THE NEW MOLECULAR MEDICINE

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Nuclear medicine is based on the concept that diseases can be characterized by in situ molecular abnormalities. Advances in our understanding of intra- and intercellular communication are revolutionizing the practice of medicine. In the future, diagnosis, treatment and the monitoring of the response to treatment will be molecular. NanoDx probes will search out molecular abnormalities within different organs and parts of organs. NanoRx probes will correct them.

Modern scientific medicine is moving away from an organ orientation, and is advancing by leaps and bounds through a holistic approach to disease, exemplified by revolutionary advances in human genetics, immunology, oncology, endocrinology, neuropsychiatry, and molecular biology, all of which are holistic approaches to disease. The new, integrated, global approach of nuclear medicine goes beyond anatomy and gross pathology to physiology and molecular medicine. In the future, localized diseases are likely to become the exclusive domain of surgeons, a field in which an anatomical orientation will continue to advance dramatically through advances in anatomical and pathological imaging.

In the practice of molecular medicine, patients' problems can be viewed as molecular dysfunction, not structural abnormalities (structure and function converge at the molecular level). Molecular "slices of life" provided by nuclear medicine will join histopathology as a way to characterize disease.

The practice of medicine remains strongly influenced by a concept described 150 years ago by the great physiologist, Claude Bernard – the concept of the extracellular fluid as the milieu interieur, or internal environment – which maintains a constancy which buffers the cells of the body from the changes that occur constantly in the external environment. Examination of the constituents of blood and urine, commonplace in medical practice today, reflects Bernard's concepts. Nuclear medicine extends Bernard's concepts by making it possible to study the cells themselves, including their importantant interface with the extracellular fluid that surrounds them. Radioactive tracers make it possible to examine regional blood flow, cellular bioenergetics and intercellular communication, the latter involving membrane and intracellular receptors, enzymes and substrate transport systems.

Although the number of radiotracers has expanded greatly, the principles of "molecular nuclear medicine" remain the same as when the first studies of the thyroid were carried out half a century ago. By studying the accumulation of radioactive iodine by the thyroid gland, it was possible to examine the accumulation of environmental iodine and track the formation of thyroxine and other thyroid hormones. The first study in which I was involved in nuclear medicine was the mapping of the accumulation of radioiodine in thyroid nodules with a hand held Geiger tube positioned at 1 cm intervals over the neck in order to determine whether a nodule of the thyroid gland was functional, that is, accumulating radioactive iodine. This laborious, time-consuming task was simplified by the invention of Benedict Cassen, who in 1951 invented the rectilinear scanner in which a crystal radiation detector was automatically moved back and forth over the neck for the portrayal of regional thyroidal function. Its commercial development occurred in the late 1950's.

In 1960, the radionuclide, technetium 99-m/molybdenum generator was developed by Stang and Richards, and advertised for sale by the Brookhaven National Laboratory at a time when no one had any idea of how the technetium-99m would be used. In 1963, Paul Harper at the University of Chicago recognized the technetium-99m's emission of 140 keV photons, decay by isomeric transition, and 6 hour half life were ideal for use in nuclear medicine.

In 1958, Hal Anger described the first scintillation camera at the meeting of the Society of Nuclear Medicine in the United States, but his exhibit drew little interest. Nevertheless, the combination of technetium 99-m and Anger's scintillation camera, and the movement of industry into the field led to a logarithmic phase of growth that continued until the 1970's, at which time a plateau was reached, related chiefly to the development of computed tomography (CT). This brought about a great decrease in nuclear brain scanning, and the first exodus from nu-

clear medicine. The development of positron emission tomography (PET) by TerPogossian, Phelps and Hoffman at Washington University moved nuclear medicine into a new logarithmic growth phase, and encouraged industry to develop single photon emission computed tomography (SPECT) to facilitate the extension of PET successes into more widespread clinical use. TerPogossian had been among the first to recognize the potential of the cyclotron for producing short-lived radiotracers, such as oxygen-15, carbon-11 and fluorine-18.

