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Review Article

Pathophysiology and treatment of focal cerebral ischemia

Part I: Pathophysiology

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This article examines the pathophysiology of lesions caused by focal cerebral ischemia. Ischemia due to middle cerebral artery occlusion encompasses a densely ischemic focus and a less densely ischemic penumbral zone. Cells in the focus are usually doomed unless reperfusion is quickly instituted. In contrast, although the penumbra contains cells "at risk," these may remain viable for at least 4 to 8 hours. Cells in the penumbra may be salvaged by reperfusion or by drugs that prevent an extension of the infarction into the penumbral zone. Factors responsible for such an extension probably include acidosis, edema, K^+/Ca^{++} transients, and inhibition of protein synthesis.

Central to any discussion of the pathophysiology of ischemic lesions is energy depletion. This is because failure to maintain cellular adenosine triphosphate (ATP) levels leads to degradation of macromolecules of key importance to membrane and cytoskeletal integrity, to loss of ion homeostasis, involving cellular accumulation of Ca^{++}, Na^+, and Cl^-, with osmotically obligated water, and to production of metabolic acids with a resulting decrease in intra- and extracellular pH.

In all probability, loss of cellular calcium homeostasis plays an important role in the pathogenesis of ischemic cell damage. The resulting rise in the free cytosolic intracellular calcium concentration (Ca^{++}) depends on both the loss of calcium pump function (due to ATP depletion), and the rise in membrane permeability to calcium. In ischemia, calcium influx occurs via multiple pathways. Some of the most important routes depend on activation of receptors by glutamate and associated excitatory amino acids released from depolarized presynaptic endings. However, ischemia also interferes with the intracellular sequestration and binding of calcium, thereby contributing to the rise in intracellular Ca^{++}.

A second key event in the ischemic tissue is activation of anaerobic glucolysis. The main reason for this activation is inhibition of mitochondrial metabolism by lack of oxygen; however, other factors probably contribute. For example, there is a complex interplay between loss of cellular calcium homeostasis and acidosis. On the one hand, a rise in intracellular Ca^{++} is apt to cause mitochondrial accumulation of calcium. This must interfere with ATP production and enhance anaerobic glucolysis. On the other hand, acidosis must interfere with calcium binding, thereby contributing to the rise in intracellular Ca^{++}.

Key Words • **cerebral ischemia** • **penumbra** • **reperfusion** • **brain injury** • **acidosis** • **ion flux**

In spite of improvements in the prevention of vascular disease, focal cerebral ischemia still represents a major cause of morbidity and mortality. Focal ischemia encompasses cerebrovascular disease (stroke), subarachnoid hemorrhage (SAH), and trauma. Progress has been made in the pharmacological and surgical treatment of SAH;[65,106] however, there is at present no adequate therapy for stroke and, until very recently, experimental work gave few useful hints. The situation has now changed since exploration of pathophysiological events has prepared the ground for pharmacological treatment along two major lines. In one, attempts are made to reduce excessive influx of calcium into cells or to prevent the toxicity of glutamate and other excitatory amino acids (EAA's), which is probably mediated by calcium influx. The second is based on evidence that

Definitions of Abbreviations

ADP = adenosine diphosphate
AMPA = amino-3-hydroxy-5-methyl-4-isoazole propionic acid
ATP = adenosine triphosphate
ATPase = adenosine triphosphatase
DAG = diacylglyceride
EAA = excitatory amino acid
ECF = extracellular fluid
GABA = γ-aminobutyric acid
NMDA = N-methyl-D-aspartate
PCr = phosphocreatine
PLC = phospholipase C
VSCC = voltage-sensitive calcium channel

sustained ischemia leads to a potentially devastating production of toxic free radical species or of nonradical species that have a similar effect in causing inflammatory reactions in the metabolically perturbed tissue, reactions that may have the microvessels as their main targets. Thus, this line of treatment is concerned with pharmacological agents that can scavenge or antagonize such species. Similar treatment principles seem applicable to SAH and trauma.

In discussing recent advances in this field, I will draw information from results on experimentally induced focal ischemia. This is because such studies give a comprehensive account of pathophysiological events and treatment strategies. A recent criticism of experimental stroke models makes this a seemingly controversial approach. Such models were criticized on grounds that they were poor replicates of human stroke and that the treatment modalities derived from them had failed to ameliorate brain damage in man.[155] However, no previous experimental work has identified therapeutic principles that were unequivocally efficacious; besides, the present clinical handling of stroke patients does not allow pharmacological intervention within the therapeutic intervals suggested by experimental studies.

The following discussion is mainly based on results obtained in focal ischemia due to experimental occlusion of a middle cerebral artery (MCA). With relatively few exceptions, citations will be confined to original articles and reviews. The reader interested in the general pathophysiology of ischemia is encouraged to consult an earlier review published in this journal.[120] I also advise the reader to consult a later article, focused on focal ischemia.[80] Since then, significant progress has been made. A useful overview of recent advances in the pharmacology of brain ischemia is provided by the proceedings of the last Marburg meeting.[62]

In Part I of this review, I will discuss current knowledge on the pathophysiology of ischemic brain damage in stroke. Part II will consider the mechanisms involved and the principles whereby damage can be ameliorated.[123]

Critical Perfusion Rates, Reperfusion, and Neuropathology

Ischemia due to MCA occlusion differs from global or forebrain ischemia in two major respects.[32,34,86,116] First, the reduction in cerebral blood flow (CBF) caused by MCA occlusion is usually less severe than that observed during global or forebrain ischemia, and it is usually much more sustained. This is dramatically illustrated by differences in the rate of changes in metabolites reflecting the cerebral energy state during complete ischemia[82,102] or following MCA occlusion.[81,139] Second, a "stroke lesion" can be considered to consist of a central core of densely ischemic tissue (the focus) and of perifocal areas with less dense ischemia.[13,35,142,144,147] The focus, usually encompassing the lateral part of the caudoputamen and the adjacent neocortex, represents tissues that depend heavily on perfusion from the occluded MCA by end-arterial branches. The periphery of the stroke lesion and the perifocal areas are perfused by the anterior cerebral (ACA) and posterior cerebral (PCA) arteries via collateral vessels, most of which are leptomeningeal. Clearly, in all but the most central parts of the lesion, perfusion depends on the adequacy of the collateral circulation. Focal tissues are usually doomed since they receive a blood supply too feeble to sustain the cells beyond the initial 15 to 30 minutes (rats) or 30 to 60 minutes (primates) of ischemia. In contrast, although the perifocal areas are at risk, they may remain viable for longer periods. With a good collateral supply, viability may be preserved for many hours. With time, though, the infarct usually grows in size because the perifocal tissues are recruited in the infarction process. Since this takes time, a therapeutic "window" should exist. This implies that perifocal tissues may be salvaged by reperfusion before time has run out or by pharmacological agents that prevent the perifocal tissues from being recruited in the infarction process. Such agents evidently must support cells at risk over a critical period. This period is critical for one of several reasons: because there is an initial mismatch between blood flow and metabolic demands, because inhibitory control over cell activity is temporarily offset, or because factors released from the mortally sick neighbors jeopardize the survival of cells in the border of the focus. As will be discussed below, such factors encompass EAA's, pro-oxidant iron, or free radicals. A likely cause of a better match is an improvement of collateral flow with time.[13]

It is unfortunate that a clear distinction has not previously been made between stroke (defined as discussed in the preceding paragraph) and global ischemia due to cardiac arrest or forebrain ischemia in rats and gerbils.[116,125] In the latter models, the ischemia is usually transient, dense, and short-lasting. I will develop the idea that its pathophysiology is different from that of stroke, explaining why drugs that lack effect on the cardiac-arrest type of ischemia may ameliorate lesions caused by stroke.[127] As a first approximation, we will assume that brief transient ischemia injures neurons

without causing major microcirculatory problems, while the penumbral zone of a stroke lesion suffers prolonged ischemia, predisposing to inflammatory reactions at the blood-endothelial cell interface.

