IS CELL MEMBRANE RECEPTOR PROTEIN DOWN-REGULATION ALSO A HYDRODYNAMIC PHENOMENON?

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ABSTRACT: It has been argued that the histaminergic neuronal system, the serotonergic neuronal system, and the central nervous renin-angiotensin system stimulated by them, are involved in the primary physiological drives for water regulation of the body. Since, next to oxygen, water is the single most essential substance for the maintenance of the homeostatic balance of the body, it would be logically imperative to satisfy the natural physiological drive of these systems, instead of blocking their activity. Before and during the time medical treatment procedures are being evaluated, the water regulatory function of these systems have to be considered. It has also been argued that from an early period of life, thirst sensation loses its fine regulatory control of water intake, until an incipient dehydration establishes and becomes a chronic physiological state in the body, to the point that from age twenty to seventy, the ratio of extracellular to intracellular water content of the body changes from a figure of approximately 0.8 to 1.1. Since water intake regulates intracellular fluid balance and sodium regulates the extracellular fluid volume, it would be logical to assume that inadequate water intake causes a very gradual chronic intracellular free water deficiency. It seems, since intracellular viscosity becomes gradually increased, as a result of cellular free water loss, proportionate to cell dehydration, a decreased protein and enzyme efficiency of function will be established. This decrease in protein function will include receptors incorporated in all the membranes within the cell as well as the plasma membrane. It seems, since all transport systems in the cell, whether carrier borne or microstream flow directed, will down-regulate as a direct resultant effect of increasing viscosity of the cytosol, or water perfusion decrease within the bilayer membrane of the cell. This hydrodynamic down-regulation of cell physiology could be reasoned to be responsible for irreversible cell damage or cancer transformation. The initiating mechanism implicated seems to be continued histamine induced sequence of calcium-calmodulin protein kinase C- protein kinase M activation within the cell. Diabetes seems to exemplify the process (1).

Key Words: histamine, thirst, free water, renin-angiotensin, vasopressin, receptor protein down-regulation, cell membrane, cation pump, hydrolysis, diabetes, insulin, interleukin-1, interleukin-6, tumor necrosis factor, CRF, DNA fragmentation, gluconeogenesis, proteolysis, neoplasia.

During the past few years the mechanisms of body water regulation have attracted attention (1). It has become a recognized fact that with increase in age the thirst mechanism becomes less acute. Since water intake depends on this sensation, an incipient and gradually establishing chronic dehydration will become a symptom producing physiological state of the human body (1,2,3,4). This state of chronic dehydration will drastically affect the cell water content. The ratio of extracellular to intracellular water content of the body, between the ages of twenty to seventy years of age, has been shown by Bruce and associates (5) to change from an approximate figure of 0.8 to 1.1. This drastic change can only mean free water

deficiency within the cell. Since water balance regulates intracellular fluid volume and sodium balance regulates the extracellular fluid volume (6), it would be reasonable to assume that the fine regulatory role of thirst sensation becomes blunted from the age of twenty onward, or even earlier. Clinically (68) and scientifically (1,52) it has been demonstrated that there are other physiological signals associated with local or general intracellular free water deficiency, among them the dyspeptic pain. Dehydration as a signal producing phenomenon should become part of any evaluation and differential diagnosis of disease etiology in the human body (1, 52, 68).

According to Katchalski-Katzir (7), proteins and polypeptides demonstrate a greater conformational freedom in solutions of low viscosity. As the viscosity of the medium increases, conformational freedom of proteins and polypeptides, essential to their efficiency of function, will also decrease. In media of higher viscosity, the peptide chains become "completely" frozen in their conformation. The same behavioral phenomenon is seen in linear peptides. The characteristics of conformational freedom in solutions of low viscosity can involve the amino acid side chains, segments of a particular protein or even its domain flexibility (7). Assuming that all physiological functions of the body are regulated by the tightly coupled protein and enzyme actions and their feedback mechanisms (8), for those enzyme systems that depend on diffusion for their action (9,10,11), or the transfer of their end-products, the efficiency of function of these proteins and peptides are inversely proportionate to the viscosity of their cytosolic or membrane environment. Then the decrease in cell free water may be assumed to be directly, as well as indirectly responsible for the decrease or even loss of some protein and enzyme functions.

