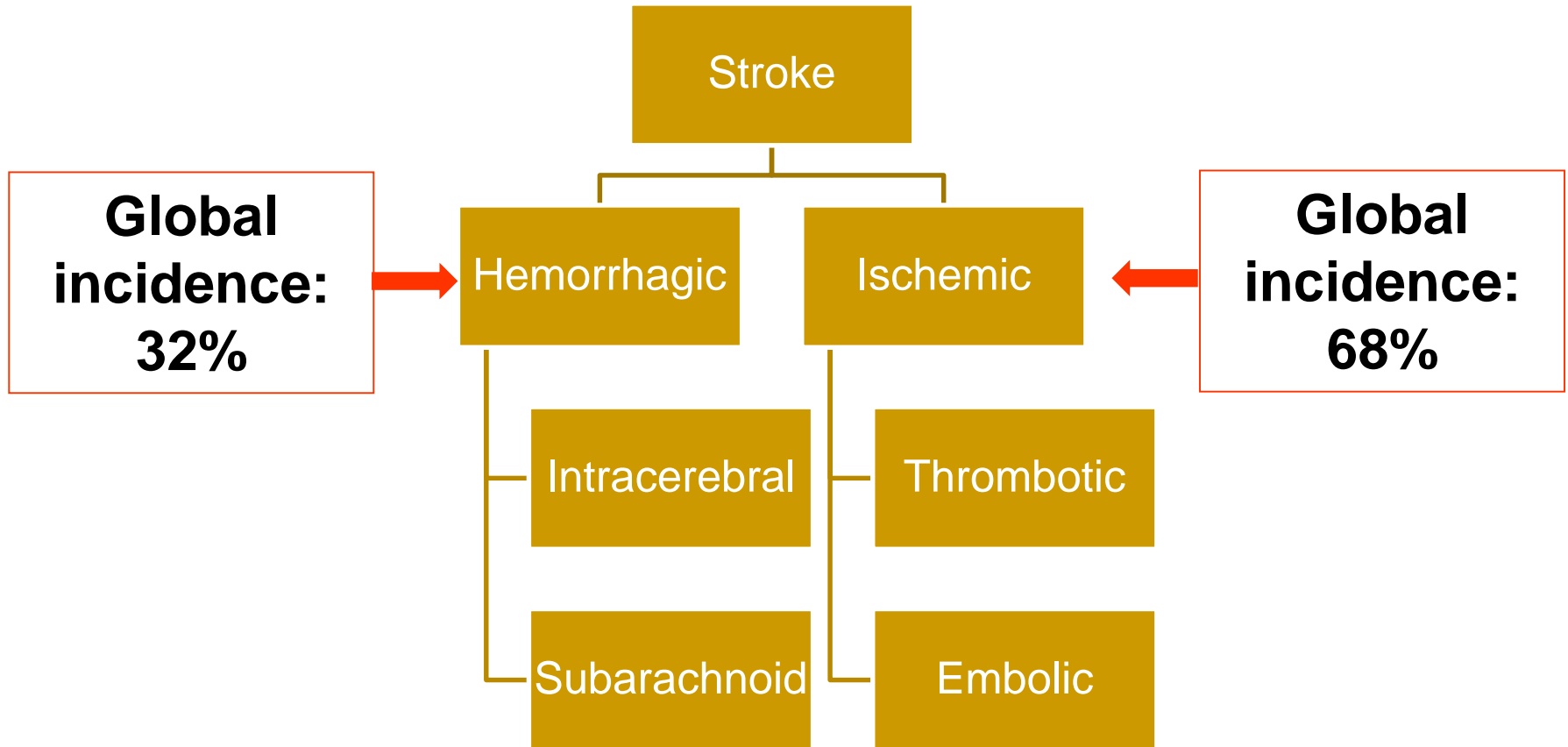

**Pathogenesis of Cerebral
Infarction
at
Cellular & Molecular Levels**

Objectives:

By the end of this lecture, the students should be able to:

- Identify the possible **cell death mechanisms** implicated in the pathogenesis of ischemic brain injury
 - Acquire the knowledge of the important role played by **oxidative stress and free radicals** in the pathogenesis of cerebral infarction
 - Understand the various factors involved in **ischemia-induced metabolic stress**
 - Identify the Neurochemical changes involved in cerebral ischemia
-

Cerebral Ischemia (Strokes) subtypes



Risk factors of strokes

- There are a number of risk factors for stroke:
 - Some increase the risk of one type of stroke (hemorrhagic or ischemic).
 - Some increase the risk of both types.
 - Occasionally, strokes occur in people who have no risk factors.

