REVIEW ARTICLE

Lactic Acidosis and Other Mitochondrial Disorders

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The ability of mitochondria to oxidize substrates and generate energy is integral to normal homeostasis and to the ability of cells to survive in the face of impending energy failure. Lactic acidosis is a common and readily apparent biochemical marker for mitochondrial dysfunction. However, lactic acidosis represents only the most obvious example in which acquired or congenital abnormalities of mitochondrial oxidative phosphorylating capacity contribute to the pathobiology and phenotypic expression of a broad spectrum of clinical disorders. Consequently, interventions that improve mitochondrial function or prevent mitochondrial energy failure may have widespread therapeutic implications.

A PATIENT with lactic acidosis commonly elicits two immediate reactions from clinicians: first, death is likely, if not imminent, and second, little can be done to alter the prognosis. This rather jaundiced view reflects the knowledge that persistent hyperlactatemia is usually due to acute or chronic multiorgan system failure[1,2] or to congenital DNA mutations that severely compromise aerobic cellular metabolism and energetics.[3] In either case, reversing the acid-base disturbance is difficult and does not necessarily improve survival.[4] Overlooked, however, is that lactic acidosis is frequently linked to a fundamental disturbance of mitochondrial function, for which hyperlactatemia is a marker. Moreover, similar or identical mitochondrial pathology contributes both to the physiology of aging and to several diseases of metabolic integration in which the efficient conversion of substrate fuel into energy is skewed, in part, by acquired or hereditary abnormalities of mitochondrial oxidative metabolism. Such a concept is predicated on the hypothesis that defective mitochondrial energy metabolism cuts across disease strata to provide a common pathogenetic denominator, albeit variably expressed as diverse clinical syndromes. In addition, the usual mendelian genetic paradigm, in which the presence of a single gene defect yields a precisely predictable inheritance pattern, is inconsistent with the pathology of aging and many degenerative diseases of mid- to late-life. This article reviews relevant aspects of lactate metabolism and mitochondrial biology, links the pathobiology of certain neurologic, myocardial, and endocrinologic disorders to mitochondrial function, and offers a common therapeutic strategy to mitigate the clinical expression and consequences of these mitochondrial diseases.

MITOCHONDRIAL BIOLOGY

An evolving symbiosis of infectious prokaryotes and their eukaryotic hosts may ultimately have led to the presence of mitochondria within eukaryotic cells. The most obvious physiological consequences of this union allow animals to oxidize substrates to produce energy and to sustain amphibolic pathways, such as the tricarboxylic acid cycle, essential for survival (Fig 1). A striking metabolic feature of eukaryotic cells is the coordinate regulation of multiple cytoplasmic and mitochondrial reactions required for the efficient conversion of substrate fuel into energy. This is illustrated by the catabolism of glucose and the differential in adenosine triphosphate (ATP) (Table 1) yielded when it is metabolized anaerobically in the cytoplasm to lactate, versus aerobically in mitochondria to carbon dioxide and water (Table 2). Under aerobic conditions, the oxidation of glucose (and lactate) is controlled by the activity of the pyruvate dehydrogenase complex (PDC). This multienzyme system is located in the mitochondrial inner membrane and decarboxylates pyruvate to acetyl coenzyme A (CoA), generating reducing equivalents in the form of reduced nicotine adenine dinucleotide (NADH). Rapid posttranslational regulation of PDC activity occurs by substrate activation, end-product inhibition, and reversible phosphorylation, in which the phosphorylated enzyme is inactive (Fig 2). Free calcium is a positive regulator of the activities of mitochondrial dehydrogenases such as PDC and α-ketoglutarate dehydrogenase, and is thus an important determinant of mitochondrial substrate utilization and energetics.

Reducing equivalents, in the form of NADH, are produced in the cytoplasm by the glyceraldehyde-3-phosphate dehydrogenase reaction, and in the mitochondria, in the form of NADH and flavin adenine dinucleotide ($FADH_2$), by the PDC reaction and the tricarboxylic acid cycle. Electrons donated by these products enter the respiratory chain at complex I (NADH dehydrogenase) or complex II (succinate dehydrogenase) and initiate a series of iron- or copper-mediated oxidation-reduction reactions that culminate in the reduction of molecular oxygen to water (Fig 1). Each complex consists of multiple protein subunits. Complexes I, III (ubiquinone c reductase), and IV (cytochrome oxidase) are believed to act as proton pumps that create an electrochemical gradient across the mitochondrial inner membrane that drives a membrane-associated ATP synthase (complex V) to produce ATP.[5]

Over 90% of the oxygen used by mitochondria is reduced to water by a series of reactions that produces potentially highly reactive oxygen intermediates. As already noted, one of these reactions is catalyzed by complex IV, which receives electrons from ferrocytochrome *c* and transfers them to cytochrome a_3-Cu, the site of oxygen reduction.[5] An intact, functional oxidative phosphorylation system minimizes "leakage" of electrons from the respiratory chain,[6] while reduced glutathione

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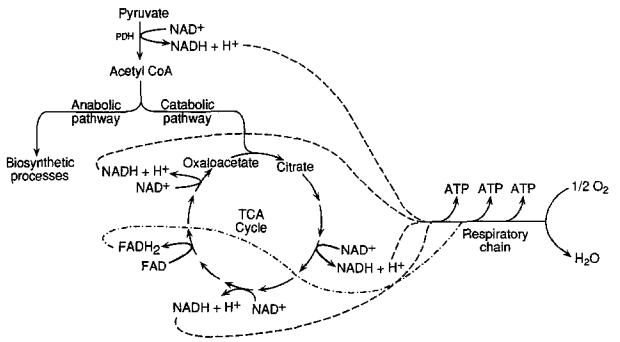

Fig 1. Mitochondrial oxidation of pyruvate and ATP production. Reducing equivalents in the form of NADH and $FADH_2$ are generated by the reactions catalyzed by the pyruvate dehydrogenase (PDH) complex and by various dehydrogenases of the tricarboxylic acid (TCA) cycle. Electrons from 1 mol NADH enter the respiratory chain at complex I (NADH dehydrogenase) to yield 3 mol ATP. Electrons from 1 mol $FADH_2$ enter the chain at complex II (succinate dehydrogenase) to generate 2 mol ATP. Complex IV (cytochrome oxidase) facilitates the reduction of molecular oxygen to water. (Reproduced with permission[1]).

and manganese superoxide dismutase provide intramitochondrial free radical quenching mechanisms.[7,8] A fundamental but rather ironic aspect of mitochondrial development is that these organelles probably existed initially in a state of extreme hypoxia and active free radical–generating reactions, both processes essential for biogenesis and evolution.

The mitochondrial genome is an example of extreme genetic parsimony. All the structural proteins of both the mitochondrial membrane and the respiratory chain complexes, all the protein subunits of PDC, all the enzymes of the tricarboxylic acid cycle, and all the proteins required for mitochondrial replication (and thus for long-term mitochondrial survival) are encoded by nuclear DNA. However, in addition to its limited information content, mitochondrial DNA is distinct from nuclear DNA in many other important respects.[9-11] The mitochondrial genome is a closed circular molecule that contains 16,569 base pairs situated on two strands that code for 12S and 15S rRNAs, 22 tRNAs, and only 13 subunits of the respiratory chain enzymes, that is, the functional (rather than structural) proteins comprising the various macromolecular complexes of the oxidative phosphorylation cascade (Fig. 3). The length of the mitochondrial genome is about 0.001% that of mammalian nuclear DNA.[12] Not only does the mitochondrial genome code for very few mitochondrial proteins, it also lacks introns and uses two predominant promoters located near the triple-stranded displacement (D) loop region.