Free Water

Free-water has many important primary functions; it generates high heat of activation that is necessary for cell differentiation, maturation and development (65,66,67). In the bone marrow cell line, a correlation between the different stages of cell maturity, nuclear development, the permeability of water through the cell membrane and the cellular content of osmotically active water has been shown (67,66). When water is said to produce high heats of activation (65,66,67), this thermodynamic effect of water will also increase the rotational properties of aromatic amino acids, tryptophan in particular (1,7). The increased rate of conformational change in the amino acid component of macromolecules will also translate to greater flexibility of the chain segment or domain flexibility essential for random coupling between the hormones and neurotransmitters and their receptors in the cell membranes (1,7). Thus the heat of activation produced by osmotically active "free water" will increase the efficiency of the cell membrane receptor proteins. Dehydration will down-regulate the rotational properties of the receptor proteins and render them less efficient.

Assuming that all actions of the body are energy dependent and the energy exchange is derived from hydrolysis of ATP, then this very activity is directly and indirectly water dependent. For hydrolysis to take place, free water is required; with increase in body activity more free water will be required for generation and release of energy. With stress and subsequent hormonal over-ride for gluconeogenesis and hydrolysis of fat and proteins (21), free water will be needed. For the phenomenon of neurotransmission and cation exchanges at the cell membranes, free water will be required. ATP driven cation pumps, H^+, Na^+ and Ca^{2+}, apart from their cation exchange, also synthesize ATP as a result of increase in chemical potential with increasing hydration of small cations and polyphosphate anions in the highly structured interfacial aqueous phase of the two phosphorylated intermediates within the channel, where free energy of hydration is assumed to be involved (12,51,65,66,67), when water is shed at

the hydrophilic selectivity filter site in the narrow section of the tunnel (14,22). Dehydration will negatively affect this physiological function, albeit, even at a local level.

A silent assumption, in recognizing hydrolysis as a source of energy formation or transformation, would logically be the acceptance of an involvement of water in tightly coupled enzyme directed dissociation of water itself when the hydroxyl ion becomes a factor in the cell buffering system for its pH regulation. The hydrated hydrogen ion, the hydronium ion is a transient by-product of hydrolysis. The free energy of H^+ is used for energy consuming transport functions, or storage in ATP synthesis, or even intracellular calcium bondage, subsequent to calcium transport. Thus, availability of free water molecule is a factor of great importance for pH regulation, as well as its being a source for energy liberation in the process of cell maturation and its functions - a series of continuously water-dependent regulatory mechanisms.

Ca^{2+}-ATPase serves to transport calcium with the stoichiometric coupling ratio of two mol of Ca2+ for each mol of ATP (13). An assumption to this action is that, when calcium from the intracellular stores is released, then, for every two mol of Ca^{2+} released, energy for formation of one mol of ATP is also liberated. These are some indications that the cation transport system, that regulates the steady-state functions of the cell, is also involved in energy generation and its storage - or its liberation through some mechanisms that involve the energy of hydration of ions by the water molecule during cation exchange. This thought gathers support from study of the proposed characteristics of single ion channels. It seems that one criterion of such channels is their specificity to the particular ion, in such a way that the ion will have to shed some of its energy of hydration, when it passes through the short and narrow selectivity filter within the tunnel (14,22). If the strongly hydrolytic metabolism of water in the cells of the body involves the formation of excess hydrogen ion which then forms hydronium ion, H_3O^+ - and it is agreed that hydrogen ion channels do exist - then the hydrogen ion must also liberate its energy of hydration during its passage through the channel, in the same way proposed for other single ion channels.

The H^+ pump proteins must be designed in such a way that they recognize the hydronium ion, when free or "nascent" H^+ will exist for an instant to join another water molecule and form the hydronium ion at the other side of the pump protein, when the "selectivity filter" shaves the water molecule off the H^+. Under such possible circumstances, free water content of the cell may have to be recognized as a ready source of energy for the intracellular energy dependent coupled reactions and pH regulation. The OH^- produced by the hydrolytic processes becomes adsorbed to the protein buffers in the cytosol. As a model for this explanation, one could assume that the parietal cells of the stomach have developed this process of hydrogen ion formation and its secretion to a fine art, and is involved in thermodynamic energy exchanges emanating from hydrolysis of water for the act of protein digestion. Thus, dehydration and cellular free-water loss would mean a proportionate decrease in some cell functions, after the stored energy in other forms is used up at a rate greater than it is supplied.