Critical Flow Thresholds

Since autoregulation is impaired or lost in moderate to severe ischemia, CBF varies passively with the perfusion pressure.[141,151] This relationship has allowed investigators to successively reduce CBF and to assess critical flow thresholds for certain functions. Both experimental studies in baboons and cats[16,44] and clinical studies during endarterectomy[112,144] have shown that spontaneous and evoked electrical activity ceased when CBF fell below 16 to 18 ml · 100 gm^{-1} · min^{-1}. This degree of ischemia thus represents a threshold for loss of neuronal electrical function. It was subsequently shown that the corresponding threshold for loss of cellular ion homeostasis was lower, requiring a reduction of CBF to below 10 to 12 ml · 100 gm^{-1} · min^{-1}. This threshold was originally established by workers measuring extracellular K^+ concentration.[3,15] Following the demonstration of a coupled K^+ efflux/Ca^{++} uptake in anoxia,[100] Harris, *et al.*,[43] showed that K^+ was released from and Ca^{++} taken up by cells at this lower CBF threshold. Rapid efflux of K^+ and uptake of Ca^{++} represent a generalized collapse of membrane function. Thus, at this point cells also take up Na^+ and Cl^-, with osmotically obligated water.[40,41,49,61]

The concept of two principal flow thresholds, separated by an intermediate zone characterized by cessation of electrical activity of cells with preservation of their membrane potential, is relatively unambiguous, and the threshold flow rates quoted may apply to cats, monkeys, and man. In rats and gerbils, the thresholds are reportedly higher,[42,93,147] probably reflecting the higher neuronal packing density and, hence, the higher cerebral metabolic rate.[56,113] However, since the control blood flow rates are also higher, the percentage of reduction required to reach a certain threshold is probably relatively similar.

Gross perturbation of cellular energy homeostasis occurs at the same CBF values as are associated with dissipation of the gradients for physiological ions.[93,105] As will be discussed below, this also follows from the fact that energy failure and dissipation of ionic gradients are mutually reinforcing events. It also seems likely that the threshold for infarction is similar to that for energy failure/loss of membrane homeostasis. However, the threshold for infarction varies with the duration of the insult.[25,45,53,70,143] For example, in monkeys subjected to ischemic periods of 1 to 3 hours, the critical level for infarction is 10 to 12 ml · 100 gm^{-1} · min^{-1} but, if the ischemia is permanent, the threshold is somewhat higher, about 17 to 18 ml · 100 gm^{-1} · min^{-1}. In rats, the corresponding threshold is reported to be about three times as high.[51]

Higher CBF thresholds have been reported for other indices of a perturbed cellular metabolism. For example, extra- and intracellular acidosis is observed at CBF values distinctly higher than those associated with energy failure/loss of ion homeostasis.[3,42,79,92,93] These results are in line with previous studies of progressive hypoxia and ischemia, demonstrating that lactate accumulates in the tissue at far higher arterial pO_2 values or mean arterial blood pressures than those associated with gross energy failure.[128,129] Furthermore, edema develops at CBF values exceeding those causing overt disruption of ion homeostasis.[52,140] Finally, the ischemic threshold for protein synthesis is even higher, suggesting that this function is extraordinarily sensitive to a reduction in CBF.[84,159]

The Concept of an Ischemic Penumbra

The finding of separate thresholds for cessation of electrical signals and for loss of ion homeostasis led to the concept of an ischemic penumbra, that is, of perifocal tissues containing electrically inexcitable but essentially viable cells.[2,39] It was even thought that such "sleeping beauties" could be viable for long periods and that they could be made to function by revascularization of the tissue.

The difficulty has been to delineate physically a penumbral zone, defined as an area with reduced CBF containing functionally depressed but viable cells with maintained ion homeostasis. For example, although Strong, *et al.*,[135,136] could characterize a perifocal zone with lowered CBF and depressed electroencephalographic (EEG) readings, extracellular K^+ was moderately increased, and scattered neuronal necrosis occurred within 2 hours. This led to a debate on the existence of the "sleeping beauties."[134] It was tempting to argue that, when one moves from the densely ischemic MCA focus toward the well-perfused areas irrigated by the ACA and PCA, flow is gradually increased from very low to normal values, and cell damage is gradually reduced, progressing from infarction to dense neuronal necrosis, sparse neuronal necrosis, and normal tissue. However, even this concept has received poor support. Thus, acute CBF measurements in MCA-occluded animals demonstrate a very sharp transition between poorly and normally perfused tissues.[13,97,147] Furthermore, in man and in subhuman primates, there is a very sharp transition between infarcted and normal tissue, with little evidence of a perifocal zone containing scattered necrotic neurons.[17,98] The zone is larger in cats[83] and rats,[94] but even in these species it is narrow unless the animals are made hypoglycemic prior to MCA occlusion.[96] One may argue from such results that, if drugs ameliorate ischemic damage by protecting cells in the penumbra, tissue salvage would indeed be modest. It has been pointed out that, at least in the rat, there is a more gradual transitional zone if one views CBF distribution in the sagittal plane.[13] However, the implication of these observations for human stroke is not obvious.

The penumbra concept, as discussed, has been useful in focusing attention on cells suspended in a twilight

state somewhere between survival and overt energy failure, with loss of basic membrane functions. However, it runs the risk of restricting the discussion to a special case, rather than describing the general one. The final lesion resulting from MCA occlusion usually consists of two parts: a central focus which always becomes infarcted unless reperfusion is quickly achieved, and a perifocal area which normally becomes part of the final infarct but which may be prevented from doing so, not only by reperfusion but also by drugs. Pharmacologically, we may thus define the penumbra as that part of an infarct which is potentially salvageable. This is a useful definition since it directs attention to tissues at risk. Such tissues are not necessarily electrically silent, nor do they necessarily have a completely preserved energy or ion homeostasis. However, they are vulnerable because they receive a subnormal blood supply, and also possibly because they live close to mortally sick neighbors. Defined in this way, the penumbra may well have a markedly reduced blood flow, albeit higher than in the focus. In the usual setting, it constitutes the periphery of the distribution territory of the occluded artery (the MCA), an area which would become nourished by collateral vessels emanating from the ACA and PCA, provided it survives the initial, critical period.

How, then, is the penumbra recruited in the infarction process? Two alternative ways of invasion seem feasible: 1) the infarction process spreads at the border in a relentless march that does not stop until well-perfused tissue is encountered; or 2) islands of necrosis develop in the underperfused tissues which, with time, coalesce to yield an extension of the infarct. The question arises: what could lead to the death of cells in the marginally underperfused perifocal tissues? Furthermore, how can drugs ameliorate damage to cells in the penumbra?