Table 1. Glossary of Terms

ATP: adenosine triphosphate
PDC: pyruvate dehydrogenase complex
NAD(H): oxidized (reduced) nicotine adenine dinucleotide
$FAD(H_2)$: oxidized (reduced) flavine adenine dinucleotide
Complex I: NADH dehydrogenase
Complex II: succinate dehydrogenase
Complex III: ubiquinone *c* reductase
Complex IV: cytochrome oxidase
Complex V: ATP synthase
SOD: superoxide dismutase
H_2O_2: hydrogen peroxide
mtDNA: mitochondrial DNA
O_2: superoxide anion
H_0: hydroxyl radical
H_0: hydroxyl anion
NADP: nicotine adenine dinucleotide phosphate
AD: Alzheimer's disease
PD: Parkinson's disease
CoA: coenzyme A
CAT: carnitine acetyltransferase

In mammals, mtDNA accounts for one% or less of total cellular DNA.[13] The fact that so few respiratory chain protein subunits are encoded by the mitochondrial genome means that the majority are synthesized in the cytoplasm and must be imported into mitochondria, where they are assembled with the appropriate mtDNA gene products into functional enzyme complexes.[14,15] These energy-dependent processes of protein importation and assembly thus require the coordinate regulation of discrete nuclear and mitochondrial DNA compartments possessing distinct genetic codes; in theory, each of these compartments and their various regulatory features are susceptible to perturbations that could influence mitochondrial function.

Mammalian mtDNA is almost exclusively maternally inherited, since the portion of the spermatozoan that contains mitochondria ordinarily does not enter the female gamete during fertilization.[16] All human cells except mature erythrocytes contain hundreds of mitochondria and up to 10 mtDNA copies per organelle,[17] so each cell contains thousands of mtDNAs. mtDNA replication uses two origins, one each for the heavy (H) and light (L) strands.[18] However, all but one of the genes that code for the respiratory chain subunits are encoded by L-strand DNA (Fig 3). Replication probably occurs throughout the cell cycle and can be influenced by changes in the metabolic needs of the cell.

Two important features of mitochondrial genetics that have crucial implications for mitochondrial pathology are the stochastic partitioning of mitochondria during cell division, a process called "replicative segregation," and the susceptibility of mtDNA to mutation. During replicative segregation, mitochondria are randomly distributed to the daughter cells (Fig 4). The biological significance of this process is highlighted by the fact that the mutation rate of mtDNA is very high, up to 17 times that of the nuclear genome.[19] Susceptibility of mtDNA to mutation is enhanced by the high concentration of reactive oxygen species formed during oxidative phosphorylation.[6,20] Moreover, mtDNA is "naked" in that it lacks histones and is thus more susceptible to direct free radical attack.[17] Some investigators consider that mitochondria contain the enzymes required to conduct excision repair processes of oxidatively modified DNA, although the mechanism is less efficient than that present in the nucleus.[21] The possibility that mtDNA polymerase may not adequately proofread DNA sequences and remove incorrectly incorporated nucleotides[21] may also markedly increase the rate of mtDNA mutation. Thus, replicative segregation provides a mechanism for unequal distribution of mutated mitochondria and hence the potential for extreme tissue heterogeneity in the phenotypic expression of the mutation.

In summary, the small size and information content of the mitochondrial genome, the lack of adequate DNA repair

Table 2. Generation of High-Energy Phosphate Ⓟ in the Catabolism of Glucose

Pathway	Reaction Catalyzed by	Method of ~ Ⓟ Production	Number of ~ Ⓟ Formed per Mol of Glucose
Glycolysis	Glyceraldehyde-3-phosphate dehydrogenase	Respiratory chain oxidation of 2 NADH	6*
	Phosphoglycerate kinase	Oxidation at substrate level	2
	Pyruvate kinase	Oxidation at substrate level	2
			10
Allow for consumption of ATP by reactions catalyzed by hexokinase and phosphofructokinase			−2
		Net	8
Citric acid cycle	Pyruvate dehydrogenase	Respiratory chain oxidation of 2 NADH	6
	Isocitrate dehydrogenase	Respiratory chain oxidation of 2 NADH	6
	α-Ketoglutarate dehydrogenase	Respiratory chain oxidation of 2 NADH	6
	Succinate thiokinase	Oxidation at substrate level	2
	Succinate dehydrogenase	Respiratory chain oxidation of 2 $FADH_2$	4
	Malate dehydrogenase	Respiratory chain oxidation of 2 NADH	6
		Net	30
Total per mol of glucose under aerobic conditions		38	
Total per mol of glucose under anaerobic conditions			2

*It is assumed that NADH formed in glycolysis is transported into mitochondria via the malate shuttle. If the glycerophosphate shuttle is used, only 2 ~ Ⓟ would be formed per mole of NADH, the total net production being 36 instead of 38. The calculation ignores the small loss of ATP due to a transport of H^+ into the mitochondrion with pyruvate and a similar transport of H^+ in the operation of the malate shuttle, totaling about 1 mol of ATP.

Reproduced with permission.[4a]

mechanisms, the high intramitochondrial level of free radicals, and the rapid turnover rate of mtDNA all contribute to the profound susceptibility of mtDNA to mutation. Furthermore, since there are no introns in the mitochondrial genome, many mutational events are likely to occur in sequences that code for respiratory chain proteins and thereby increase the probability of inducing functional alterations in oxidative phosphorylation

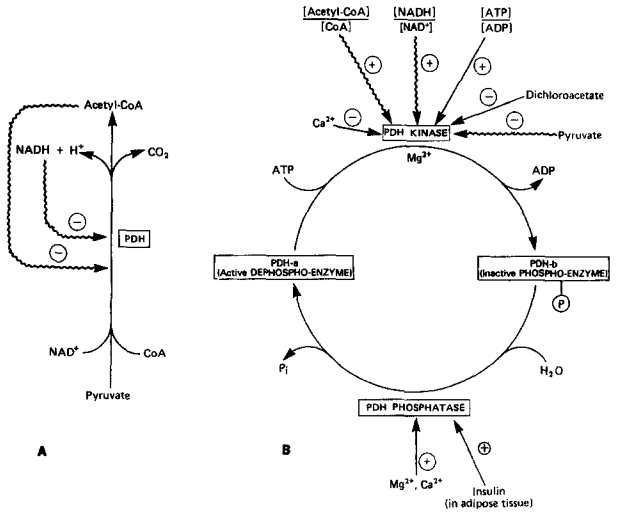

Fig 2. Regulation of the pyruvate dehydrogenase (PDH) complex. Arrows with wavy shafts indicate allosteric effects. (A) Regulation by end-product inhibition. (B) Regulation by interconversion of active and inactive forms. (Reproduced with permission[4a]).

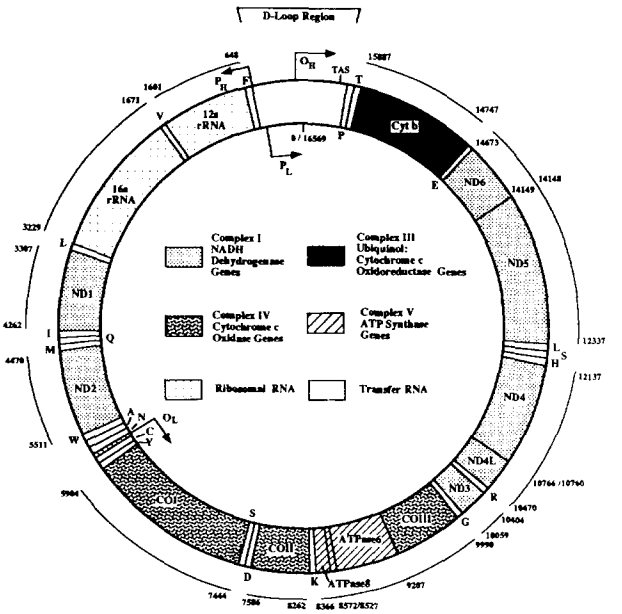

Fig 3. Human mtDNA map. mtDNA encompasses 16,569 nucleotide pairs with numbering starting at O_H (heavy-strand origin) and proceeding counterclockwise around the circular map. The function of each gene is identified by shading according to the designations within the circle. The first and last nucleotides of the rRNAs and mRNA genes are shown on the external arcs. tRNA genes are indicated by the letter of their cognate amino acid. The D-loop region is a triple-stranded region that contains a new heavy (H)-strand segment base-paired to the parental light (L)-strand. The D-loop contains a separate promoter for the H-strand (P_H) and L-strand (P_L). The origins of replication for both H-strand (O_H) and L-strand (O_L) are indicated. (Reproduced with permission, from the Annual Review of Biochemistry, vol 61, ©1992, by Annual Reviews Inc.[18a])