Since histamine is said to induce hydrogen ion formation and its secretion into the stomach, at the same time that it induces calcium release within the cell, it may be justified to consider the action of histamine as a component of energy management, including amino acid mobilization for cell functions.

Biological membranes have similar characteristics, they are bilayer in form (15,18,19) and, depending on the amount of their cholesterol content (1,57), are semipermeable to water and almost impermeable to ions. The diffusion coefficient for water permeation through the mem-

brane is approximately $1x10^{-2}$ to $2x10^{-5}$ centimeters per second (15, 16), whereas for sodium and potassium, it is 10^{-12} (16). Rand and Parsegian have shown that water separates the bilayer structure of lipids to a fixed distance of 20-30 Å in case of electrically neutral bilayer or to a large separation of up to 100 Å in case of charged bilayers. A lateral diffusion pressure is also developed when water gets into the bilayer (17).

On the basis of the above information and the consensus of opinion that biological membranes have the same properties as reconstituted lipid bilayer membranes (15,18,19), it has been proposed that with increase in water permeation through the cell membrane, the "tuning-fork-like" hydrocarbon projections within the cell membrane act as mixers, or stirrers, by becoming responsible for the lateral diffusion pressure, at the same time as the two sides of the membrane are separated by the diffusing water (1). When water in the membrane is deficient, the same "tuning-fork-like" projections act as barrier rods to the process of lateral diffusion of enzymes that function within the bilayer.

This lateral diffusion system in the bilayer, a free water flow-dependent system, seems to be another mechanism for the "proportionate" integration of cell into the main stream of body physiology. Silver (18) explains the separation of the hydrocarbon chains as a result of an approximately 35 degree tilt, with an schematic diagram indicating the creation of a free space between the two layers of the membrane. This separation of the membrane bilayer by water is a very convenient development for unhindered lateral diffusion of the enzymes functioning within the bilayer space. Silver (18) and Edidin (20) confirm the existence of the phenomenon of lateral diffusion in the bilayer structure of the cell membrane, as well as the mitochondrial membranes, demonstrated by Sowers, et al.(23).

It is generally agreed that the adenylate cyclase activation by the cations, H^+, Na^+ and Ca^{2+}, as well as that stimulated by beta-adrenergic receptors is a diffusion controlled process, inversely proportional to the microviscosity of the membrane (9,10,11). It seems that the adenylate cyclase in the membranes exists in single pools, to be stimulated by the frequency of collisions from any number of agonists dependent on its messenger system activation (20). Hydration of the cell membrane, to the extent that there is a build up of the lateral diffusion pressure, as well as separation of the hydrocarbon chains, would increase the efficiency of the cation exchange, the beta-adrenergic neurotransmission, as well as other receptors, whose functions depend on diffusion rate in the bilayer membrane - many of the receptors involved in the feedback mechanism of neurotransmission and hormonal systems. Conversely, dehydration of the cell membrane would proportionately decrease, or down-regulate the cation exchange or hormone receptor activity.

Microstream Axonal Transport

This microviscosity alteration, as a result of water diffusion decrease through the cell membrane, could become serious where neurotransmission mechanisms is concerned. If neurons regulate function by means of axonal transport, delivering formed products for incorporation in the end plates or synaptic junctions (24), the two way axonal transport must also be microviscosity sensitive. According to Gross and Weiss (25,28,29), the process of axonal transport is also a hydrodynamic phenomenon (25). There seems to exist in the axons, low viscosity microstream flow regions surrounding the microtubules. This low viscosity domains in the axons possessing flow properties could be directly responsible for the transport of substances in solution, such as amino acids, electrolytes and proteins, or even formed products, such as vesicles. Indirectly, the function of microtubule-dependent carrier proteins, such as kinesin (26,27), an ATPase-dependent activity (28,29), coupled to

calcium calmodulin activation (30), would be facilitated in the low viscosity microstream flow region around the microtubules. It appears that microtubules act as "drainage pipes" in the cytosolic medium and draw water and create low viscosity regions around themselves. At the same time, they act as "guide-lines" for the transporter proteins and their floating cargo. Accordingly, a direct volume-dependent result of free water deficiency in the body would be a proportionate down-regulation of protein activity. This decrease in activity or down-regulation will also include the neurotransmission systems, receptors involved in the pathway of neurotransmission, neuroendocrine systems and finally, the ionic pumps, the backbone of cell functions (1).