Figure 1 summarizes events in focal and perifocal tissues, events that may influence the fate of the underperfused tissue. It suggests that a CBF lower than 40% to 50% of control values puts the tissue at risk. This value is chosen because a reduction of CBF to that level is achieved by brisk hyperventilation, a condition that is not known to damage brain cells.[113] Figure 1 emphasizes that the factors that can jeopardize the survival of cells in the penumbra zone include acidosis, edema, transients of K^+ efflux/Ca^{++} influx, and depressed protein synthesis.

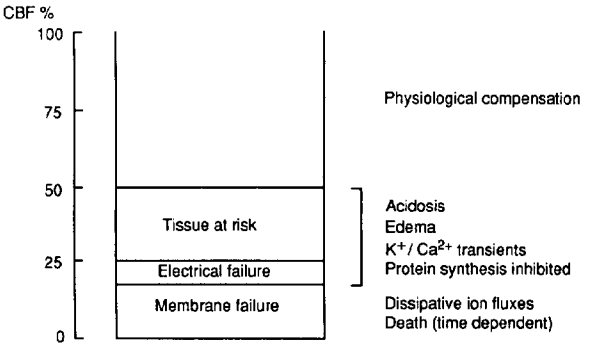

FIG. 1. Schematic diagram illustrating cerebral blood flow (CBF) thresholds for cell dysfunction or death in ischemia. The diagram shows that the threshold of membrane failure, with loss of ionic homeostasis, is also the threshold of cell death; however, cell death only occurs when ion homeostasis has been lost for some time. Electrically quiescent cells with a preserved ion homeostasis are conventionally considered to constitute the penumbra. The diagram suggests that any perifocal tissue with a CBF of less than 50% of control values is at risk and that several factors can jeopardize the survival of cells in this extended penumbra. (Diagram modified from that in Astrup J, Symon L, Branston NM, et al: Cortical evoked potential and extracellular K^+ and H^+ at critical levels of brain ischemia. *Stroke* 8:51–57, 1977.)

Reperfusion and Brain Damage

Experiments with complete ischemia in cats and monkeys have shown that, provided measures are taken to optimize reperfusion, short-term recovery of electrical and metabolic functions is possible even after such long ischemic periods as 60 minutes.[47,48] I suggest that this is possible only because these optimizing procedures (mainly hemodilution with hyperosmotic agents and an abrupt rise in perfusion pressure prior to release of vessel occlusion) prevent adhesion of neutrophils/thrombocytes to the endothelial cell wall, thereby preventing or reducing an inflammatory reaction that causes either "no-reflow" or a secondary shutdown of capillary circulation. However, under ordinary circumstances or following dense ischemia of the global or forebrain type, there is usually only a brief revival time, defined as the maximum period of ischemia that can be sustained without permanent neurological deficit or structural damage. Since flow is better preserved following MCA occlusion, the revival times are longer. However, they vary between regions since focal tissues can have flow rates below 10% of control values and penumbral tissues may be only moderately underperfused. Predictably, very early reperfusion can prevent damage from occurring altogether, while a further delay could salvage penumbral tissues and leave the focus infarcted. Two questions arise. First, is the "reperfusion penumbra" identical to the "pharmacological penumbra?" Second, what are the actual revival times?

The classic studies of Sundt, *et al.*,[138,139] demonstrated that MCA occlusion periods of up to 6 hours in cats and up to 1 hour in squirrel monkeys did not necessarily lead to infarction. Subsequent studies more closely defined the revival times and the nature of the lesions observed following relatively brief occlusion periods. Crowell, *et al.*,[24] studied *Macaca mulatta*, emphasizing that MCA occlusion in that species regularly causes nonfatal hemiparesis mimicking human stroke. The lesions also resembled those observed in cats. The authors occluded the MCA in anesthetized subjects for 1 to 24 hours and determined that ischemia from this cause requires 4 to 8 hours of evolution to yield the maximum extent of infarction.[23,24]

A subsequent study by DeGirolami, *et al.*,[25] on *Ma-*

caca mulatta and *fascicularis* examined the reperfusion window in even greater detail.[53,70] The information conveyed by these studies is the following: First, 15 to 18 minutes of MCA occlusion is tolerated without histological lesions (or neurological deficit). Second, an occlusion period of 30 minutes usually yields either no lesion or microscopic lesions confined to the subcortical gray matter. Third, although a 60-minute occlusion period may be tolerated with no damage or with only selective neuronal necrosis, macroscopic lesions are occasionally observed. With longer periods, the severity of the lesions increases. In general, animals with 4 to 8 hours of occlusion or longer had single confluent infarcts involving both deep and superficial structures, while those with occlusion periods of 4 hours or less showed either multiple nonconfluent infarctions involving deep structures only or multiple foci of selective neuronal necrosis, which often had a perivascular localization.

Important pieces of information arise from these studies.[25,53,70] First, at least in the monkey, an MCA occlusion that lasts for 30 minutes or longer often produces some tissue damage, and occlusion periods of 60 minutes or longer frequently create infarction. However, they also show that reperfusion within 4 to 8 hours can be expected to ameliorate the final lesion. Second, with relatively brief occlusion periods, the (micro)-infarcts are often multiple, suggesting that the final infarct represents the coalescence of multiple foci of cell necrosis. Third, rather than following the pattern of selective neuronal necrosis known from studies of global ischemia, the neuronal lesions observed after MCA occlusion often have a pericapillary and perivenular localization, suggesting that the pathophysiology of the microinfarcts encompasses microcirculatory disturbances. Interestingly, such a vascular pattern of tissue necrosis has also been seen following 10 minutes of forebrain ischemia in rats in which the head temperature was inadvertently raised above 37°C.[132]

The data obtained in the monkey studies have been confirmed and extended by more recent experiments in cats[154] and rats.[56,76,77] Results on cats seem similar to those reported for the monkey, while those obtained on rats suggest a smaller window. The latter results justify scrutiny, since the rat is now extensively used in studies of pharmacological protection or treatment. Two major models are used in that species. In one, a tandem occlusion of the common carotid artery and of the distal part of the MCA is used in spontaneously hypertensive rats, yielding neocortical infarction without affecting the caudoputamen.[51,56] Studies based on this model have shown that 1 hour of occlusion yields no or only a small infarction, while an increase in occlusion time from 3 to 4 hours induces infarction of maximum size.[56]

These results are not directly comparable to those obtained in cats or monkeys, since the caudoputamen is not affected. The other major model described for rats is that of Tamura, *et al.*,[144] In that model, the lateral part of the caudoputamen is densely ischemic and therefore prone to develop early damage. In support of this contention, early results obtained in rats suggested that 30 minutes of MCA occlusion may yield minor infarction.[95] With the advent of reproducible reperfusion models,[58,69,76,90] revival times could be studied in more detail. As shown in Fig. 2, with this model a 30-minute occlusion period in rats usually gives rise to an infarct localized to the caudoputamen and, less consistently, to selective neuronal necrosis in the neocortex.[77] The ischemic lesion is not ameliorated if reperfusion is delayed beyond the first 90 minutes. All this probably reflects a high cerebral metabolic rate and a relatively poor collateral blood supply in rats. This fact should be taken into account when attempts are made to extrapolate findings in rats to the clinical setting. As mentioned above, most subhuman primates show longer revival times and could therefore benefit from reperfusion even if delayed for 3 to 6 hours.