Whitehead has written: "It is a well-founded historical generalization that the last thing to be discovered in any science is what the science is really about. Men go on groping, guided merely by a dim instinct and a puzzled curiosity until at last some great truth is loosened." Nuclear medicine has become molecular medicine. Its images are concerned with the temporal as well as spatial distribution of the molecules of the living human body. Health requires the proper integration of hundreds of billions of chemical reactions. Proper functions require proper chemistry, and diseases can be defined at the molecular as well as cellular level, often before structural changes have occurred.

Nuclear medicine has the technology that can extend to medical practice the enormous advances in molecular biology and genetics. Two principles of genetics are of special relevance to nuclear medicine: pleotropism and genetic heterogeneity. Pleotropism is the condition in which a single gene defect does not affect a single tissue or organ, or even a single organ system, but can lead to many different manifestations of disease. Genetic heterogeneity describes the fact that abnormalities in different genes can result in the same clinical syndrome. Thus, it is necessary to examine molecular phenotypes as well as genotypes – if one is to see the whole picture. Genes are blueprints for molecules. Genes don't cause disease directly. They express themselves through molecules. The genome is the map; nuclear medicine images the territory.

The microscope revealed the neuron as the fundamental unit of the nervous system. PET/SPECT and more simple radiation detectors make it possible to examine molecular intercellular communication – to examine how neurons (and other cells) "talk" to each other. Electrical "action potentials" transmit information along axons, and there is some electrical neuron-to-neuron communication, but most is by molecular "messengers", carrying information among the 1,000 to 10,000 synapses that connect hundreds of billions of neurons in the human brain. Membrane receptors are involved in the recognition and transfer of information between presynaptic neurons and second and subsequent "messengers", including channels through which sodium, calcium and potassium ions travel. Energy-carrying molecules, such as adenosine tri-phosphate (ATP), modulate the over-all activity of the neurons. With evolution, the increasing complexity of the nervous systems does not seem to have resulted in the development of new neurotransmitters, but rather in the development of incredibly more complex circuitry of neuronal connections, most of which involve molecules, including biopolymers, which "recognize" specific "messenger" molecules that are released locally or circulate through the body until they encounter the appropriate biopolymer on the surface of neurons or other cells which fit their specific configuration. The neurotransmitters have the right shape, charge, and other physicochemical properties to bind to the receptor biopolymers. The patterns and quantities of these "recognition sites" integrate individual cells of the body to make the person a unique, whole individual. "Disintegration" results in disease or death. In the brain, neurotransmitters and receptors affect how we think, feel and act. Molecules are the words that make up the story of our lives – past and present.

The maintenance of life requires intercellular communication, which in turn requires energy, that needed to generate the ion gradients that produce electrical "action potentials" and that needed to synthesize transmitters and receptors. The rate of consumption of the principle source of energy – glucose – can be measured by positron emission tomography.

Since the 1950's, more than 100 neurotransmitters, or "chemical messengers", such as serotonin and dopamine, have been discovered. Many single neurotransmitters, such as acetyl choline, dopamine or serotonin, have different effects on different receptors. For example, acetyl choline stimulates skeletal muscle cells to contract, but causes heart muscles to relax. Muscarinic acetyl choline receptors are excitatory; nicotinic acetyl choline receptors are inhibitory.

The production of the molecules of life is directed by the desoxyribose nucleic acids (DNA) within each cell – including somatic cells – that serve as blueprints for the growth, development, and repair of the molecules that make up our bodies. Molecules must repair themselves when injured, and reproduce themselves when they wear out. When the processes stop, we age or die. Wrinkled skin, poor healing of wounds, and poor memory reflect damaged molecules that accumulate with aging. Antibiotic molecules may be needed to selectively overcome the effects of invading microorganisms. Other molecules may be needed to stimulate or inhibit defective chemical reactions. Many drugs act by stimulating or blocking "recognition sites" on the surface of cells, such as cimetidine that blocks histamine receptors, propranolol that blocks nor-epinephrine receptors, and haloperidol that blocks dopamine receptors. One of the characteristics of chemical receptors, such as those on post-synaptic neurons, is their exceedingly low concentrations, in the range of picomoles/gram. The great sensitivity with which radioactivity can be measured is needed in order to measure receptor concentrations in different parts of the body, such as the brain.