So far, the two neurotransmission systems recognized for their body water intake regulation are the serotonergic and the histaminergic neuronal systems. Both these systems also activate the renin-angiotensin system for water intake of the body. Because of its apparent exposed characteristics, it has been proposed that histamine, apart for its water intake regulation, also acts as an emergency substitute to water in cation pump activation, and release of energy for this action (review of references included in ref. No.1). It is now apparent that histamine, through H_1 receptor activation, produces a cytosolic free calcium release and oscillation, which in turn produces a cyclical potassium channel activation (31). Since K^+ pumps initiate a regulatory ion exchange, the role of histamine in initiating the calcium-dependent potassium channel must be considered to be a cell volume regulatory process, as well as its other functions. In support of this statement, it is important to note that histamine has been demonstrated to exert a strong stimulatory effect on vasopressin release in animals and man (32). Vasopressin is also secreted in stress (21).

According to Finkelstein's review(15) of the research about water permeation through epithelial membranes, it seems that an antidiuretic hormone-receptor coupling is considered to have the capability of producing a wide base channel through the external aspect of the bilayer membrane. A cluster of perforations are created at the inner side of the membrane at its wide base. The final transformation of the receptors seems to resemble a "shower head" with perforation at the inner leaf of the bilayer membrane. The perforations are considered to be approximately 2 Å units, enough only for a single-file passage of one water molecule at a time (15). The brain is very sensitive to its volume change, and a 1% change in osmolality will stimulate the secretion of vasopressin. The direct water intake through the cell membrane - diffusion and shower-head filtration must be an important function of osmoregulatory systems, of which histaminergic neuronal systems must be considered to be an initiating link. In the same way renin-angiotensin system is now considered to be involved (33), and even stimulated by the action of histamine (46,52), histamine itself seems to be the primary regulator of cell water content in the cells of some organs during the time there is dehydration.

In view of the existence of a low viscosity microstream flow of the axonal transport system, proposed by Gross and Weiss (25,28,29), and the fact that some of the transported materials will take days or even weeks to be delivered to their ultimate sites of action to prevent a delicate water-dependent transport system of the brain function to become down-regulated, satisfying the natural drive of the histaminergic neuronal system and its generated signal systems for water intake, instead of blocking its actions by enumerable variety of antihistamines medications used in the different disciplines of clinical medicine, seems imperative (1,52). Furthermore, alcoholic beverages further hamper vasopressin regulation of cell volume by their inhibitory action on vasopressin secretion. The result of chronic dehydration coupled to the wrong choice of fluid intake (alcohol, coffee, tea and caffeine-containing beverages) may be the main contributory factors in production of some

neurological disorders, such as multiple sclerosis.

Calcium-Dependent Proteolysis

If constant reliance on a failing thirst mechanism (2,3,4) produces a chronic dehydration and increased cytosolic microviscosity, inducive to continuous histaminergic and renin-angiotensin systems' drive for water intake, then the increased cytosolic calcium turnover for cation regulation must be a logical expected outcome, in the way shown by Sauve and associates (31). Mellgren characterizes a class of non-specific calcium-dependent proteases (CDP) that are active at the level of the cell membrane (34); they are non-lysosomal proteolytic enzymes that are activated by free calcium. They consist of two types: one is activated in the presence of micromolar calcium levels and the other in the presence of millimolar calcium levels in the cytosol.

It seems that calcium initiates proteolysis of the enzyme protein into a large and a small proenzyme subunit, with increased affinity for the calcium ion; as a result the proteases undergo autoactivation. Although there are also calcium-dependent protease inhibitors (CDPI), their action is neutralized by CDP at higher activated rates, indicating that these inhibitors are transient membrane buffers. Certain cells have more CDPI than proteases (34). The continuous action of these proteases in the brain, where a greater type one variety of CDP capable of proteolysis at lower calcium concentration is present, is being considered to be the cause of damage to the membrane-cytoskeleton attachment protein fodrin in some conditions (34). Protein Kinase C (PKC), responsible for phosphorylation of some intracellular proteins, (activated through Ca^{2+}-calmodulin simulation and ATP hydrolysis at a calcium concentration of about 10^{-8} to 10^{-5} M, an activity inhibited at Ca^{2+} concentration of about 10^{-4} M -- 35,13), is proteolysed to protein kinase M (PKM). PKM is a kinase that is independent of calcium activation or inhibition; this protein kinase is being suspect in cancer transformation of the cells (34). Thus, if proteolysis is an essential component for maintenances of the amino acid pool balance and gluconeogenesis, again, histamine is directly involved in the energy conversion processes and mechanisms.