Reperfusion may be a double-edged sword. Thus, although return of flow within the revival times of the ischemic tissues usually salvages cells, at least in some extracerebral tissues restitution of oxygen supply may exacerbate damage, giving rise to what is commonly called "reperfusion damage."[60,75] It was observed decades ago[138] that reperfusion of an occluded MCA may be followed by massive tissue swelling and secondary compromise of circulation, and it was later established that reperfusion following 3 or 6 hours of MCA occlusion aggravates the edema, particularly the vasogenic component.[50] Such damage may be due to the resupply of water and osmotic equivalents (which exacerbates edema) or of oxygen (which triggers production of injurious free radicals).[29,59,119,124] It has been argued that reperfusion damage in the brain does not exist in the sense that reperfusion gives rise to a larger infarct than does permanent occlusion.[56] This may depend on the size of the infarct. At any rate, it is perhaps more profitable to discuss the amelioration of damage due to medications given just after reperfusion is achieved. Clearly, if such drugs are efficacious, they interfere in one way or the other with injurious factors operating in the reperfusion period.

Pathophysiology

The concept of cellular energy failure is central in ischemia research. Very likely, cell necrosis is secondary to a deranged or perturbed energy metabolism.[120,122] This is because failure of synthesis of adenosine triphosphate (ATP) and other nucleoside triphosphates (such as cytidine triphosphate and uridine triphosphate) threatens cell survival in three ways. First, in the absence of an adequate energy source, anaerobic glycolysis is stimulated, leading to intra- and extracellular acidosis. As will be discussed below, this is a potential threat to the viability or recovery of ischemic tissue. The second major way in which energy failure may jeopardize the survival of ischemic tissue is by disrupting ion homeostasis. Two components of this loss are in the

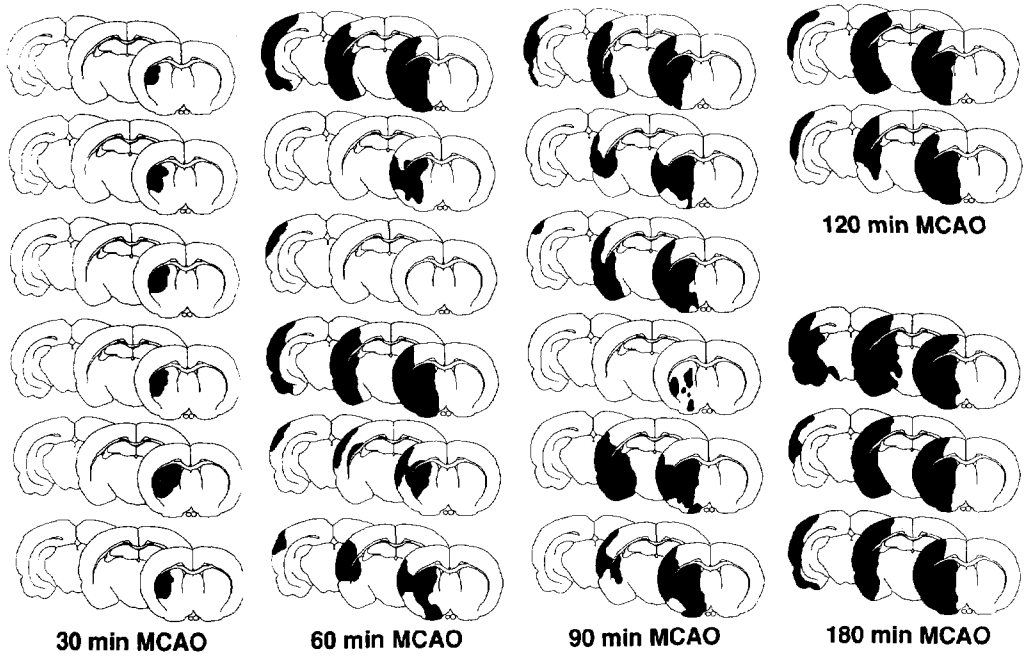

FIG. 2. Cross sections of rat brains illustrating the "reperfusion penumbra," that is, the effect of reperfusion on the final infarct resulting from transient middle cerebral artery occlusion (MCAO). The left artery was occluded for 30 to 180 minutes, and the resulting infarct evaluated after 7 days of recovery. The *large black areas* represent infarcted tissue, while *black dots* denote areas of selective neuronal necrosis. (Data drawn from Memezawa H, Smith ML, Siesjö BK: Penumbral tissues salvaged by reperfusion following middle cerebral artery occlusion in rats. *Stroke 23*:552–559, 1992.)

focus of interest: the cellular influx of Na^+ and Cl^- with osmotically obligated water and the influx of calcium, a major pathogenetic event. The third consequence of energy failure is that the structural integrity of the cell is threatened. Since high-energy phosphates are required to support resynthesis of macromolecules and macromolecular assemblies following their spontaneous or enzyme-catalyzed degradation, loss of ATP leads to the breakdown of cell structure. In this area, research is usually focused on protein and lipid constituents. For example, it has been emphasized that ATP failure leads to proteolytic cleavage of components of the cytoskeleton, such as microfilaments and microtubuli, and their anchorage to the cell membrane.[66] Another effect of ATP failure is the degradation of phospholipids, with the accumulation of breakdown products such as lysophospholipids, diacylglycerides (DAG's), and free fatty acids, including arachidonic acid.[4–6,119,120,158] However, since many of the degradation enzymes involved are activated by calcium, the breakdown of structure is due both to a loss of ATP and to a rise in calcium concentration.

Linkage of Energy Metabolism to Ion Flux

The coupling of energy metabolism and ion homeostasis is exemplified by Fig. 3, which illustrates both passive and energy-driven ion flux across normally polarized cell membranes. An unequal distribution of physiological ions and a membrane potential of about 60 mV (the inside being negative) are created by "uphill" ion transport, which directly or indirectly depends on ATP. Such active transport normally balances the passive ("downhill") flux of ions via membrane channels. This is driven by both the chemical gradients and the membrane potential.

Energy harnessed by ATP can be used to drive directly the extrusion of $3Na^+$ in exchange for $2K^+$, and extrusion of one Ca^{++} in exchange for $2H^+$. In this way, the extracellular concentrations of Na^+ and Ca^{++} become about 10- and 10,000-fold higher than the intracellular levels, respectively, while the intracellular concentration of K^+ is about 40-fold higher than the extracellular level. Since the membrane is quite permeable to K^+, the distribution of K^+ determines to a large extent the membrane potential. However, since the permeability to Na^+ is not negligible, the membrane potential is less negative than the equilibrium potential for K^+. This means that K^+ normally tends to diffuse out of cells, driven by a concentration gradient (see below).