Not only administered drugs, but naturally-occurring neurotransmitter molecules, affect intercellular communication. These include amines, such as nore-

pinephrine, dopamine and serotonin; amino acides, such as gamma-aminobutyric acid (GABA), glutamic acid, aspartic acid, and glycine, and peptides, such as endogenous enkephalins. Neurotransmitters are secreted in varying amounts depending on the number and rate of electrical impuses traveling down the axon of the presynaptic neuron from which the neurotransmitter is secreted. Neurotransmitters bind to enzymes that degrade the unbound neurotransmitter.

In addition to involvement in neuron-to-neuron information transfer, neurotransmitters, including enkephalins, act as modulators of regional neuronal activity. Some chemical "messengers" act within fractions of a second while others have an effect over hours or even days. Thousands of synapses connecting with a single post-synaptic neuron are integrated and determine whether the post-synaptic neuron fires an action potential. The availability of 20 different amino acids means that a vast number of different combinations are possible, and can encode a large amount of information.

Each cell of the body contains billions of molecules of thousands of different types. At birth, molecular blueprints are encoded in the inherited DNA within each cell of the body, making up the "genome" or "genotype", which determine cell structure and, eventually, function. The location of over 1,800 genes on specific chromosomal regions is known. As a result of the "human genome" project, established in the United States in 1988, eventually all 50,000 or more genes will have been assigned locations on chromosomes. Approximately 4,000 human diseases are believed to be genetic in orgin.

The human genome initiative is a joint effort sponsored by the National Institutes of Health (NIH) and the Department of Energy (DOE). The proposal for the project was made originally by the DOE in order to study the genetic effects of radiation on DNA. The NIH was included because of the potential impact on health care. According to Victor A. McKusick: "What Vesalius' anatomy text of 1543 was to medicine in the past, the anatomy of the human genome is to medicine today – a body of knowledge that will inform all future clinical practice."

Where does nuclear medicine fit into the picture? Genes direct the production of molecules that make up the structure of the body, provide energy or carry information. Genetic mutations result in biochemical abnormalities, such as enzyme deficiencies. For example, an abnormality in the gene that signals the development of the pigment melanin results in the phenotype called albinism. Such molecular abnormalities have been identified in about 430 disorders out of the over 1 million classifiable human diseases. Mutations occur in human being at a rate of about one in one hundred thousand cell divisions and affect about one in a million genes per generation. Thus, there is approximately one mutation per 20 persons per generation. Approximately 2,000 specific mutations have brought about identifiable human diseases. About 300 of these affect the brain.

Radioactive tracers make it possible to detect and quantify the molecular abnormalities that the genes bring about. The molecular studies are needed because (as described above), abnormality of a single gene may produce several different types of diseases, or phenotypes. One can illustrate the role of nuclear medicine in the field of cancer. Multiple genes are involved, some resulting in the excessive production of growth factors, and other involving deficient production of growth suppressor molecules, such as somatostatin.

Cancer is the result of genetic and environmental factors. For example, in small cell cancer of the lung, a genetic defect sets the stage for an environmental factor, such as smoking, that finally results in the disease. Many types of cancer, including carcinoid, neuroendocrine cancers, and almost half of the cancers of the breast contain increased concentrations of somatostatin receptors. The increase in somatostatin receptors suggests that a deficiency in the growth-suppressor somatostatin may be the cause of the increase – "up-regulation" – of somatostatin receptors.

Geneticists search for genetic abnormalities in patients with specific syndromes. Another approach, called "reverse genetics", is to search for manifestations of disease when abnormal genes are found. Initially, the search is for an abnormal protein, such as an enzyme. Then the search is for a clinical disorder.