Continued



Risk factors of strokes

Ischemic stroke risk factors

Age older than 40 years
Heart disease
High blood pressure
Smoking
Diabetes
High blood cholesterol levels
Illegal drug use
Recent childbirth
Previous history of transient ischemic attack
Inactive lifestyle and lack of exercise
Obesity
Current or past history of blood clots
Family history of cardiac disease and/or stroke

Hemorrhagic stroke risk factors

High blood pressure
Smoking
Illegal drug use (especially cocaine and "crystal meth")
Use of warfarin or other blood thinning medicines

The cell death mechanisms
implicated in the pathogenesis of
ischemic brain injury

Cell death mechanisms in cerebral ischemia: Necrosis and Apoptosis

- **Necrosis** is commonly observed **early** after **severe** ischemic insults
- **Apoptosis** occurs with more **mild** insults and with **longer** survival periods
- The mechanism of cell death involves calcium-induced **calpain-mediated proteolysis** of brain tissue
- **Substrates for calpain include:**
Cytoskeletal proteins, Membrane proteins and Regulatory and signaling proteins

Biochemical Responses to Ischemic Brain Injury

Oxidative stress

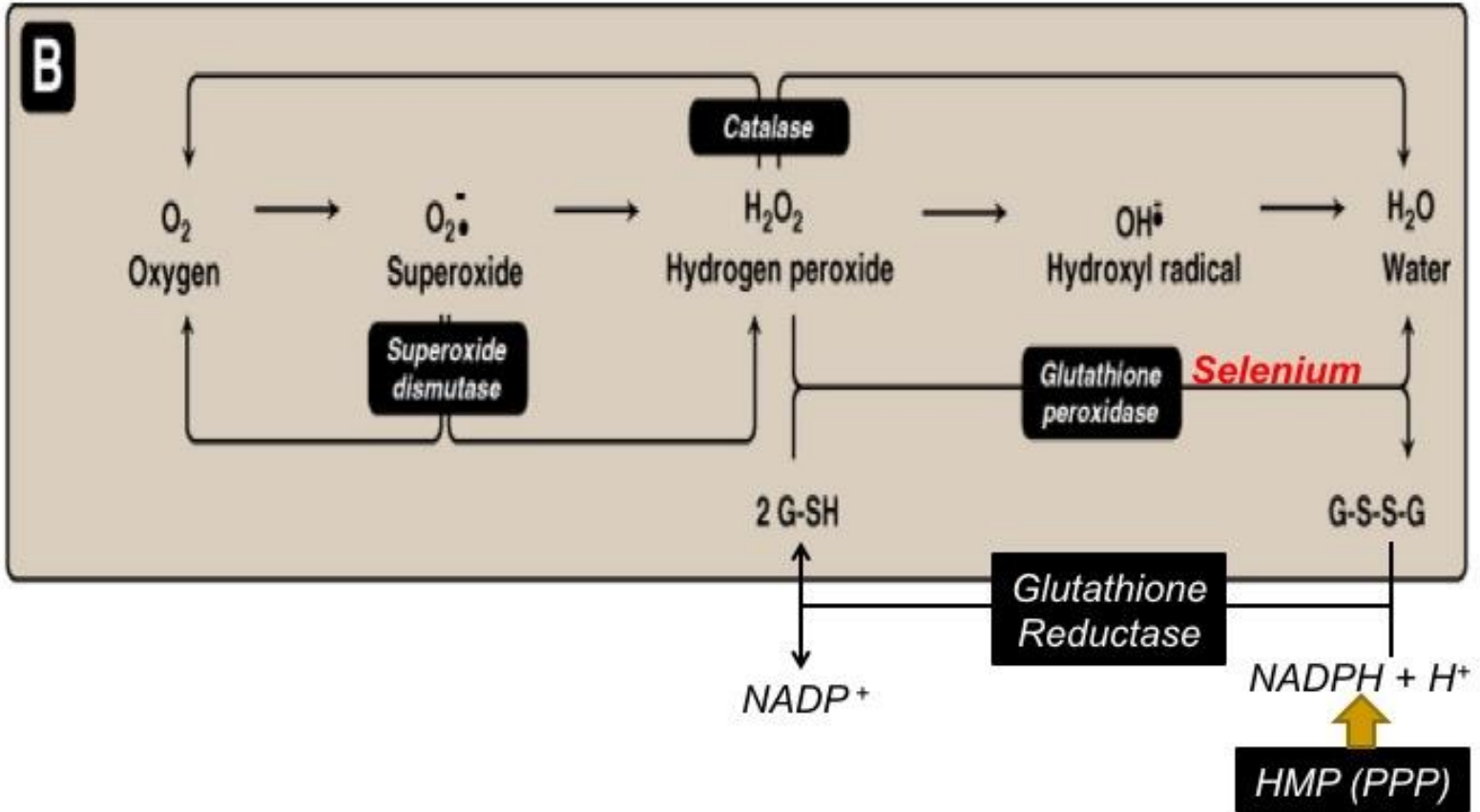
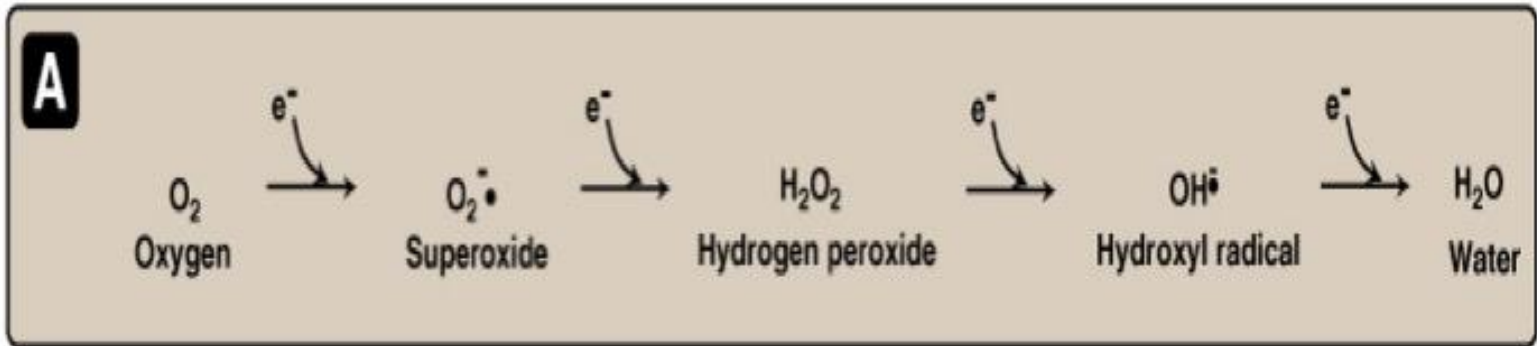
Metabolic stress

Neurochemical response

Oxidative stress

The Role of Reactive Oxygen Species (ROS) & Reactive Nitrate Species (RNS) in Normal Brain Physiology