The Na^+ gradient created by ATP-driven transport is a source of energy that can be utilized either to drive Na^+ back into the cell via a conductance channel or in exchange for another ion such as Ca^{++} or H^+. The $3Na^+/Ca^{++}$ exchange mechanism ("antiporter") aids in the efflux of Ca^{++} from cells, while the Na^+/H^+ antiporter forms a ubiquitous mechanism for extrusion of H^+. The $3Na^+/Ca^{++}$ exchanger is electrogenic; that is,

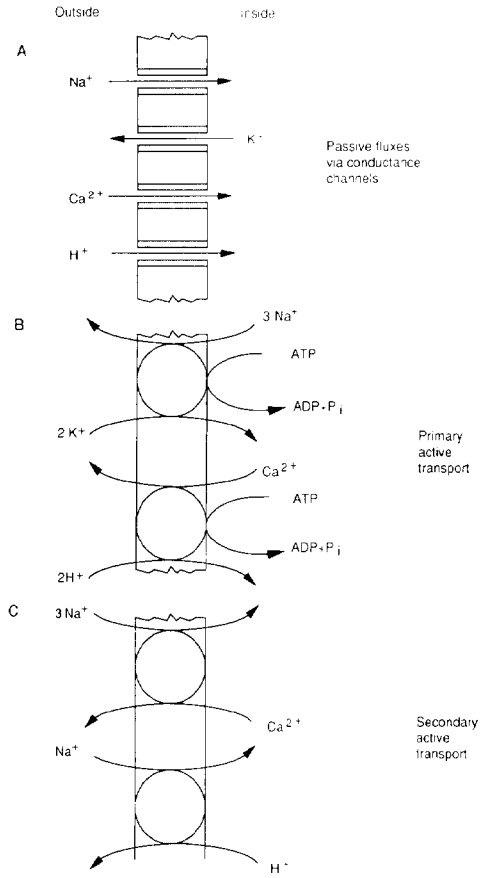

FIG. 3. Schematic diagram illustrating major mechanisms of passive and energy-dependent ion fluxes across polarized membranes in normal tissue. The passive fluxes are driven by the electrochemical gradients, while active fluxes directly or indirectly depend on energy furnished by adenosine triphosphate (ATP). ADP = adenosine diphosphate; P_i = intracellular phosphorus.

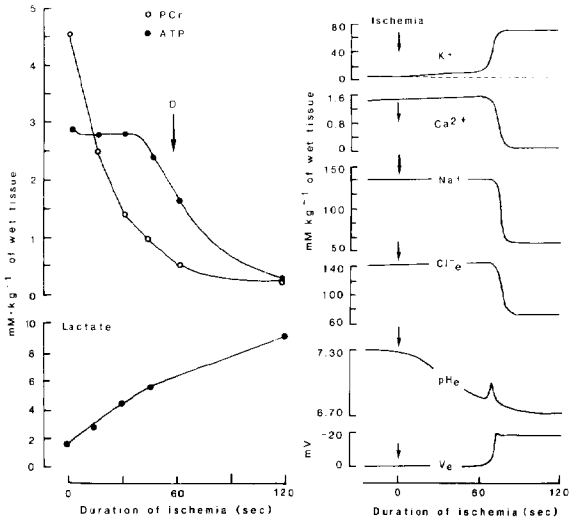

FIG. 4. Graphs depicting the coupling of energy failure and ionic fluxes during ischemia. *Left:* Tissue concentrations of phosphocreatine (PCr), adenosine triphosphate (ATP), and lactate during the first 120 seconds of complete ischemia. D = depolarization. *Right:* Changes in extracellular concentrations of K^+, Ca^{++}, Na^+, Cl^- (Cl^-_e), and pH (pH_e) and in DC potential (V_e). Note that K^+ increases when the ATP concentration is greater than 90% of control values, and that ion homeostasis is lost when ATP has decreased to below 50% of control values.

it carries both ions and electrical charge across the membrane. This means that the membrane potential is one of the determinants of $3Na^+/Ca^{++}$ exchange. In this context, I wish to remind readers that the Na^+ gradient, created by the $3Na^+/2K^+$ adenosine triphosphatase (ATPase), can be used for other transport tasks. To take only one example, cotransport of Na^+ and amino acids such as glutamate forms an important reuptake mechanism, terminating transmitter action.

What determines the distribution of negative ions such as Cl^- and HCO_3^-? Some processes tend to achieve uphill transport of Cl^-. For example, since the Na^+/K^+-ATPase is electrogenic, expelling $3Na^+$ in exchange for $2K^+$, the carrier can only operate at useful rates if the excess charge is balanced by efflux of an anion such as Cl^-. Export of one extra Na^+ together with a Cl^- means that water must follow passively. In other words, the Na^+/K^+-ATPase functions as a water pump that could balance the leakage flux of Na^+ and Cl^- (see below). Leakage flux for H^+ and HCO_3^- could occur via conductance channels or via a Cl^-/HCO_3^- exchanger, which mediates loss of HCO_3^- (gain of H^+) and uphill transport of Cl^-. This means that mechanisms exist for Cl^- transport. However, under normal circumstances, the membrane permeability to Cl^- is so high that Cl^- is distributed passively in the electrical field, meaning that the extra- to intracellular concentration ratio is about 10:1.

Clearly, under normal circumstances the active and passive ion flux is balanced. Dissipation of the normal ion gradients (loss of ion homeostasis) can occur because the pumps fail, because the leaks are enhanced, or both. In normal polarized cells, leakages flow in the directions shown in Fig. 3. If the membrane depolarizes, the electrical force is attenuated, and the direction of the flux is mostly determined by the concentration gradients. This means that Na^+ and Ca^{++} will still enter cells, but K^+ will now rapidly leave and Cl^- enter.

Figure 4 correlates changes in cerebral energy state during complete ischemia and dissipative (downhill) ion fluxes, and shows that ion fluxes occur in two major phases.[40,118,150] During the first phase, the extracellular K^+ level slowly rises. There is also a fall in extracellular pH; however, this is not necessarily due to membrane flux of H^+ via a cation channel since lactic acid may leave the cell by nonionic diffusion and, besides, accumulation of CO_2 would acidify extracellular fluid (ECF). Since some contraction of the ECF volume may occur in this early phase, the changes in the extracellular concentrations cannot be rigorously interpreted. However, it seems likely that a K^+ conductance is activated.

If this is so and if the positively charged K^+ leaves the cell, the membranes containing such K^+ channels will tend to hyperpolarize (see below). We also observe that extracellular Ca^{++} does not decrease.

During the second phase, K^+ rapidly leaves cells, while Na^+, Cl^-, and Ca^{++} are absorbed. These changes, which occur simultaneously with the development of a DC electrical potential shift and a transient alkaline shift in the pH record, suggest that a nonspecific cation conductance is activated. The transient alkaline shift may arise because H^+ enters cells with an upheld membrane potential and leaves again when the membrane depolarizes.[104,117] Alternatively, HCO_3^- moves in the opposite direction via activated Cl^- channels known to be permeable to HCO_3^-.[54]

The changes in phosphocreatine (PCr) and ATP can be interpreted as follows. Anoxia halts ATP synthesis but not ATP utilization. This leads to ATP hydrolysis via the reaction ATP + HOH → adenosine diphosphate (ADP) + intracellular phosphorus + H^+. However, since ADP concentration rises, the creatine kinase reaction (PCr + ADP + H^+ → creatinine + ATP) maintains ATP at about 95% of control until the PCr stores are depleted, then ATP concentration falls. Glucolysis with production of lactate yields some additional ATP but this feeble source of ATP cannot prevent rapid energy failure. A correlation between changes in energy balance and ion homeostasis shows two things: first, the initial rise in extracellular K^+ level occurs in the absence of major energy failure and, second, depolarization with rapid dissipative ion flux is preceded by extensive energy failure.

The patterns of ion flux shown in Fig. 4 are pathogenically important because they illustrate two putative mechanisms of cell damage.[109] One is related to the influx of Na^+ and Cl^-. Since this must occur with osmotically obligated water, it leads to cell swelling (osmolytic damage). The other mechanism is related to the influx of calcium and to the resulting increase in cytosolic calcium concentration (calcium-induced damage). The first mechanism may not be important in complete ischemia, simply because the source of Na^+ and Cl^- is restricted, but it may be more deleterious if some flow persists, as with stroke.