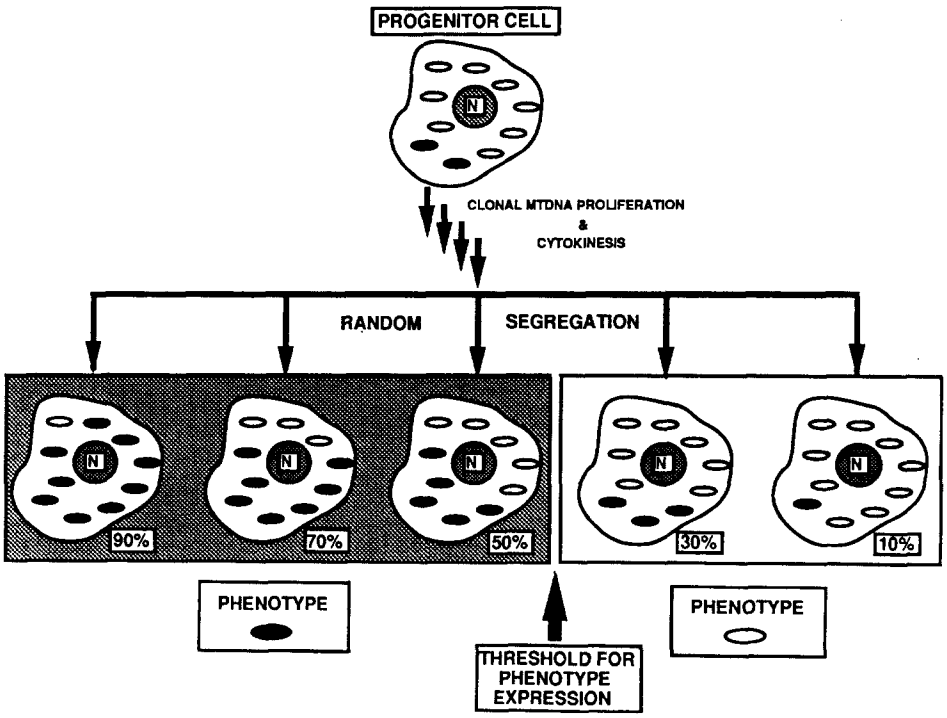

Fig 4. Replicative segregation of mtDNA. Mitochondria segregate randomly during cell division (cytokinesis). Once the mutant mtDNA reaches a certain threshold, the cell begins to express the mutant phenotype. Solid ovals represent mutant mitochondria, open ovals represent normal mitochondria, and N represents the nucleus. (Reproduced with permission.[19])

capacity. Such propensity of mtDNA for sustaining phenotypically meaningful mutations lends major pathological import to the process of random segregation of mitochondria during mitochondrial and cellular division. For a given mtDNA mutation, random segregation ensures that daughter cells receive unequal amounts of normal and mutated DNA. The existence of multiple mitochondrial genomes in the same cell confers a state known as heteroplasmy. Accordingly, the cell's threshold for manifesting the altered phenotype depends on the "dose" of mutated gene and the functional significance of the mutation. Thus, in the example illustrated in Fig 4, at least 50% of a cell's mitochondria must contain the specific DNA mutation in order to express an altered phenotype. Consequently, in organs and tissues that consist of nondividing cells and that depend on a high rate of oxidative metabolism, the acquisition of mitochondrial mutations and their heteroplasmic distribution may lead, over time, to diverse and important clinical manifestations in the host (Table 3).

MITOCHONDRIAL PATHOLOGY

Lactic Acidosis

Perhaps the most common and obvious clinical condition due to mitochondrial dysfunction is lactic acidosis. Hyperlactatemia is synonymous with lactic acidosis and ensues whenever the production of lactate exceeds its removal. It is hypothesized[1] that the majority of acquired cases of lactic acidosis reflect a fundamental abnormality in the mitochondrial metabolism of pyruvate and an imbalance between the production and hydrolysis of ATP. Septic shock typifies such a state in which tissue hypoperfusion and hypoxia lead to accelerated glycolysis and lactate production and to impaired pyruvate oxidation, possibly through inhibition of PDC activity.[22] Although mammalian mitochondria are not usually considered targets of pathogenic viruses, patients with the acquired immune deficiency syndrome may exhibit marked and persistent hyperlactatemia in the absence of other precipitating causes[23] such as hypotension or treatment with zidovudine, a known mitochondrial toxin.[24] Many other drugs such as sedatives-hypnotics, anesthetics, carbon monoxide, and cyanide can inhibit respiratory chain enzyme activity and may precipitate lactic acidosis.[1] By far the most common biochemically proven causes of congenital forms of lactic acidosis are substitution or deletion mutations in mitochondrial respiratory chain genes or in one of the PDC enzyme subunits. Highly oxidative tissues such as the brain, heart and skeletal muscle are the primary target organs for these congenital diseases.

Table 3. Basic Characteristics of Mitochondrially Inherited Diseases

1. Mode of inheritance frequently maternal (nonmendelian)
2. Impaired glucose, lactate, and pyruvate aerobic oxidation
3. Susceptibility to oxidative stress
4. Marked phenotypic heterogeneity
5. Heteroplasmic tissue distribution of genetic defect
6. Clinical expression often delayed, due to gene-dosage threshold effect

For both acquired and congenital forms of lactic acidosis, hyperlactatemia (or elevated cerebral spinal fluid lactate, or both) is a surrogate marker for impending or ongoing cellular energy failure, due to deficient mitochondrial pyruvate oxidation. The role of the lactate ion per se in the pathobiology of lactic acidosis of acute onset is controversial and, in most cases, probably does not directly influence the clinical course once blood levels exceed about 5 mmol/L.[1,2,4] By that time, widespread cellular energy deficits have contributed to multiorgan system failure and to a mortality that exceeds 80%.

Why do only a subset of critically ill patients, including those

with hypotension, develop fulminant lactic acidosis? One hypothesis to explain such variable susceptibility is that individuals who express this condition harbor subtle congenital or acquired defects in one or more mitochondrial oxidative enzymes that are manifested under conditions of increased energy demand. This is illustrated most strikingly in infants and children with congenital forms of lactic acidosis, in whom intercurrent illness such as infection may acutely elevate the chronic acid-base disturbance to life-threatening proportions. Supportive evidence for the operation of a similar mechanism in adults with "acquired" lactic acidosis would be the identification of deficiencies in enzyme activity and their correlation with the presence of mutations in the relevant genes in tissues obtained by biopsy or at autopsy.