In the 1950's and 60's, inborn errors of metabolism could be detected by amino acid analysis, spectroscopy, spectrophotometry, and the use of newly available radioisotopes. Abnormalities of proteins included enzymes, receptors, and structural proteins. In some cases, a predisposition to disease can be identified by finding an abnormal gene locus even before birth, while the fetus is still in utero. Nuclear medicine makes it possible to extend the search to the molecules involved in intercellular communication.

As described above, there is not a one-to-one relationship between a gene and a disease. As McKusick has written, "it would be ludicrous to conclude that when we have determined the last nucleotide in the human genome, we will know all it means to be human." Radioactive tracers play a major role in trying to connect the genome to human disease. Nuclear medicine can be defined as "in vivo molecular medicine".

Molecular nuclear medicine not only makes it possible to classify disease biochemically, it also provides a new approach to the design and development of drugs. One can determine the relationship between a specific molecular configuration and the in vivo biochemical effects of a drug. The effects of drugs on energy metabolism, synthetic processes, communication and regulatory mechanisms can be expressed in molecular terms. Such knowledge not only provides a more homogeneous selection of patients, but also a way to plan and monitor drug treatment. In the past, pharmacological research consisted of an alliance between organic chemists to make new compounds and pharmacolo-

gists to screen them for possible effectiveness in animals. Molecular nuclear medicine is now involved in drug design, development, evaluation, and monitoring in human beings, as well as animals.

Nuclear medicine techniques can be used to assess the effectiveness of surgery or radiation therapy, and can document the extent of tumors, and progression or regression in response to different forms of treatment. Such data permit modifications of the treatment plan sooner than can be determined by the clinical response of the patients or changes in the size of the lesions. Thus, treatment need no longer be based solely on clinical response, gross morphology or the lesions and histopathological examination of biopsies.

An important characteristic of neoplastic tissue is its increased rate of cell division. In general, accumulation of thymidine into neoplasms is increased in the presence of increased DNA synthesis. Amino acid transport across tumor cell membranes has also been found to differentiate many malignant from non-malignant tumors. Not only membrane transport, but also protein synthesis can be examined if suitable mathematical modeling is used in data anlysis. The accumulation of fluoro-deoxyglucose can be used to measure both aerobic and anerobic regional glucose utilization. Many malignant tumors have accelerated glycolysis compared to surrounding tissues.

In addition to measuring blood flow to tumors, blood volume, substrate incorporation or DNA synthesis, PET and SPECT can be used to measure the number and affinity of hormone receptors which characterize certain tumors. Estrogen receptors are increased in many breast tumors, in both the primary and metastatic sites. Dopamine receptors are often increased in pituitary adenomas.

Fluorine-18 estradiol accumulation as determined by PET makes it possible to tailor the treatment of a specific patient on the basis of the number of estrogen receptors. A tumor containing estrogen receptors is more likely to be treated successfully with estrogen-receptor blocking drugs, such as Tamoxifin, than cancers which do not contain estrogen receptors. The presence of progesterone receptors as well as estrogen receptors is the best prognostic sign. Radioactive tracers that bind to estrogen receptors make it possible to assess the status of the primary breast cancer and regional metastatic deposits. Histopathology alone need no longer be the only criterion for diagnosis, prognosis and therapy.

Receptors are also found on pituitary tumors. Using the depamine receptor binding agent (11-C)-N-methylspiperone, it has been possible to classify pituitary adenomas according to whether they possess dopamine receptors. If the tumors contain such receptors, they can be treated chemically rather than surgically, that is, by administering the dopamine receptor agonist, bromocryptine.

After treatment, measurement of the metabolic activity of the tumor makes it possible to detect persistence or recurrence of the tumor and damage to normal brain tissue, such as that resulting from radiation. For example, (11-C)-methionine is useful for delineating the boundaries of brain tumors, providing information of value in the planning and performance of brain surgery, by permitting differentiation of the metabolizing brain tumor from simple disruption of the blood brain barrier.