The results shown in Fig. 4 should not be construed to demonstrate that the intracellular Ca^{++} concentration does not rise until the ion homeostasis is suddenly lost (after 60 to 70 seconds). Thus, surface fluorescence measurements[148] and direct intracellular microelectrode measurements[130] suggest that the intracellular Ca^{++} concentration rises shortly after induction of anoxia and that phosphorylase *b* to phosphorylase *a* conversion, a calcium-dependent event, occurs within 15 seconds following induction of ischemia.[30] Obviously, we must consider the possibility that calcium is released from intracellular stores before it enters from the ECF. This would be an important event since a rise in intracellular Ca^{++} concentration could increase membrane conductance to K^+, Cl^-, and other ions (see below).

Increased membrane permeability to ions is probably a major cause of a perturbed ion homeostasis. Since loss of K^+ as well as uptake of Na^+ and Ca^{++} serve as triggers for ATP consumption, pump failure and enhanced leakage must be mutually reinforcing events in ischemia. In other words, if membrane permeability to ions is increased, the ATP produced could be wasted during a futile cycling of ions. This contention is supported by the fact that, in other conditions, depletion of ATP stores may not even be the trigger of the loss of ion homeostasis. In hypoglycemia, rapid downhill flux of K^+ and Ca^{++} precedes energy failure.[157] Ion fluxes of the type illustrated in Fig. 4 may even occur in normal tissues. This is observed in spreading depression elicited by electrical stimulation, a stab wound, or local application of K^+ or glutamate.[41,61,71,89,150] The difference between spreading depression and ischemia (or hypoglycemia) is that, in the former, the ion fluxes are spontaneously reversible.

The concept of a deranged pump/leak relationship for ions as a trigger to ischemic tissue damage has important implications for focal ischemic lesions. Thus, it is no longer tenable to regard the metabolic defect exclusively in terms of pump failure secondary to depletion of ATP stores. It is equally necessary to consider the ion leak. Thus, if local depolarization in penumbral tissues leads to activation of ion conductances, this may trigger overt energy failure and that, in turn, may prolong the ionic transient. Clearly, if the leak can be tightened, the effects of the pump failure with its metabolic consequences may be dramatically reduced. This argument applies to the pump/leak relationship for calcium, and to the coupling between the severity and duration of a rise in intracellular Ca^{++} and cell necrosis. Clearly, we are justified in scrutinizing the leak pathways, particularly those mediating rises in intracellular Ca^{++}, and in briefly reviewing the mechanisms that regulate intracellular Ca^{++}.

Pathways of Dissipative Ion Fluxes

Figure 3 illustrates pathways of dissipative (downhill) ion fluxes as simple membrane pores, allowing positively or negatively charged ions to pass through the membrane. Figure 5 depicts schematically an excitatory nerve ending and helps to define and characterize the channels involved (see two useful reviews on ion channels[107,108]). We envision that, when presynaptic depolarization due to successive activation of Na^+ conductances (channels) invades the nerve terminal, a voltage-sensitive calcium channel (VSCC) is opened, allowing Ca^{++} to enter and trigger release of an EAA such as glutamate. Presumably, the VSCC involved is of the N type, while the T and L types could be localized mainly to postsynaptic membranes in dendrites and soma.[37,85,146,155] It should be emphasized, however, that release of EAA's may be unrelated to Ca^{++} influx. This is because reuptake of EAA's following their release is due to an electrogenic $2Na^+$/glutamate$^-$ cotransporter which is driven by both the Na^+ gradient and the membrane

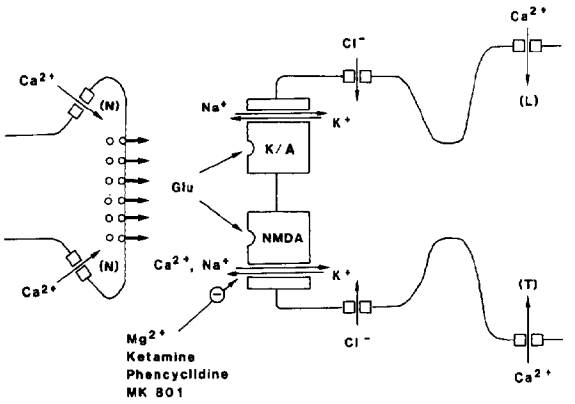

FIG. 5. Schematic diagram illustrating pre- and postsynaptic ion channels, with emphasis on voltage-sensitive (VSCC) and agonist-operated calcium channels. Presynaptically, the VSCC involved in transmitter release are assumed to be of the N type, while the L and T types are assumed to be localized to dendrites. Release of the excitatory transmitter glutamate (Glu) is shown to activate two types of receptors, selectively sensitive to kainate/amino-3-hydroxy-5-methyl-4-isoazole propionic acid (AMPA) (K/A) and to N-methyl-D-aspartate (NMDA), respectively. The K/A receptor gates a channel that is permeable to monovalent cations (Na^+, K^+, and H^+), while the NMDA-gated channel is permeable to Ca^{++} as well. Normally, this channel is blocked by Mg^{++}, but this block is voltage-dependent. The AMPA receptor activation and Na^+ influx relieve the block, allowing Ca^{++} to enter. Depolarization also allows Ca^{++} to enter via VSCC of the L and T types. Inhibition is assumed to be mediated by activation of K^+ and Cl^- conductances. (Reproduced from Siesjö B: Calcium, excitotoxins, and brain damage. *News Physiol Sci* 5:120–125, 1990, with permission.)

potential.[55,99] If the Na^+ gradient is dissipated and the membrane is depolarized, the symporter could reverse its mode of action and catalyze release of EAA's. Very likely, this occurs in ischemia.

Postsynaptic membranes probably contain Na^+ conductances allowing "fast" excitation. However, the predominant mechanism for excitation in the brain encompasses ion channels gated by glutamate receptors.[22,28,68,72,152,153] These are now usually classified into five categories depending on the agonist that most efficiently activates them: high- and low-affinity kainate, amino-3-hydroxy-5-methyl-4-isoazole propionic acid (AMPA), N-methyl-D-aspartate (NMDA), and quisqualate receptors. Since only the first four are ionotropic in the sense that they act as ion channel gates and since K and AMPA receptors could be linked to the same type of ion channel, the discussion will be confined to AMPA and NMDA receptors. As Fig. 5 shows, we envision that these are predominantly localized to dendritic spines (and distal dendrites).

Glutamate is a mixed agonist. When released from presynaptic endings, it will active both AMPA and NMDA receptors. The AMPA receptor is linked to a channel providing a nonselective conductance mechanism for the monovalent cations Na^+ and K^+, and probably also H^+. By allowing Na^+ to enter, the opening of this channel leads to depolarization. The NMDA subtype of glutamate receptor gates a channel permeable to both monovalent cations and calcium. This channel is normally blocked by Mg^{++} but, since this block is voltage-dependent, it is relieved when the membrane depolarizes.[73,103] By causing depolarization, Na^+ influx via the AMPA receptor-linked channel thus sets the stage for calcium influx via the NMDA channel. Clearly, depolarization caused by Na^+ (and Ca^{++}) influx also must allow entry of Ca^{++} through VSCC's of the L and T types. An additional pathway of entry is the $3Na^+/Ca^{++}$ exchanger. Thus, reduction of the Na^+ gradient and/or depolarization can reverse the direction of the exchanger, which then shuttles Ca^{++} inward.[27,111,126] It has been speculated that the reduction in Na^+ gradient required to reverse $3Na^+/Ca^{++}$ exchange can be the result of intracellular acidosis stimulating Na^+/H^+ exchange.[64] It is also conceivable that massive depolarization, or the rise in intracellular Ca^{++}, activates nonspecific cation conductances separate from those gated by glutamate receptors (a discussion and further literature review are presented elsewhere[125,126]). Obviously, activation of glutamate receptors can trigger an avalanche type of reaction in which multiple calcium conductances are activated. We also recognize that the release of EAA's and the activation of ionotropic glutamate receptors provide a likely mechanism for an increase in membrane permeability to several ions.