Increased calcium availability to the tissue can promote increased granulation tissue formation and faster repair rate (39), indicating that higher calcium turnover in the tissue can promote angiogenesis, a phenomenon seen in neoplastic tissue and AIDS associated Kaposi sarcoma. (47,48,...).

Another water regulatory process, operational at the cell membrane level, is the mechanism of vesicle formation, when the extra cellular environment is sampled, taking water in at the same time. This process seems to be involved in cell water and volume regulation. In a similar way, water can be extruded from the cell by the formation and extrusion of water vesicles. The process of vesicle formation and release of content seems to be under Cl^- regulation (36).

The cell membrane seems to undergo constant internalization and reconstitution, carrying with it the surface proteins and receptors. Ligand activated receptors are stimuli to vesicle formation and internalization of the membrane, at the same time as the receptor protein and its ligand attached to the membrane - "receptor mediated endocytosis." The pinocytic vesicles fuse to form endosomes, where, through decrease in pH brought about by H^+ build up in the endosome, the ligand is separated from the receptor. The receptor protein is released to be recycled back to the membrane for its reincorporation. If the hydrogen ion concentration is not sufficient to separate the ligand from the receptor, both the receptor and the ligand is delivered to be degraded by lysosomal enzymes. Dependent upon the rate of receptor protein synthesis, recycling efficiency and degradation of the receptor-ligand

complex, a down-regulatory process is possible (37), if the proton pumps and ATPase activity do not keep up with the cell requirements. This receptor-mediated endocytosis mechanism regulates many of the external stimulatory and supply processes of the cell. Catecholaminergic receptors are also recycled after internalization of the receptor. By a process of phosphorylation-dephosphorylation reaction, the receptor is recycled and reincorporated in the membrane. This receptor exhibits two types of desensitization of the adenylate cyclase, "homologous" and "heterologous". Homologous desensitization is rapid and lasts during the period of internalization of the receptor, when the adenylate cyclase is responsive to other hormonal agonists. Heterologous desensitization of the adenylate cyclase affects more than just the one agonist, it becomes desensitized to other hormonal agonists; this type of desensitization is in part mediated by cAMP; it seems phosphorylation events are involved in heterologous desensitization (38).

Taking the above membrane receptor recycling characteristics into consideration, it would seem justifiable to assume, if the recycling process of receptors is an important physiological function, then a continuous and unabated histaminergic drive for water intake would also deregulate the cell function by increased release of calcium from the cytosolic stores, in an attempt to regulate cation pumps of the cell. This increased calcium turn over, not only would continue to activate the membrane proteases but, by increased activity of protein kinase C and eventually protein kinase M would also deregulate the catecholaminergic, as well as some other receptors on the cell membrane. This process could lead to an autonomous cell function, disconnected from the other regulatory mechanisms that would stimulate as well as inhibit the cell in a normal integrated manner, the very autonomy seen in neoplastic tissues of the body.

In this way, continuation of what should be transient regulatory actions of histamine, slowly reversible, or completely irreversible alterations to cell function and structure will come about, when regulation of water metabolism is continually disrupted or its signals misunderstood. Because histamine, the water intake regulator and its emergency substitute, has been shown to be a growth promoting factor (40,41,42,43,44), whilst abundant water supply decreases the mast cell histamine (45). Thus, the continued action of histamine during chronic dehydration may ultimately be the cause of an autonomous cell function, independent of most other regulatory mechanisms established at the time of integration of the cell-type in the total systems of the human body. Depending on the hierarchical importance of the cell, the action of histamine would seem to fall within a process of saving that cell-type from destruction as a result of long lasting dehydration. Since the excess action of histamine at a local level can only be inhibited by the feedback with its H_3 receptor, or its production in the mast cells or basophils decreased by the direct effect of water, the action of histamine should be presumed to remain in force throughout the period of dehydration until the cell changes become irreversible.