Aging

Aging and life span are probably as much a function of mitochondrial as of nuclear genomic integrity and stability. Indeed, apoptosis, or programed cell death, among mammalian species is linked to the genetic controls of the rate of oxygen utilization, which in part reflects the rate of free radical–mediated mitochondrial damage[25,26] (and vide infra). In vivo and in vitro investigations of various animal species, including humans, show that a decline in mitochondrial respiratory capacity is a fundamental characteristic of aging.[27-36] This phenomenon may be most apparent in tissues, such as heart, skeletal muscle, and brain, in which the rate of postnatal cell division is least and in which the demand for oxidative metabolism is greatest.[37] Decrements in the activities of mitochondrial oxidative enzymes have been reported most frequently for PDC,[38-40] complex I (NADH dehydrogenase),[40-42] and complex IV (cytochrome oxidase).[33,39,41-46] An age-dependent decrease in complex IV activity is particularly noteworthy because of the differential expression of this enzyme relevant to metabolic requirements of individual tissues, cells, and subcellular sites.[29,30,36] The age-dependent decrease in respiratory activity has been linked to a decrease in the cellular content of mitochondria[28,30,36,47,48] and to an increase in the structural and functional abnormalities of mitochondrial membranes,[20,27,34,41,48] in the frequency of mitochondrial DNA mutations,[49-53] and in the production of oxygen radicals, due to respiratory chain dysfunction[25,48,52] and reduced antioxidant defenses.[28,54-56]

The age-related decline in mitochondrial oxidative phosphorylating capacity in overtly healthy humans begs the question as to whether these subtle defects become clinically relevant when tissue oxidative reserve becomes taxed by serious, intercurrent illnesses such as infection or shock. Thus, just as some genotypes (and families) exhibit specific polymorphisms in oxidative phosphorylation genes that correlate with enhanced sensitivity to aerobic training,[57] so may other genotypes (and families) define individuals at greater risk to develop age-dependent deterioration of mitochondrial oxidative function. Age-associated reduction in the reserve capacity of cells for oxidative phosphorylation means that the "threshold effect" for tissue-specific expression of that diminished reserve will occur sooner if the organism is stressed, for example, by intercurrent illness. Under these circumstances, critical deficiencies in mitochondrial fuel supply and energetics could ultimately result in the overproduction or underutilization of lactate, or both. It is within this mechanistic framework that subsets of critically ill individuals would exhibit an increased tendency to develop lactic acidosis.

Diabetes Mellitus

Several lines of evidence point to a possible role of mitochondrial dysfunction in the etiology of at least some forms of diabetes mellitus. A reciprocal relationship between the oxidative metabolism of carbohydrate and that of nonesterified long-chain fatty acids appears to exist in pancreatic β cells,[58] as it does in many other tissues.[59] The functional significance of this reciprocity is illustrated by the ability of long-chain fatty acids, added to the diet or directly to islet cells, to blunt the stimulation of insulin secretion induced by glucose.[58] PDC represents a major inhibitory site of action of fatty acids, which decrease PDC activity by feedback inhibition and by stimulation of pyruvate dehydrogenase kinase (Fig 2). Thus, the rate of mitochondrial oxidation of glucose is decreased in lieu of an accelerated oxidation of fatty acids. Oxidation of glucose beyond pyruvate appears to be essential for glucose-stimulated insulin secretion,[60] but an absolute dependency of this process on PDC activity has not been proved.

Diminished PDC activity is believed to underlie the mild, basal hyperlactatemia and the inhibition of glucose oxidation measured in the peripheral tissues of some patients with non–insulin-dependent diabetes mellitus (NIDDM).[61,62] Under these circumstances, diminished oxidative removal of glucose accounts for a portion of the decrease in whole-body glucose disposal and is not corrected by insulin administration.[62,63] Although diet and insulin resistance may be factors in the expression of abnormal tissue glucose oxidation in some patients, in others the etiology of the reduced inhibition of PDC activity is obscure. Congenital mutations of PDC, a nuclear DNA-encoded enzyme complex, have not been reported in patients with any form of diabetes, but there is no evidence that they have been sought. Such cases might arise sporadically or be transmitted in a mendelian mode of inheritance.

Another etiologic link between mitochondrial dysfunction and diabetes risk is the association of NIDDM with mtDNA mutations. A distinct clinical subtype has emerged in which NIDDM and neurosensory hearing loss occur in patients who harbor a point mutation that leads to an A to G transition at nucleotide 3,243, a conserved position in the mitochondrial gene for $tRNA^{Leu(UUR)}$.[64-75] This mutation is also present in about 80% of children with a form of congenital lactic acidosis termed MELAS, for mitochondrial encephalomyopathy, lactic acidosis, and stroke-like episodes.[3] Cardiomyopathy[64] and short stature[69] may coexist in some NIDDM patients with the MELAS mutation, but none of the classic clinical features of the syndrome affecting children has been reported in diabetic adults who possess the identical mtDNA mutation. A possible exception to this rule is the association of both NIDDM and IDDM in a family with the MELAS mutation, in whom the proband developed lactic acidosis, myopathy, and circulating insulin antibodies.[67] Thus, phenotypic hereogeneity, a hallmark of inherited or acquired mitochondrial disorders, is reflected in the point mutation for $tRNA^{Leu(UUR)}$.

Maternal transmission of NIDDM and hearing loss occurs in

families with the MELAS mutation, a finding consistent with a mitochondrial etiology of disease. A maternal mode of inheritance has also been described for a family in which NIDDM and IDDM occurred in members with a large (10.4-kilobase) deletion of the mitochondrial genome.[74] Although the prevalence of any specific mtDNA mutation in the general diabetic population is unknown, it is noteworthy that several studies indicate that an increased frequency of a maternal history of NIDDM exists in diabetic offspring,[77-80] a finding not consistent with the traditional view that NIDDM is inherited as a number of autosomal gene variants.[78] Increased maternal inheritance of gestational diabetes has also been observed.[81,82]

Finally, a mitochondrial etiology of diabetes is consistent with the fact that known diabetogenic agents, such as alloxan and streptozotocin, are mitochondrial toxins or cause mtDNA mutations and lead to inhibition of cellular oxidative phosphorylating capacity.[83,84] Other mitochondrial poisons, such as cyanide, azide, and dinitrophenol, have variably been reported to cause deafness, basal ganglia disorders, neuromuscular deterioration, and inhibition of insulin secretion,[84] ie., effects similar to known mitochondrial mutations that give rise to congenital lactic acidosis and diabetes.

In children with the MELAS mutation, mitochondrial respiratory chain activity is decreased, and lactic acidosis ensues early in life, due to the block in pyruvate oxidation.[3] In such patients, highly oxidative tissues such as the nervous system become the principal anatomic targets, resulting in lactate accumulation in both cerebrospinal fluid and blood. Therefore, why do some patients with the identical mtDNA mutation present clinically later in life with islet cell failure and insulin resistance? Recall that mitochondrial heteroplasmy results in unequal distribution of normal and mutated mtDNA during cell division and accounts for the variability in time of onset and tissue expression. In those cases in which a high frequency of mutation occurs in cells of the developing nervous system, a threshold effect for clinical expression of the genetic abnormality is achieved early and is reflected by neuromuscular deterioration. In contrast, individuals in whom mtDNA mutations are either less frequent or less severe may express their disease when the cumulative environmental stresses of obesity, sedentary lifestyle, high-fat intake, and aging unmask the congenital defect in islet cells and insulin-sensitive tissues. Thus, whether the genetic etiology involves nuclear DNA, as in the case of PDC, or mtDNA, as in the case of the enzymatically more distal MELAS defect, a bottleneck occurs in the oxidative metabolism of glucose and its glycolytic intermediate, pyruvate. If the resulting accumulation of lactate exceeds the capacity of cells with enzymatically intact mitochondria to oxidize it, hyperlactatemia results. Regardless, individual cells that are unable to efficiently convert carbohydrate fuel into energy fall below a critical functional threshold, begin to fail, and, in the case in point, exhibit insulin deficiency, insulin resistance, or both.

Ischemia

The most common disorders of humans are those of the heart and brain, which are also the organs most dependent on aerobic glucose metabolism. Hence, it is not surprising that mitochondrial dysfunction is integral to the pathway of tissue damage in many forms of acute or chronic cardiac and neurological diseases.