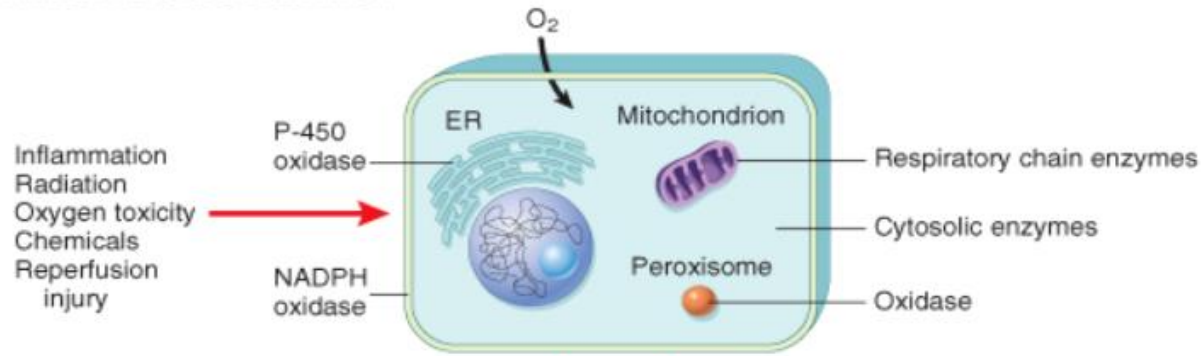
- They are mainly generated by microglia & astrocytes
- They modulate synaptic transmission & non-synaptic communication between neurons & glia
- During periods of increased neuronal activity, ROS & RNS diffuse to the myelin sheath of oligodendrocytes activating Protein kinase C (**PKC**) → posttranslational modification of myelin basic protein (**MBP**) by phosphorylation
- They regulate neuronal signaling in both central & peripheral nervous systems
- They are required for essential processes as learning & memory formation



Oxidative stress

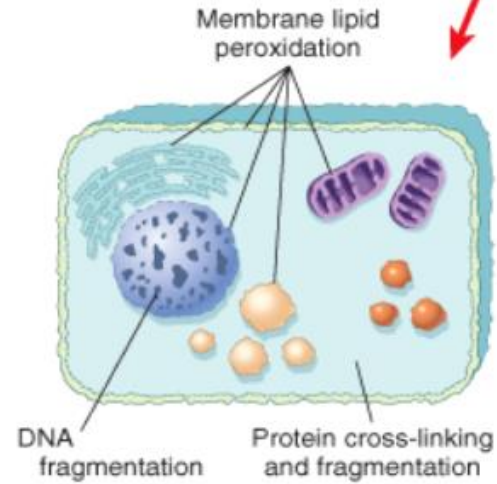
- A condition in which cells are subjected to excessive levels of Reactive oxidizing species (ROS or RNS) & they are unable to counterbalance their deleterious effects with antioxidants.
 - It has been implicated in the ageing process & in many diseases (e.g., atherosclerosis, cancer, neurodegenerative diseases, stroke)
-

A. FREE RADICAL GENERATION

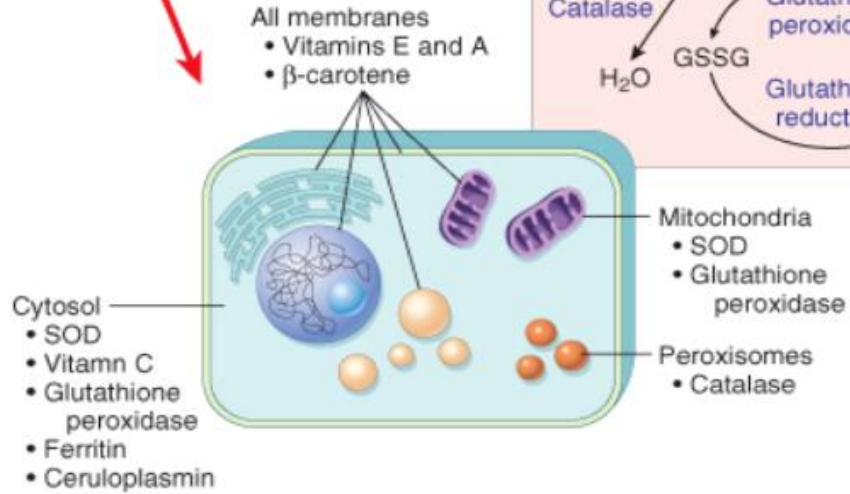


Reactive oxygen species:
 $O_2^{\cdot-}$, H_2O_2 , OH^{\cdot}

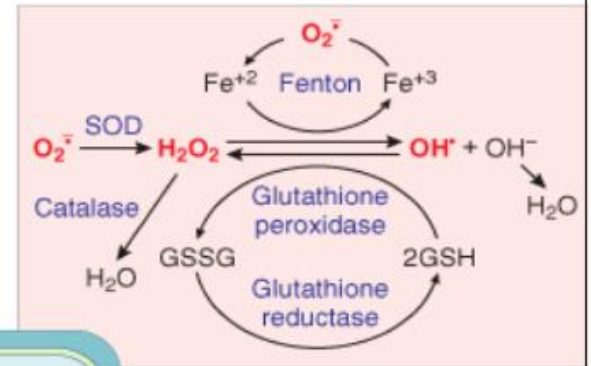
Reactive oxygen species:
 $O_2^{\cdot-}$, H_2O_2 , OH^{\cdot}