The excitatory events, including a release of glutamate and glutamate activation of receptors which cause Na^+ influx and depolarization, followed by calcium influx via multiple channels, form part of a physiological signaling system that utilizes calcium as a second messenger. Both termination of the excitatory events and tonic control are achieved by inhibitory events and by reuptake of the EAA's released.[19,107,108] Inhibition is mainly due to ion fluxes which tend to hyperpolarize membranes. In general, activation of K^+ or Cl^- conductances leads to inhibition. This is because efflux of K^+ hyperpolarizes membranes, while an increase in Cl^- conductance tends to clamp the membrane potential at close to normal values. Inhibition may also be exerted by transmitters or other messengers that modulate calcium channels. Important control of excitatory events, both pre- and postsynaptic, is exerted by γ-aminobutyric acid (GABA)-ergic, purinergic, and noradrenergic tone. This tonic control is secondary to activation of receptors linked to channels (exemplified by GABA) or to modulation of calcium channels (by adenosine or noradrenaline). Evidence accumulates that agonists acting on noradrenergic and purinergic receptors or those activating GABA receptors may reduce damage; however, definitive data have not emerged on their usefulness in focal ischemia.

It now seems likely that an important inhibitory control is exerted by activation of K^+ conductances.[9,40,63,87] It was stated above that, once a K^+ channel is opened and the positively charged K^+ ion leaves the cells, the membrane hyperpolarizes. Two types of K^+

channels of interest to the present discussion are those activated by a rise in intracellular Ca^{++} level and by a fall in ATP/ADP ratio. The former type may be responsible for hyperpolarization of postsynaptic membranes and synaptic block, perhaps explaining the early loss of EEG activity during anoxia.[26,40] Channels regulated by the ATP/ADP ratio, now assumed to be mainly presynaptic, could be important modulators of transmitter release.[8]

In summary, a rise in the extracellular K^+ concentration, depolarization, and presynaptic calcium influx trigger a cascade of reactions, including release of glutamate and other transmitters and activation of cation and anion conductances. In the absence of an adequate energy source, the Na^+ gradient is reduced or dissipated, retarding or preventing reuptake of transmitters. These events and the associated rise in intracellular Ca^{++} level may keep both cation and anion conductances activated. In other words, the membranes become so leaky that any ATP formed is wasted by futile cycling of ions. Clearly, drugs that may prevent such a loss of ion homeostasis or that may reverse the changes once elicited include those that reduce presynaptic depolarization, calcium influx, and transmitter release, and those that reduce activation of glutamate receptors by their agonists, block VSCC's, or enhance the conductance of Cl^- or K^+.

Cellular Calcium Homeostasis

Enhanced influx and reduced extrusion of calcium is but one aspect of a perturbed cellular calcium homeostasis. Thus, other factors contributing to a rise in intracellular Ca^{++} are accelerated calcium release from intracellular stores, compromised sequestration, and reduced binding. Figure 6 gives an overview of the mechanisms involved in regulating Ca^{++} (for further details, see references[12,18,74,115,121]).

As with other cells, neurons release calcium from intracellular stores in response to an appropriate stimulus.[10,11,36,88,91] This stimulus may be an agonist activating a surface receptor coupled to phospholipase C (PLC), which then splits phosphatidylinositol biphosphate to DAG and inositol triphosphate, the latter acting to release calcium from the endoplasmic reticulum. Acetylcholine and some peptides are the classic agonists at such receptors. However, it is now known that, at least in some cells glutamate, acting on the quisqualate type of metabotropic receptors, initiates this cascade of events.[101,131,137] The stimulus for Ca^{++} release may also be a rise in intracellular Ca^{++} concentration by other means, triggering calcium-induced Ca^{++} release, an amplification mechanism that may be involved in Ca^{++} oscillations. The effect of calcium on this amplification mechanism is duplicated by caffeine, while ryanodine and dantrolene reduce intracellular Ca^{++} release.[31,36]

Most of the calcium that enters cells via VSCC's and agonist-operated calcium channels or that is released intracellularly, is buffered by being bound by a host of high- and low-molecular-weight compounds or sequestered into organelles. The most important buffering sites are provided by effector proteins such as calmodulin and by specialized binding proteins such as calcibindin.[74] Binding of Ca^{++} to negative groups is similar to the buffering of H^+ which, like Ca^{++}, is maintained in the cell at a concentration of about 10^{-7} M. In fact, Ca^{++} and H^+ may compete for occupancy at the same buffer sites. Thus, acidosis is likely to release calcium from some of its binding sites.

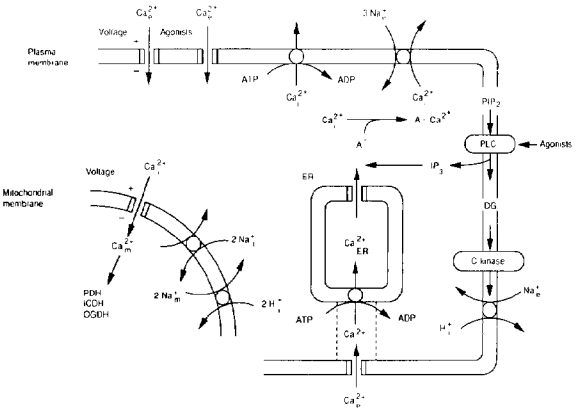

FIG. 6. Schematic diagram illustrating major components of cellular calcium homeostasis. The cytosolic intracellular calcium concentration (Ca^{++}_i) represents the balance between influx, efflux, binding, and sequestration. Normally, influx of calcium occurs via voltage-sensitive and agonist-operated calcium channels, and possibly via channels associated with the endoplasmic reticulum (ER). Efflux is due to an adenosine triphosphate (ATP)-driven transporter and to electrogenic $3Na^+/Ca^{++}$ exchange. A similar pump/leak relationship is envisioned to exist at the level of intracellular membranes. Efflux is depicted to occur in response to formation of inositol triphosphate (IP_3), while resequestration occurs at the expense of ATP. Interaction of agonists with receptors coupled to phospholipase C (PLC) is shown to give rise to both IP_3 and diglycerides (DG), the latter activating protein kinase C which modulates membrane-bound proteins such as the Na^+/H^+ antiporter. ADP = adenosine diphosphate; PIP_2 = phosphatidylinositol biphosphate; PDH, ICDH, OGDH = dehydrogenases. Subscript i = intracellular; subscript e = extracellular; subscript m = mitochondrial. (Reproduced from Siesjö BK, Bengtsson F: Calcium fluxes, calcium antagonists, and calcium-related pathology in brain ischemia, hypoglycemia, and spreading depression: a unifying hypothesis. *J Cereb Blood Flow Metab* 9:127–140, 1989, with permission.)