A dramatic illustration of this point is the pathobiology of tissue injury due to ischemia and subsequent reperfusion. Sudden deprivation of oxygen and other nutrients (glucose in the case of brain cells and glucose and fatty acids in the case of cardiac cells) precipitates a cascade of metabolic events[84-89] in which cellular energy failure assumes an early and prominent role (Fig 5). A common response to abrupt withdrawal of oxygen and metabolizable substrates is an upregulation of glucose transport and glycolysis,[83,90,91] as cells attempt to maintain ATP levels despite diminished fuel supply. As ATP is catabolized, intracellular levels of adenosine, inosine, and hypoxanthine increase, and as the oxidative phosphorylating capacity of the mitochondria diminishes, more electrons "leak" from the respiratory chain enzymes, principally at complex I and ubisemiquinone, which normally are the sites for generating most respiratory chain free radicals.[6,20,88,92,94] In the setting of advancing mitochondrial energy failure, loss of adenosine diphosphate (ADP) diminishes the functional capacity of complex IV to use electrons, resulting in further leakage from the respiratory chain. Intramitochondrial levels of reduced glutathione and the activity of manganese superoxide dismutase also decline during ischemia.[95] Thus, a vicious cycle ensues in which intramitochondrial free radical accumulation decreases mitochondrial membrane integrity, leading to further respiratory chain dysfunction, increased electron leakage, and diminished antioxidant activity. Ultimately, lactate and hydrogen ions accumulate extracellularly, due to the increased glycolytic rate and the inability of mitochondria to oxidize pyruvate.

Energy failure itself can lead to irreversible tissue injury, but it is not the only or immediate cause of cell death. Extreme intracellular lactic acidosis, in which lactate levels exceed 15 mmol/L and pH decreases to less than 6.5, may also be toxic to cells.[88,94,96-103] This is thought to occur, in part, because of the ability of acidosis to increase the rate of membrane lipid peroxidation and depolarization.[88,94,97,101] Combined energy failure and intracellular lactic acidosis stimulate proteolysis and inhibit the synthesis and release of neurotransmitters.[104] It has been reported that ischemia-associated lactic acidosis may protect myocytes by augmenting ATP synthesis[105] and may preserve neurons by decreasing *N*-methyl-D-aspartate (NMDA) and kainate receptor activation and neurotoxicity.[102,106-109] However, other studies indicate that accumulation of lactate ions, protons, or both correlate inversely with synaptic function, recovery of electroencephalographic activity, and survival of myocardial and neuronal cells.[94,108,110-113]

Because the NMDA (glutamate) receptor in brain is voltage-dependent, its normal activity relies, in part, on the integrity of the pathway of aerobic glucose oxidation for maintenance of energy-dependent ion pumps and membrane potentials within the cells. In this regard, studies of cultured cerebellar neurons deprived of oxygen and glucose[114,115] are consistent with the premise that the transition of glutamate from neurotransmitter to neurotoxin is facilitated when energy failure occurs. In similar fashion, energy failure may be instrumental in converting nitric oxide from a neurotransmitter to a neurotoxin.[116,117] Thus, substrate metabolism, ATP synthesis, and functional sodium, potassium ATPase enzymes are required to generate a

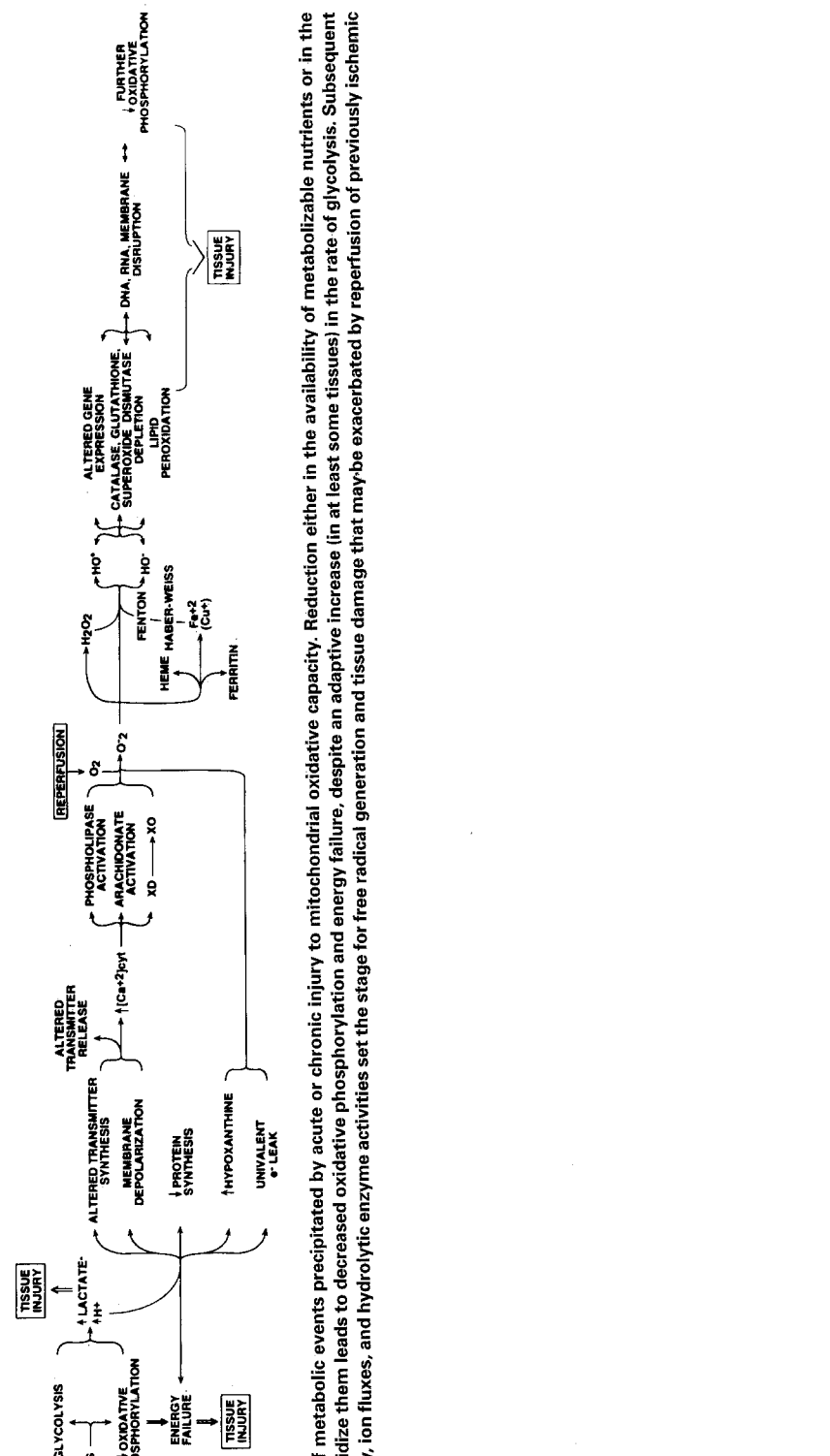

Fig 5. Postulated sequence of metabolic events precipitated by acute or chronic injury to mitochondrial oxidative capacity. Reduction either in the availability of metabolizable nutrients or in the capacity of mitochondria to oxidize them leads to decreased oxidative phosphorylation and energy failure, despite an adaptive increase (in at least some tissues) in the rate of glycolysis. Subsequent changes in membrane integrity, ion fluxes, and hydrolytic enzyme activities set the stage for free radical generation and tissue damage that may be exacerbated by reperfusion of previously ischemic cells.

resting membrane potential sufficient to maintain voltage-dependent regulation of the NMDA receptor channel. When energy failure relieves the inhibition of the channel, excitatory amino acid neurotoxicity may ensue.