Diabetes as a model of dehydration-induced receptor down-regulation-initiated pathological disorder.

Within the "free water-dependent systems' approach" to disease emergence in diabetes, before establishment of the associated irreversible pathology, it seems the emergency and naturally predetermined physiological steps are designed in such a way to mobilize water from its storage in the intracellular compartment. The regulatory design around the flow of water initially delivers water and glucose and then the necessary amino acids that are carried by water to the brain tissue for the maintenance of its total function (1). Water,

in its 3 nanometer thick "adhesive sheet" form, is said to be directly and physically involved in the maintenance of the total integrity of the cytoskeletal structure (50, 51,52). The brain tissue is said to be 85% water (50) and extremely sensitive to the slightest fluctuation in its water content. Water at some point of its passage through the cell membrane seems to form a defined "adhesive sheet" conformation that is said to hold the cytoskeletal cell structure together (50). The brain tissue has no insulin barrier to its glucose metabolism (1). The main by-product of glucose metabolism of the brain is water itself.

One of the organs that must also receive an adequate supply of water is the pancreas. This organ has to regularly secrete copious amounts of watery bicarbonate solution into the duodenum to neutralize the acid that is emptied into the intestine from the stomach. Under a paradigm shift that establishes water regulation of the body as a primary objective of the regulatory systems (1,52), the action of histamine and PGE production become the operative drives in the water distribution system(s) in chronic dehydration (52). Within such an integration of function, the production of insulin will need to be inhibited. Otherwise, through the effect of insulin on transport of K^+across the cell membrane and its flow into the cell, the flow of "coupled water" will also establish a "counter-productive" step to the emergency need to redistribute some of the "free water" held in the intracellular compartment. In this light, the *in vivo* exposed inhibitory action of PGE on insulin production by Robertson and Chen(53) and Robertson and Associates (54,55), involving PGE stimulation of Ni components of the b-cells' adenylate cyclase can be interpreted as a confirmation of the above logic. Furthermore, PGE can be manufactured in the capillaries (52) and distributed in the pancreatic tissue because of the possession of a special "intra-islet interstitial fluid flow system" in the pancreatic islets of Langerhans, demonstrated by Weir GC and Bonner-Weir S (56).

Other processes in this systematic series of actions, "to prevent water from getting into the cells that have insulin receptors," involve xanthurenic acid "masking" of the insulin hormone itself, rendering it biologically less effective (1). Other insulin receptor down-regulation procedures involve the action of tyrosine aminotransferase on the exposed 5 tyrosine molecules needed for the autophosphorylation of the insulin receptor in the liver (52). The continued action of PGE in the liberation of Ca^{2+} (52) will activate the "non-specific Ca^{2+}-dependent proteases" at the cell membrane (34) and may ultimately become responsible for production of the many different pathological "steady-state" pictures seen in diabetes, all of them the result of the predetermined brain crisis response to a threshold determined persistent "free water shortage" of the body.

Chronic dehydration can alter the efficiency of tryptophan metabolism of the body (1,51,57). Dehydration can alter the permeability of the cell membrane by alteration of its cholesterol content (1,57) and establish a cholesterol adaptation process associated with dehydration, but attributed to diabetes in patient evaluation. The alteration of cell membrane "structural homeostasis" and the possible disturbance of the amino acid metabolism with the establishment of a different composition of the "amino acid" pool can bring about many structural changes in the manufacturing systems within the cells.

While diabetes begins as an insulin-independent (insulin secretion inhibited) form, at some time during the course of the disease, it often transforms to an insulin-dependent variety (B cell destruction), frequently seen in the younger diabetics. The steps in this direction seem to become established as a side effect of stress that is associated with dehydration. Chronic dehydration establishes a stress physiology in the human body, in the same way "stress" itself causes cellular free-water depletion(1,51,52). An associated secretion of cortisone release factor (CRF) in stress-in-

duced physiological response of the body is already recognized (1,52). It seems that cortisone release factor stimulates the secretion of interleukin-1 (IL-1 --59), when IL-1 could also promotes the secretion of CRF (60).