The uptake of calcium into the endoplasmic reticulum and similar sequestration sites requires ATP. Under normal circumstances, mitochondria do not take up calcium in appreciable amounts, but if the intracellular Ca^{++} concentration rises markedly, mitochondria may accumulate large amounts of calcium. The driving force is the potential across the inner mitochondrial membrane (the inside being negative). This potential is only upheld in the presence of O_2 or ATP.

Clearly, regulation of the intracellular Ca^{++} level represents the balance between calcium pumps and calcium leaks, not only at the level of the plasma

membrane but also intracellularly. Figure 6 demonstrates that a rise in intracellular Ca^{++} concentration may occur before calcium has entered from the ECF, possibly explaining the early activation of Ca^{++}-regulated K^+ channels (see above). Obviously, all that is required is release of Ca^{++} from intracellular stores (for example, in response to activation of receptors coupled to PLC). We recognize the possibility of manipulating Ca^{++} release by drugs that act on receptors coupled to PLC, or on intracellular receptors that regulate Ca^{++} release.

Acidosis

The major cause of a reduction in intracellular and extracellular pH during ischemia is a mismatch between glycolysis and oxidative phosphorylation. Figure 7 illustrates the principal steps in the aerobic generation of ATP from the breakdown of glucose and in the way hypoxia/ischemia alters metabolism. Under aerobic conditions, glucose is broken down in a series of enzymatic steps to 2 M pyruvate. During these reactions, the oxidized form of nicotinamide-adenine dinucleotide is reduced and 2 M each of ADP and intracellular phosphorus are converted to 2 M of ATP. In the presence of oxygen, pyruvate is first metabolized by pyruvate dehydrogenase and then by a series of mitochondrial reactions to CO_2 (and H_2O) with the formation of 36 M of ATP. This is the maximum ATP yield. It may be less (for instance, if the mitochondria have to sequester Ca^{++} during intense activity). In other words, ATP production and Ca^{++} sequestration are alternative ways of harnessing the energy released during pyruvate oxidation.

Hypoxia/ischemia blocks or retards this sequence of events at the stage of pyruvate oxidation and leads to the reduction of pyruvate to lactate. This reaction, catalyzed by the pyruvate reductase part of the lactate dehydrogenase enzyme, does not produce H^+. However, if the 2ATP formed during glucosis is hydrolyzed to ADP and intracellular phosphorus (meaning that the ATP concentration stays constant), the net result of the anaerobic metabolism of 1 M of glucose is the production of 2 M each of lactate$^-$ and of H^+.[46] The energy disadvantage of this aerobic-anaerobic transition is that ATP production is reduced; in addition, the cell is acidified. Also, since mitochondria deprived of oxygen lose their ability to sequester calcium, any calcium entering the cell or released within the cell will raise the intracellular Ca^{++} level. We recognize the complex interplay between acidosis and changes in the intracellular Ca^{++} concentration: on one hand, a rise in the level will force partially oxygen-deprived mitochondria to take up calcium, further reducing their ability to produce ATP and enhancing the acidosis. On the other hand, at very low oxygen tensions (or ATP concentrations) mitochondria can no longer take up calcium, and the intracellular Ca^{++} level may rise to excessive levels. Furthermore, H^+ can then displace Ca^{++} from its binding sites, contributing to the rise in intracellular Ca^{++}.

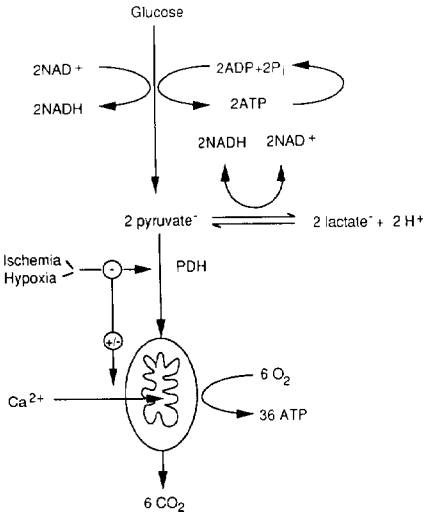

FIG. 7. Schematic diagram illustrating the aerobic/anaerobic metabolism of glucose. The oxidation of pyruvate to CO_2 (and H_2O) by pyruvate dehydrogenase (PDH) and citric acid cycle enzymes is retarded or blocked by oxygen deficiency, causing a reduction of pyruvate to lactate. If the adenosine triphosphate (ATP) formed during glycolysis is hydrolyzed (that is, if the ATP concentration stays constant), one molecule of H^+ is released for each molecule of lactate formed. If the mitochondria retain a membrane potential, they will sequester excess calcium entering the cell; however, if they are de-energized (with collapse of their membrane potential), they will release their calcium content. NAD = nicotinamide-adenine dinucleotide; NADH = reduced nicotinamide-adenine dinucleotide; ADP = adenosine diphosphate; P_i = intracellular phosphorus; PDH = pyruvate dehydrogenase.

What determines the severity of intra- and extracellular acidosis? With complete or near-complete ischemia where energy failure is at hand and glucose delivery is arrested, the amount of lactate accumulated depends on the preischemic glucose and glycogen content of the tissue.[67] Thus, the severity of the acidosis correlates to the preischemic plasma glucose concentration. With complete or near-complete ischemia of the global or forebrain types, intracellular pH falls to values of 6.2 to 6.4,[7,20,149] and it is further reduced in hyperglycemic animals.[14,21,133] In ischemia due to MCA occlusion, acidosis of comparable magnitude has only been observed in the low-flow focus of the lesion,[1,57,110] while there are many reports of much less severe degrees of acidosis.[38,78,79] Nakai, *et al.*,[92] nicely illustrated both the flow dependence of the reduction in intracellular pH and the aggravating effect of hyperglycemia. However, their data showed that, unless CBF fell below about 20 ml · 100 gm^{-1} · min^{-1}, the acidosis was relatively moderate. This leads to the question: what determines the severity of the lactic acidosis? One can discern two major factors: the availability of carbohydrate substrates and the degree of ATP failure. If glucose is readily available, as it is in hypoxia with maintained CBF, the amount of lactate accumulated is determined by the degree of energy failure, and not by the plasma glucose

concentration.[33] In ischemia, where flow is reduced, glucose supply can be limiting for lactic acid production; if it is, enhancement of glucose supply by hyperglycemia may exaggerate the acidosis. However, the other important determinant is the cellular ATP concentration. If ATP content is only marginally reduced, hyperglycemia is not likely to cause much further accumulation of lactate$^-$ plus H^+, but if it is very low, most of the glucose delivered will be anaerobically metabolized. Clearly, if we wish to reduce the acidosis, one strategy is to reduce plasma glucose concentration; another is to improve mitochondrial function and thereby to increase ATP production.

Conclusions

The main cause of cell dysfunction and cell death in ischemia is a failure of ATP synthesis. By itself, energy failure threatens cell viability by retarding or preventing resynthesis of macromolecules of key importance for cell structure and function. However, these threats are reinforced by the effects of energy failure on ion homeostasis and on acid-base balance. This is because dissipative ion fluxes and glycolytic production of lactate$^-$ plus H^+ lead to influx/intracellular release of calcium and to intra- and extracellular acidosis. Tertiary events encompass lipolysis, proteolysis, and inhibition of protein synthesis. These events lead to rapid cell death in the densely ischemic focus of a stroke lesion, and threaten the viability of cells in the less densely ischemic penumbra. The mechanisms involved and ways of ameliorating the damage are the subjects of the second part of this review.[123]

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