Jorgensen PF, Scholz T, Reinholt FP, Wang JE, et al. Systemic administration of enamel matrix derivative to lipopolysaccharide-challenged pigs: effects on the inflammatory response. *Surg Infect (Larchmt)*. 2008; 9(2):161–169. [PubMed: 18426348]

40. Miura M, Gronthos S, Zhao M, Lu B, Fisher LW, Robey PG, et al. SHED: stem cells from human exfoliated deciduous teeth. *Proc Natl Acad Sci U S A*. 2003; 100(10):5807–5812. [PubMed: 12716973]
41. Sonoyama W, Liu Y, Fang D, Yamaza T, Seo BM, Zhang C, et al. Mesenchymal stem cell-mediated functional tooth regeneration in swine. *PLoS ONE*. 2006; 1:e79. [PubMed: 17183711]
42. Wada N, Menicanin D, Shi S, Bartold PM, Gronthos S. Immunomodulatory properties of human periodontal ligament stem cells. *J Cell Physiol*. 2009; 219(3):667–676. [PubMed: 19160415]
43. Sun L, Akiyama K, Zhang H, Yamaza T, Hou Y, Zhao S, et al. Mesenchymal stem cell transplantation reverses multiorgan dysfunction in systemic lupus erythematosus mice and humans. *Stem Cells*. In Press.
44. Lypka M, Afshar A, Pham D, Fortman K, Yamashita DD, Yen S. Implant-supported distraction osteogenesis: a technique to advance the deficient maxilla. *J Craniofac Surg*. 2009; 20(2):525–527. [PubMed: 19169158]
45. Chai Y, Maxson RE Jr. Recent advances in craniofacial morphogenesis. *Dev Dyn*. 2006; 235(9): 2353–2375. [PubMed: 16680722]
46. Ishii M, Han J, Yen HY, Sucov HM, Chai Y, Maxson RE Jr. Combined deficiencies of *Msx1* and *Msx2* cause impaired patterning and survival of the cranial neural crest. *Development*. 2005; 132(22):4937–4950. [PubMed: 16221730]
47. Chung IH, Yamaza T, Zhao H, Choung PH, Shi S, Chai Y. Stem cell property of postmigratory cranial neural crest cells and their utility in alveolar bone regeneration and tooth development. *Stem Cells*. 2009; 27(4):866–877. [PubMed: 19350689]
48. Glasser OWC. Roentgen and the discovery of the Roentgen rays. *AJR Am J Roentgenol*. 1995; 165(5):1033–1040. [PubMed: 7572472]
49. Fields RT Jr, Goaz PW. Notes on the history of dental radiology. *Tex Dent J*. 1995; 112(2):15–22. [PubMed: 8633301]
50. Hubar JS. C. Edmund Kells, Jr., pioneer in the field of dental X-rays. *J Hist Dent*. 2000; 48(1):11–15. [PubMed: 11794327]
51. Langland OE. C. Edmund Kells. *Oral Surg Oral Med Oral Pathol*. 1972; 34(4):680–689. [PubMed: 4560600]
52. Ledley RS, Ayers WR. Computerized Medical Imaging and Graphics evolves from Computerized Tomography. *Comput Med Imaging Graph*. 1988; 12(1):v–xviii. [PubMed: 3383154]
53. Ledley RS. Innovation and creativeness in scientific research: my experiences in developing computerized axial tomography. *Comput Biol Med*. 1974; 4(2):133–136. [PubMed: 4616781]
54. Odland G, Ross R. Human wound repair. I. Epidermal regeneration. *J Cell Biol*. 1968; 39(1):135–151. [PubMed: 5678445]
55. Ross R, Odland G. Human wound repair. II. Inflammatory cells, epithelial-mesenchymal interrelations, and fibrogenesis. *J Cell Biol*. 1968; 39(1):152–168. [PubMed: 5678446]
56. Ross R, Glomset JA. Atherosclerosis and the arterial smooth muscle cell: Proliferation of smooth muscle is a key event in the genesis of the lesions of atherosclerosis. *Science*. 1973; 180(93): 1332–1339. [PubMed: 4350926]
57. Ross R, Glomset J, Kariya B, Harker L. A platelet-dependent serum factor that stimulates the proliferation of arterial smooth muscle cells in vitro. *Proc Natl Acad Sci U S A*. 1974; 71(4):1207–1210. [PubMed: 4208546]
58. Ross R, Vogel A. The platelet-derived growth factor. *Cell*. 1978; 14(2):203–210. [PubMed: 352535]
59. Ross R. Atherosclerosis—an inflammatory disease. *N Engl J Med*. 1999; 340(2):115–126. [PubMed: 9887164]
60. Ross R. The pathogenesis of atherosclerosis: a perspective for the 1990s. *Nature*. 1993; 362(6423): 801–809. [PubMed: 8479518]
61. Fullmer HM, Gibson W. Collagenolytic activity in gingivae of man. *Nature*. 1966; 209(5024):728–729. [PubMed: 5924404]
62. Birkedal-Hansen H, Cobb CM, Taylor RE, Fullmer HM. Serum inhibition of gingival collagenase. *J Oral Pathol*. 1974; 3(6):284–290. [PubMed: 4377156]