Loss of energy-dependent ion pumps and ionic homeostasis contribute to an increase in the flux of calcium ions into the cytoplasm.[88,118] Although some studies have failed to find a close correlation between cytoplasmic calcium levels and cytotoxicity,[119,120] others have demonstrated that calcium-mediated activation of cellular phospholipases and nucleases, conversion of xanthine dehydrogenase to xanthine oxidase, and stimulation of excitatory neurotransmitter release can establish an intracellular environment conducive to the generation of free radicals upon reoxygenation of the ischemic tissue.[85-88,92,100,120-122] With resumption of perfusion, oxygen becomes available as a substrate for several reactions that yield hydrogen peroxide (H_2O_2) and various reactive oxygen species such as the superoxide anion radical (O_2^-), formed by the reaction of hypoxanthine with xanthine oxidase and by cyclooxygenase-mediate metabolism of arachidonate, hydroxyl radical ($HO^{\cdot}$), and hydroxyl anion (HO^-). Of these, the most reactive specie is $HO^{\cdot}$, which is generated when H_2O_2 interacts with transition metals (iron and copper) in hemoglobin, ferritin, or cytochromes in the Fenton and Haber-Weiss reactions. Oxygen radicals subsequently attack proteins and unsaturated fatty acids, causing inactivation of enzymes, autoxidation of catecholamines and lipids, and disruption of membranes.[123-126] Oxygen radicals also alter the cellular NAD(P) redox state, which, in turn, is thought to contribute to altered gene expression and DNA breaks.[86,127] Continued derangements of the mitochondrial electron transport system lead to increased univalent reduction of oxygen and further loss of oxidative phosphorylating capacity.

Early in the ischemic (or hypoxic) episode, ATP is used faster than it is synthesized. Although the cytoplasmic ATP/ADP · Pi ratio is considered the primary sensing mechanism for changes in cellular energy requirements, the mitochondrial NAD/NADH ratio allows the rate of ATP production to change at a given ATP/ADP · Pi ratio.[84] Depletion of oxygen and mitochondrial carbon substrates during ischemia inhibits electron transport and reoxidation of NADH, thereby making the mitochondrial redox state more reduced. Thus, mitochondrial respiration fails and electrons increasingly leak from the respiratory chain during ischemia, due to the combined consequences of hypoxia, damage to mitochondrial respiratory chain proteins, imbalance in ATP production relative to ATP consumption, and failure to maintain a normal intramitochondrial redox state.

Congenital or acquired mutations of mitochondrial oxidative enzymes may be expected to increase the susceptibility to tissue damage due to ischemia/reperfusion injury. Other age-related decrements in mitochondrial number and functional capacity and in cellular antioxidant defenses[28,56,128-130] further undermine the ability of host tissues to defend against an ischemic insult. Mutational deficiencies of PDC or any cytochrome should facilitate free radical generation, as do such specific respiratory chain poisons as antimycin and cyanide, because of the accumulation and autoxidation of reduced cytochrome flavins and quinones proximal to the enzymatic block.[48,85] Both PDC and PDC kinase, which inactivates PDC by reversible phosphorylation (Fig 2), may be susceptible to modulation by free radicals by altering their thiodisulfide status.[128,131,132] Under normal conditions in brain, PDC is in its predominantly active, unphosphorylated state and pyruvate flux through PDC is estimated to be only about twice that required to sustain normal oxidative capacity.[133,134] Thus, a congenital or acquired defect in PDC sufficient to decrease flux more than 50% should inhibit brain carbohydrate oxidation, ATP synthesis, and acetylcholine formation, since the acetyl CoA produced by the PDC-catalyzed step is used in the formation of this neurotransmitter.[104,135,136] Consequently, transsynaptic cholinergic transmission may be impaired and may further reduce PDC activity, since repetitive synaptic stimulation is reported to increase brain PDC activity via dephosphorylation.[135-137]

In contrast to neural tissue, healthy myocardial cells at rest rely predominantly on long-chain fatty acids as substrate fuel; thus, myocardial PDC is predominantly phosphorylated and inactive.[138] Increased cardiac work normally increases the requirement by myocytes for glucose as an energy source, and PDC activity increases. However, the capacity to increase myocardial aerobic carbohydrate oxidation may be impaired in patients with congestive heart failure,[139] and recent evidence indicates that patients with ischemic[140] or hypertrophic[141-143] cardiomyopathies may have a significantly increased frequency of mtDNA mutations in myocardial cells that could further limit cardiac reserve during ischemic stress. Whether the apparent change in the rate of mtDNA mutations is the cause or consequence, or both, of the chronically ischemic state in these individuals is not known. However, it is noteworthy that hypertrophic cardiomyopathies, conduction disturbances, and other cardiac abnormalities are also found in children with congenital lactic acidosis due to mtDNA mutations.[144]

Neurodegenerative Diseases

The cellular metabolic requirements and pathways of the nervous system are relatively straightforward: glucose is the principal substrate fuel, and aerobic oxidation of glucose is required to sustain normal neurologic function. Therefore, a premise linking mitochondrial dysfunction to neurodegenerative diseases should be compatible with evidence of acquired or congenital defects in mitochondrial energy production. The most obvious examples of such linkage are the congenital lactic acidoses, most of which are due to duplications, deletions, or point mutations in genes coding for PDC or a respiratory chain enzyme.[145] In these disorders, the number of defective mitochondria exceeds the threshold for phenotypic expression early in life and leads to progressive neuromuscular deterioration.

In other neurodegenerative diseases, particularly those in which the clinical onset occurs in adults, a mitochondrial etiology is more tenuous. However, in Alzheimer's disease (AD), diminished cerebral glucose oxidation, estimated by positron emission tomography, is reported to precede the appearance of significant cognitive defects,[146,147] and analysis of brain biopsies from AD patients shows evidence consistent with uncoupling of oxidative phosphorylation.[146] Morphologic abnormalities in mitochondria[30] and diminished activities of PDC[148-151] and cytochrome oxidase[30] in various brain regions have been observed in autopsy specimens from AD subjects compared with tissues from age-matched controls. Indeed, the

apparent decrease in PDC activity, determined in both histologically involved and uninvolved cortical brain areas of AD patients,[150] is consistent with the hypothesis that morphologic damage is a consequence rather than a cause of mitochondrial energy failure. Moreover, the suggestion that PDC activity determines the rate of acetylcholine synthesis in neurons[152] is consistent with reports in which the degree of inhibition of PDC is proportional to the cholinergic defect,[148,153] as measured by diminished choline acetyltransferase activity and regional decrements in the number of cholinergic neurons.

Recent evidence also indicates that mitochondrial defects may underlie the pathobiology of Parkinson's disease (PD). First, a mitochondrial mode of inheritance for PD is consistent with the discordance of the condition in identical twins.[154] Second, mitochondrial DNA deletions are reported to occur more frequently in the corpus striatum of PD patients at autopsy than in age-matched controls,[155] as does a decrease in the activity or protein level of complex I in specific brain regions,[146,156,157] skeletal muscle,[156] and platelets[154] of PD individuals. Third, some of the neurological findings in PD also occur in subjects exposed to such mitochondrial poisons as carbon monoxide and cyanide[154,158] or to drugs that may enhance the production of free radicals.[151]

Evidence of abnormal mitochondrial function has been reported for several other neurodegenerative diseases,[159,160] including Huntington's disease (HD), in which regional decreases in cortical glucose oxidation in vivo[161] or reduced PDC or complex I activities in autopsied brain[149] or platelets[162] have been observed. The fact that HD is an autosomal dominantly transmitted disease implies that congenital defects of oxidative metabolism must derive from nuclear gene mutations if they are to have etiologic significance.