This mutually complementing secretory pattern of CRF and IL-1, which seems to become dose dependent, establishes a series of physiological events that are basically designed to remobilize some of the stored amino acid composition of the large muscle mass for effective and adequate gluconeogenic events in the liver. At this stage of events, the processes involved seem to operate as though there is a priority crisis management. IL-1 is a potent inducer of IL-6 (61). Interleukin-6 seems to be able to act as its own inducer in an autocrine/paracrine manner in some cell lines *(in vitro)*. Tumor necrosis factor (TNF) is also capable of stimulating IL-1 and IL-6 production (62). Both IL-1 and IL-6 are able to act in an inhibitory capacity on B cells of pancreas (61). However, IL-6 seems to also have a different type of action on B cells. In cell cultures, it has been shown that a prolonged action of IL-6 on B cells seems to bring about a gross depletion of the DNA content of the nucleus in the cell and eventual inability to produce and secrete insulin by the IL-6 treated B cells (61).

The design of the human metabolism seems to show an initially silent inhibition of insulin secretion as a component of "stress," which includes chronic dehydration. The purpose seems to point at mobilization of water by preventing it from being transported into the cells. Insulin has the ability to shift K^+ from the extracellular compartment into the intracellular compartment, and by this action, the transport of K^+-coupled water across the cell membrane that possess insulin receptors. At the same time, insulin is an anabolic hormone, it also promotes amino acid transport across the cell membrane. It now seems that one of the ways the body taps on its manufactured stores of protein to replenish the amino acid pool available to the liver is by proteolysis of the large muscle mass. The released amino acids are delivered to the liver where the manufacture of new proteins and peptide could take place. The agents that seem to bring about this proteolysis are tumor necrosis factor and interleukin-1 (63,64). Thus, the DNA depleting action of IL-6 on B cells of the pancreas will develop a logic in the design of water and amino acid mobilization systems within the body. Although the experiments that exposed the proteolytic property of TNF (DNA/RNA fragmentation) and IL-1 concentrated around septic conditions in the body, in non-septic traumas or other types of stresses, the same physiological events will result if prolonged cortisone release factor release becomes strongly established (1, 52).

Water Metabolism and Future Scientific Research of the Human Body

Fluctuations in the composition of the "amino acid" pool can bring about many structural changes in the manufacturing systems within the cells. These structural changes are reflected in the genetic markers attributed to the different disease processes. In the "solute" dominated sphere of thought, the genetic markers are assumed to be the indicators for disease emergence. In the "solvent" considered sphere of thought, that begins to recognize a gradually establishing chronic dehydration in the human body, the disease processes and their accepted genetic markers reflect a "primary" solvent metabolism disturbance as a ***common factor*** to a myriad of processes involved in the establishment of the new "steady state" physiology of the human body that finally includes DNA/RNA structural alterations.

One such indicator is the very process of histamine and PGE production in the physiological regulation of fluid distribution in the body. This is an adaptation process to "drought management" by the body. The same histamine regulated

mechanisms are involved in the natural growth of tissues, plant or animal (40-44,52). If the physiological state of the body places a particular demand on a set of functions that involve the natural and regional increased production and activity of histamine as a "water regulator," the underlying and determining factors for "drought management" (known or unknown) will also be made available to the tissues in the region. Under such circumstances, it would not be out of the question that the regional tissues, because of their APUD characteristics, would begin to make histamine or preserve the produced histamine. Indicators for this prediction are given by Nolte and Associates (52 - reference 140), they describe the ability to produce histamine by some tumor cells; and Kierska and Associates demonstrate a disturbance of histamine breakdown mechanisms in some tumor bearing animals (52 - reference 141).

The immune system's role in prevention of tumor production, the other end of the broken loop to DNA/RNA damage, seems to become proportionately inhibited with the increased activity of histamine, when it becomes engaged in the regulation of water intake and its regional or total distribution in a dehydrated body (52). PGE_2 inhibits the immune system and is beneficial to tumor cells (58). It seems that the design of the body, as an integrated multi-system system, is such that automatic consequential steps to advanced dehydration - an extremely stressful state of physiology for the brain - brings about a crisis management that will include a destructive phase-out initially of some cells and eventually some anatomical parts of the body other than the brain. The progressive pathology associated with diabetes is already well recognized.