63. Sabeh F, Ota I, Holmbeck K, Birkedal-Hansen H, Soloway P, Balbin M, et al. Tumor cell traffic through the extracellular matrix is controlled by the membrane-anchored collagenase MT1-MMP. *J Cell Biol.* 2004; 167(4):769–781. [PubMed: 15557125]
64. Caterina JJ, Skobe Z, Shi J, Ding Y, Simmer JP, Birkedal-Hansen H, et al. Enamelysin (matrix metalloproteinase 20)-deficient mice display an amelogenesis imperfecta phenotype. *J Biol Chem.* 2002; 277(51):49598–49604. [PubMed: 12393861]
65. Martin GR, Kleinman HK, Terranova VP, Ledbetter S, Hassell JR. The regulation of basement membrane formation and cell-matrix interactions by defined supramolecular complexes. *Ciba Found Symp.* 1984; 108:197–212. [PubMed: 6240392]
66. Zimmermann BG, Wong DT. Salivary mRNA targets for cancer diagnostics. *Oral Oncol.* 2008; 44(5):425–429. [PubMed: 18061522]
67. Ellison SA, Mashimo PA, Mandel ID. Immunochemical studies of human saliva. I. The demonstration of serum proteins in whole and parotid saliva. *J Dent Res.* 1960; 39:892–898. [PubMed: 13726515]
68. Mandel ID. The diagnostic uses of saliva. *J Oral Pathol Med.* 1990; 19(3):119–125. [PubMed: 2187975]
69. Madonia JV, Bahn AN, Calandra JC. Salivary excretion of Coxsackie b-1 virus in rabbits. *Appl Microbiol.* 1966; 14(3):394–396. [PubMed: 5970825]
70. Oppenheim FG, Xu T, McMillian FM, Levitz SM, Diamond RD, Offner GD, et al. Histatins, a novel family of histidine-rich proteins in human parotid secretion. Isolation, characterization, primary structure, and fungistatic effects on *Candida albicans*. *J Biol Chem.* 1988; 263(16):7472–7477. [PubMed: 3286634]
71. Schlesinger DH, Hay DI. Complete covalent structure of statherin, a tyrosine-rich acidic peptide which inhibits calcium phosphate precipitation from human parotid saliva. *J Biol Chem.* 1977; 252(5):1689–1695. [PubMed: 838735]
72. VanDyke TE, Levine MJ, Herzberg MC, Ellison SA, Hay DI. Isolation of a low molecular weight glycoprotein inhibitor of calcium phosphate precipitation from the extra-parotid saliva of macaque monkeys. *Arch Oral Biol.* 1979; 24(2):85–89. [PubMed: 299145]
73. Kauffman DL, Bennick A, Blum M, Keller PJ. Basic proline-rich proteins from human parotid saliva: relationships of the covalent structures of ten proteins from a single individual. *Biochemistry.* 1991; 30(14):3351–3356. [PubMed: 1849422]
74. Wong RS, Hofmann T, Bennick A. The complete primary structure of a proline-rich phosphoprotein from human saliva. *J Biol Chem.* 1979; 254(11):4800–4808. [PubMed: 438215]
75. Scannapieco FA. Salivary biochemistry in Buffalo: the legacy of Michael J. Levine. *J Dent Res.* 2003; 82(2):76–81. [PubMed: 12562877]
76. Murray PA, Levine MJ, Tabak LA, Reddy MS. Specificity of salivary-bacterial interactions: II. Evidence for a lectin on *Streptococcus sanguis* with specificity for a NeuAc alpha 2, 3Gal beta 1, 3GalNAc sequence. *Biochem Biophys Res Commun.* 1982; 106(2):390–396. [PubMed: 7104000]
77. Herzberg MC, Gong K, MacFarlane GD, Erickson PR, Soberay AH, Krebsbach PH, et al. Phenotypic characterization of *Streptococcus sanguis* virulence factors associated with bacterial endocarditis. *Infect Immun.* 1990; 58(2):515–522. [PubMed: 2137112]
78. Herzberg MC, Brintzenhofe KL, Clawson CC. Aggregation of human platelets and adhesion of *Streptococcus sanguis*. *Infect Immun.* 1983; 39(3):1457–1469. [PubMed: 6188697]
79. Costerton JW, Stewart PS, Greenberg EP. Bacterial biofilms: a common cause of persistent infections. *Science.* 1999; 284(5418):1318–1322. [PubMed: 10334980]
80. Michalek SM, McGhee JR, Mestecky J, Arnold RR, Bozzo L. Ingestion of *Streptococcus mutans* induces secretory immunoglobulin A and caries immunity. *Science.* 1976; 192(4245):1238–1240. [PubMed: 1273589]
81. Taubman MA, Nash DA. The scientific and public-health imperative for a vaccine against dental caries. *Nat Rev Immunol.* 2006; 6(7):555–563. [PubMed: 16729013]
82. Smith DJ, Taubman MA, Ebersole JL. Local and systemic antibody response to oral administration of glucosyltransferase antigen complex. *Infect Immun.* 1980; 28(2):441–450. [PubMed: 6447110]
83. Dean HT. Fluorine in the control of dental caries. *J Am Dent Assoc.* 1956; 52(1):1–8. [PubMed: 13271016]

84. Evans CA, Kleinman DV. The Surgeon General's report on America's oral health: opportunities for the dental profession. *J Am Dent Assoc.* 2000; 131(12):1721–1728. [PubMed: 11143736]

Biographies

U.S. National Library of Medicine