In neurons, mitochondria are concentrated in dendrites and axon terminals,[146,163] ie, at the sites of ion flux and synaptic transmission. The normal heterogeneous distribution of respiratory chain enzymes among and even within neurons of varied anatomic location[30,36] and the existence of isoforms for certain subunits of these proteins[30] confer altered degrees of mitochondrial oxidative capacity and hence varying regional, cellular, and subcellular susceptibility to acquired or hereditary metabolic stresses. A critical unresolved issue is whether the reported mitochondrial mutations or other alterations in mitochondrial energy metabolism are unrelated to disease onset or whether they represent important factors in the etiology of neurodegenerative diseases. Regardless, it is intuitively obvious that if pyruvate oxidation is indeed rate-limiting for energy production by brain cells, inhibition of that process is certain to produce functional consequences for those cells that will be expressed clinically in proportion to the severity of the inhibition and to the number and location of affected cells. Whether the clinical threshold of expression is reached in infancy or childhood as in the case of congenital lactic acidosis, in mid-life as, for example, in HD, or in old age as in AD or PD, a fundamental biochemical feature of neurodegenerative diseases is mitochondrial energy failure.

TREATMENT OF MITOCHONDRIAL DISORDERS

Preservation of residual mitochondrial (and cellular) function in the face of acquired or congenital mitochondrial disorders would seem to require, at a minimum, successfully addressing two goals: first, maximize mitochondrial ATP synthesis by facilitating aerobic glucose oxidation, and second, minimize free radical production by reversing cellular energy failure and increasing the efficiency of endogenous free radical scavenging mechanisms.

Stimulation of Glucose Oxidation

Since PDC catalyzes the rate-limiting step in the aerobic oxidation of glucose, lactate, and pyruvate, it is reasonable to consider this enzyme complex a therapeutic target. Stimulation of PDC activity has the potential advantages of reducing intracellular lactic acidosis and priming the tricarboxylic acid cycle with substrate in the form of acetyl CoA. Oxidation of pyruvate by PDC also generates reducing equivalents that could be used by the respiratory chain to synthesize ATP. However, a theoretical disadvantage of PDC activation is the possibility that the increase in reducing equivalents could tax a respiratory chain already compromised by damage to one or more of its enzymes and exacerbate, rather than alleviate, energy failure and lactic acidosis.

As noted previously, PDC is regulated posttranslationally by reversible phosphorylation, product inhibition, and substrate activation. In tissues such as myocardium, skeletal muscle, and pancreatic islets in which the oxidation of carbohydrate and fatty acids are reciprocal processes, inhibition of fatty acid oxidation stimulates PDC by relieving feedback inhibition from elevated intramitochondrial ratios of NADH/NAD and acetyl CoA/CoA.[59] Accordingly, drugs that inhibit adipose tissue lipolysis, long-chain fatty acid uptake by mitochondria, or mitochondrial fatty acid or ketone body oxidation can potentially activate PDC and facilitate pyruvate oxidation.[164] However, such agents would be unlikely to alter PDC activity and aerobic glucose metabolism in neurons, since fatty acid oxidation is not normally an active process in these cells.

The naturally occurring zwitterion carnitine [$(CH_3)_3N^+$-CH_2-$CH(OH)$ -CH_2-COO^-] is widely distributed in tissues and derivatizes long-chain fatty acids for transport across the mitochondrial inner membrane.[165,166] It also interacts with the intramitochondrial enzyme carnitine acetyltransferase (CAT) to catalyze the transfer of short-chain acyl groups between CoA and carnitine:

$$\text{acetyl CoA} + \text{carnitine} \xrightarrow{\text{CAT}} \text{acetylcarnitine} + \text{CoA}.$$

Thus, provision of free carnitine to cells affords the possibility of shifting the CAT reaction to the right, liberating CoA and relieving the block on PDC induced by a high ratio of acetyl CoA/CoA. Indeed, in vitro addition of carnitine to mammalian tissues results in stimulation of PDC activity, presumably by this mechanism.[165,168] Although carnitine has been administered safely to children with congenital lactic acidosis, its efficacy in the acute or chronic enhancement of mitochondrial energy metabolism in vivo remains to be determined.

Pyruvate, at millimolar concentrations, can stimulate PDC activity. In this regard, it is noteworthy that substrate levels of this intermediate improve aerobic glucose oxidation and left-ventricular function following reperfusion of ischemic, isolated

heart preparations[169-173] and reduce glutamate excitotoxicity in cultured neurons.[115] The in vitro cardioprotective effect of pyruvate is as great as or greater than when glucose is the only metabolizable sustrate present in the reperfusion medium.[169] Pyruvate replenishes, through anaplerotic reactions, critical intermediates of the tricarboxylic acid cycle that are depleted by ischemia.[174] Moreover, its oxidized product, acetyl CoA, activates pyruvate carboxylase and thus also stimulates anaplerosis.[175] However, provision of increased levels of intracellular pyruvate by stimulation of glycolysis might exacerbate tissue lactic acidosis, unless its rate of oxidative removal were increased accordingly. Whether intravenous administration of pyruvate salts would help maintain mitochondrial oxidative phosphorylating capacity and increase cell survival during acute ischemic episodes is unknown.

Dichloroacetate (DCA) is a potent stimulator of aerobic glucose, lactate, and pyruvate oxidation by virtue of its ability to inhibit pyruvate dehydrogenase kinase and thereby "lock" PDC in its unphosphorylated, catalytically active form.[176] Activation of PDC occurs within minutes of DCA administration and occurs in virtually all tissues, including brain.[177] Blood, cerebrospinal fluid, and intracellular lactate concentrations are decreased by the drug under conditions of experimental myocardial or cerebral ischemia, sepsis, hypotension, and liver failure.[176] DCA activates myocardial PDC, stimulates glucose oxidation and inhibits fatty acid oxidation. It is reported to and, in some cases, increases ATP levels when added to the perfusion media following global ischemia of isolated rodent hearts, and increase recovery of left-ventricular function during reoxygenation.[176,178,179] Intravenous DCA reduces blood lactate levels and increases blood bicarbonate levels and pH in adults with acquired causes of lactic acidosis,[4,176] but does not alter survival. The drug also improves acid-base status in children with congenital lactic acidosis due to partial deficiencies of PDC, complex I, or complex IV of the respiratory chain.[176,177,180,181] DCA exerts a positive inotropic effect in some patients with hypotension,[59] congestive heart failure,[182] or coronary atherosclerosis,[183] and increases both the contractile efficiency and the extraction of lactate by myocardium under these conditions. The drug has not been administered to patients with unstable angina or acute myocardial infarction, and it is not known if preservation of myocardial tissue in humans can be enhanced by DCA administered during or after an ischemic insult.

Phosphorus magnetic resonance spectroscopy has been used to demonstrate that treatment of rats with DCA before or after induction of global cerebral ischemia decreases tissue lactate and increases intracellular pH during subsequent reperfusion.[184] An interesting correlate to these findings is that DCA administration increases intracellular glutamate levels. By improving tissue acid-base status during the reperfusion period, DCA may limit excitatory amino acid release from neurons threatened with energy failure and improve cell survival, but this postulate remains untested.

However, several lines of evidence indicate that DCA may, in fact, reduce neuronal cell damage from acute or chronic mitochondrial energy failure and tissue lactic acidosis. In rats, DCA significantly improves central nervous system function following brain ischemia or spinal cord injury.[185-187] Brain PDC activity is stimulated by DCA both under basal conditions and following ischemia/reperfusion injury,[188-195] and this is associated with increased tissue glucose[196] and pyruvate[188] utilization, decreased tissue lactate levels,[184,197-200] and, under certain conditions, decreased cell swelling.[199] Both PDC activity and pyruvate oxidation are reported to be tightly coupled to membrane depolarization and synaptic transmission,[137,188] and the intracellular lactic acidosis due to inhibition of these processes can cause cell damage either directly or by potentiating other mechanisms, such as cytoplasmic free calcium accumulation, lipid peroxidation and diminished neurotransmitter synthesis (Fig 4).