At the same time, the pancreas - whose main action seems also to be "water and bicarbonate" secretion into the intestine, as well as its insulin secretion that indirectly facilitates the shift of water into the insulin "sensitive" cells - is now "functionally" required to concentrate only on its direct "water secretory" actions. The more there is dehydration, the more the "water and bicarbonate" secretory gland cells will need to be increased to cope with the state of dehydration and the burden of the "solid food" metabolism. It may not be too speculative to think the local "paracrine" effectors, in the process of transforming the pancreas into an entirely "water and bicarbonate" secretory organ, will, effectively and in the process, cause the *transformation* of the cell structure in this organ.

There seems to be a strong "pack" relationship between IL-1, IL-6 and TNF. When one becomes active, the others will also become activated. Another highly active agent in this group is known as transforming growth factor (TGF). The action of TGF is designed to brings about the process of growth and remodelling of tissue. In reality, these cytokines are involved in the repair system after trauma. One must assume that chronic dehydration will also cause a form of trauma to the dehydrated tissues. Thus, cancer of the pancreas, often seen as a final complication of diabetes, may find its early root in the initial dehydration that eventually progressed to diabetes.

DISCUSSION

While mitogens and phorbal esters are being investigated for activation of protein kinase C and M in cancer cell transformation, it may serve the patients better, if it is recognized that continued cell free water deficiency, to the point of chronic dehydration, would bring about the same cytosolic molecular and physiological states inducive to cell transformation, blamed on mitogens and phorbal esters or even viruses. Up to now medical research scientists have paid attention only to the highly reactive solutes in the body, without much regard to the reactive and hydrolytic functions of the solvent. While the word hydrolysis is frequently used, yet, by some in the forefront of research, the terminology, "metabolism of water"

is considered unscientific.

In the same way iron filings recognize and obey the force generations of a magnet and align themselves accordingly. It seems that proteins and enzymes of the body (only about 15-20 percent of the estimated total of 5 to 10 thousand cellular enzymes have been identified so far -- 47) recognize and align their functions to water's regulatory role in the body physiology. Decreased water intake does not mean that these proteins and enzymes have become water-independent; it would mean that some of these functions become decreased and eventually lost. Dependent on the different transient steady-state stages of physiology, different signal systems are given by the body. Not recognizing and accepting the fact that water regulation for the body is a primary concern and therefore the concerned systems' drive to be of primary importance, we consider the signals associated with these very same drives as indications of different disease states, with the unfortunate consequences associated with their inaccurate manipulation.

We need to become cognizant of a simple fact, it is not the sole function of ions to regulate water intake of the body, that protein regulators and sensors are also involved, the very proteins that demonstrate increasing conformational change and efficiency with increasing hydration. We need to disregard the teaching that dry mouth is the only indication of thirst; it has been shown that this is not the case (2,3,4). Instead, we should recognize that the histaminergic, the serotonergic and the renin-angiotensin systems are involved in body water intake and regulation. It would be a disservice to the patient to block their actions, without testing the effect of increased water intake to satisfy the natural physiological drives of these systems (1). The human body has been shown to have a series of direct and indirect thirst signals, of which dyspeptic pain is one (1,52,68). When this and other signals are not recognized and the associated dehydration markings at different tissue sites treated as markers of different diseases, the pathology that will continue to establish would be so vast that receptor down-regulation is only a small factor in the constantly changing steady-state physiology of a dehydrated body, within the span of time it has established. A well planned and eloquent exposure of the consequences of chronic dehydration in the body, which could establish gradually and from an early period of life, will make it possible to establish a preventive approach to health-care, where it could be most effective - in the home and family environment.

CONCLUSION

If protein kinases are being implicated in cancer transformation, then it should be recognized that chronic dehydration that seems to have been repeatedly demonstrated in the elderly and suspected to establish from a much earlier age, can become responsible for their increased activity. At the same time, receptor down-regulation can be the direct and the indirect resultant effect of cell free water deficiency, the consequence of decreased and irregular water intake with increase in age. This disparity of regulation may possibly be implicated in the development of a physiological state, either by itself, or by the accentuating effect of other factors, to push the cell in the direction of neoplastic transformation. Because histamine, the regulator of body water intake and its emergency substitute, has been shown to be a growth promoting factor (40, 41, 42, 43, 44), whilst abundant water supply decreases the mast cell histamine formation (45).

Acknowledgment. I thank the *Almighty* for *His* light and fine detailed guidance that has made this presentation in *His* name possible.

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