Finally, DCA has shown promise in the long-term (up to 5 years) treatment of infants and children with congenital lactic acidosis.[176] Approximately 80% of the patients have demonstrated a lactate-lowering effect in blood, cerebrospinal fluid, or both. In those cases appropriately studied, improvement in acid-base status has frequently been associated with increased neurological function. The fact that the drug appears to benefit not only patients with PDC deficiency but also those with defects in complex I or complex IV suggests that the respiratory chain is normally not saturated under these conditions and can accommodate the presumed increase in reducing equivalents due to PDC stimulation.

In summary, DCA may be a prototype for a class of drugs that would act rapidly and specifically to preserve mitochondrial function in the face of acute or chronic conditions that threaten cellular energy homeostasis and thereby cell survival.

Free Radical Quenching

Although reactive oxygen species are products of normal respiratory chain activity, the vulnerability of mitochondria to free radical attack makes them an important, albeit challenging, target in attempting to limit cell damage from free radicals. The utility of so-called "lazaroids" in this regard awaits the outcome of controlled clinical trials. These amino-steroid–based antioxidants have the capacity to bind to transition metal ions and prevent their participation in radical-generating reactions.[86,94] A potential shortcoming of some lazaroids and other antioxidants such as deferoxamine and superoxide dismutase is the difficulty in delivering these molecules intracellularly with sufficient rapidity and at an acceptable concentration to be beneficial. Even if these agents were to gain ready access to the cytoplasmic compartment of cells, their uptake by mitochondria, whether by diffusion or active transport, is problematic.

What characteristics might be required of an effective antioxidant/free radical scavenger if it is to be administered exogenously as a drug? First, the ideal compound would be integral to the normal diet and metabolic milieu of the host. Thus, it would be a naturally occurring substance. Second, it should be soluble in plasma and sufficiently safe to be administered as a bolus or rapid infusion immediately upon establishing venous access, either in a hospital setting or elsewhere. Third, the kinetics of its transport across the blood-brain barrier and its uptake by target cells should be rapid, enabling therapeutic levels to be achieved within minutes of its administration. In this regard, the ideal compound would have a low molecular weight, be relatively lipophilic, and not require active transport across membranes, ie, it would not further drain the already limited energy reserves of cells. Rapid uptake and even

concentration by mitochondria would also be highly desirable. Fourth, a clinically effective drug would not only facilitate quenching of free radicals already generated as a result of injury, but would inhibit their further production. This latter property could be achieved, for example, (1) by augmenting levels of endogenous antioxidants; (2) by maintaining a high level of tricarboxylic acid activity and oxidative phosphorylation, thus preserving ATP levels and intracellular pH and reducing electron leakage from respiratory chain enzymes; (3) by decreasing ATP breakdown, thereby limiting the amount of available substrate for xanthine oxidase; (4) by preventing cytoplasmic accumulation of calcium ions; (5) by preventing conversion of xanthine dehydrogenase to xanthine oxidase; and (6) by stabilizing plasma and intracellular membranes to reduce their vulnerability to lipid peroxidation.

Pyruvate itself may possess several qualities inherent in a useful antioxidant/free radical scavenger. Alpha-ketoacids such as pyruvate are capable of metabolizing H_2O_2 to carbon dioxide and water and thus preventing its conversion to ·OH by the iron-catalyzed Fenton reaction.[201] Substrate levels of pyruvate administered to isolated, perfused rat hearts following no-flow ischemia improved the recovery of mechanical function, attributed, at least in part, to decreased free radical generation.[172,173] In addition, similar studies indicate that pyruvate blunts the loss of both intracellular glutathione and intramitochondrial reduced sulfhydryl groups during reperfusion of ischemic hearts,[202] although the mechanism for these effects is unknown. These antioxidant properties, coupled with the metabolic actions of pyruvate previously discussed, make it a potentially useful therapeutic agent to prevent tissue injury due to mitochondrial energy failure and increased free radical production.

Other important endogenous antioxidants are vitamins C and E and the vitamin A derivative, β-carotene. Ascorbate can replace O_2^- as a reducing agent and may thereby decrease O_2^--dependent formation of ·OH in the presence of transition metals. This property of ascorbate probably occurs only at millimolar concentrations within cells, but these levels exist normally in some mammalian tissues such as the eye, nerves, pneumocytes, and neutrophils.[94] Vitamin C may also increase glutathione levels within certain cells,[203,204] which may improve insulin sensitivity in both healthy and diabetic subjects.[205] Experimental and epidemiological data indicate that vitamins C and E and β-carotene may protect against coronary heart disease.[206-209] Like vitamin C, vitamin E also reportedly improves insulin action,[210] perhaps by decreasing the excess free radical formation and lipid peroxidation thought to occur in diabetes.[211]

DCA may exert an indirect antioxidant effect by its ability to promote mitochondrial oxidative reactions and thus, in theory, decrease electron leak from the respiratory chain and preserve normal electron flow. Allopurinol probably decreases myocardial ischemia/reperfusion injury by inhibiting xanthine oxidase activity.[212] Neither drug has been used clinically to inhibit radical-mediated tissue injury.

Finally, marine polyunsaturated fatty acids of the omega-3 family represent an intriguing class of nutrients that may protect against cellular energy failure. Dietary supplementation or emulsified infusions of omega-3 fatty acids improve acid-base status in guinea pigs with endotoxin-induced lactic acidosis.[213,214] Similar effects were achieved by treatment with pharmacologic inhibitors of prostaglandin synthesis or thromboxane A2 action. Clinical investigations suggest that long-term consumption of marine lipids may reduce the risk of cardiovascular mortality.[215-217] Although this has been attributed to various putative antiatherogenic effects of omega-3 fatty acids,[218] recent animal studies indicate that they also acutely reduce myocardial injury and ventricular arrhythmias induced by ischemia/reperfusion, compared with other classes of long-chain fatty acids.[219,220] The rapidity of the antiarrhythmic effect of fish oils cannot be accounted for by their incorporation into the plasma membranes of myocytes, but rather by an apparent inhibition of cytoplasmic calcium accumulation.[220] Thus, by as yet unclear mechanisms, omega-3 fatty acids may act at relatively proximal steps in the metabolic paradigm of tissue ischemia (Fig 5) to improve mitochondrial oxidative metabolism and acid-base status and modulate ion transport across cell membranes.

CONCLUSIONS

Fully functional mitochondria are integral to the ability of cells to survive in the face of sudden or chronic energy failure. Loss of mitochondrial integrity is thus consistent with and integrates the pathobiology of aging and of numerous acquired and congenital disorders of metabolism. Although hyperlactatemia is the most common and readily apparent biochemical marker of mitochondrial energy failure, it represents a relatively late expression of this phenomenon and one that only occurs when the inability to use the excess lactate and protons becomes widespread among host cells. Thus, by the time acute systemic lactic acidosis becomes manifest, it is often little more than an epiphenomenon in the course of a dying human being.

Treatment of acute systemic and intracellular lactic acidosis should, in general, be directed at improving oxidative removal of lactate and hydrogen ions, rather than at inhibiting their formation, since inhibition of glycolysis in hypoxic or ischemic cells may further reduce energy stores. Moreover, unless the process of reoxygenating these cells is coupled temporally to stimulating pyruvate oxidation, reactive oxygen species will be generated in increased amounts and recovery of mitochondrial ATP synthesis will lag or be prevented by radical-induced damage to respiratory chain enzymes. Neither treatment of an acute ischemic crisis nor the prevention of more chronic causes of mitochondrial disease will likely be achieved by any single therapeutic intervention currently available. However, specific intramitochondrial sites that may be attractive targets for chemotherapy are PDC and complexes I and IV of the respiratory chain, since defects at these sites have been identified repeatedly in the pathogenesis of aging and of acute and chronic degenerative disorders. Additional laboratory and clinical investigations are required to evaluate the additive, possibly synergistic, effects of naturally occurring and synthetic agents that act to enhance mitochondrial oxidative capacity and reduce free radical generation.

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