B. CELL INJURY BY FREE RADICALS



C. NEUTRALIZATION OF FREE RADICALS – NO CELL INJURY



The brain and Oxidative stress

- The brain is highly susceptible to ROS-induced damage because of:
 - ❑ High concentrations of peroxidisable lipids
 - ❑ Low levels of protective antioxidants
 - ❑ High oxygen consumption
 - ❑ High levels of iron (acts as pro-oxidants under pathological conditions)
 - ❑ The occurrence of reactions involving dopamine & Glutamate oxidase in the brain
-

Molecular & Vascular effects of ROS in ischemic stroke

■ Molecular effects:

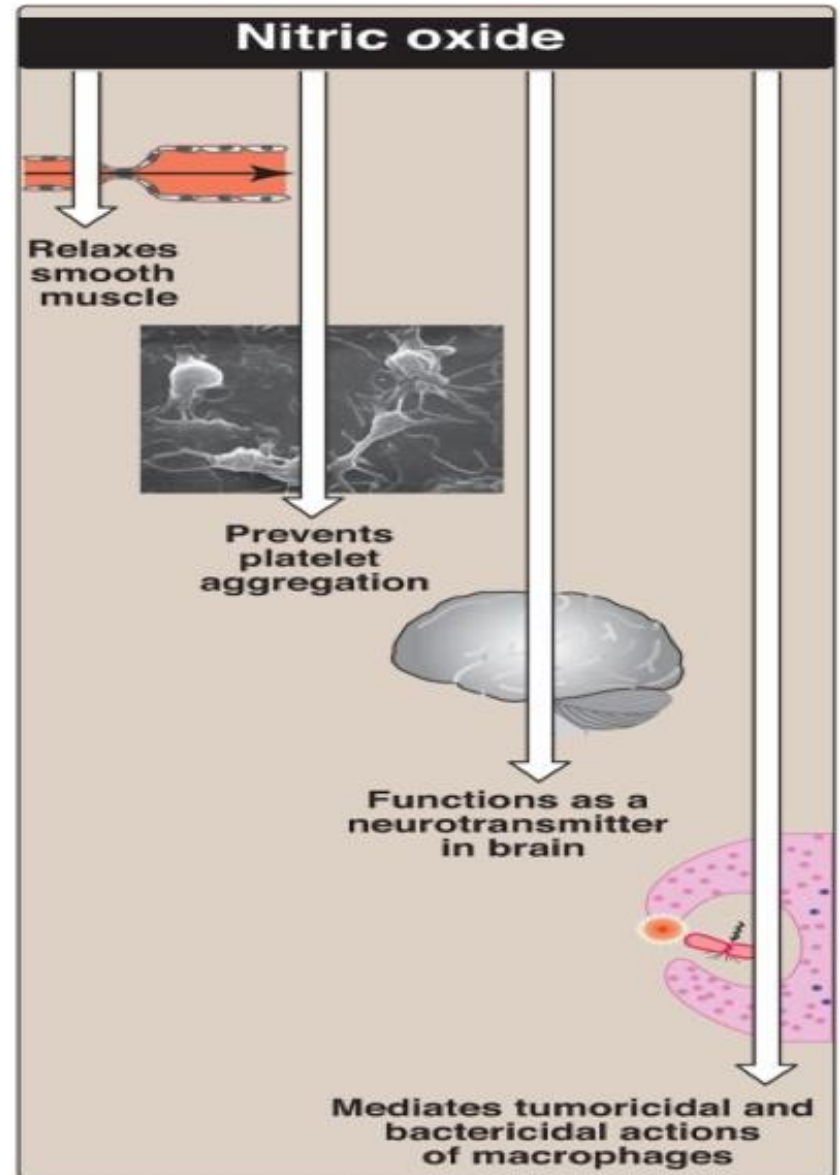
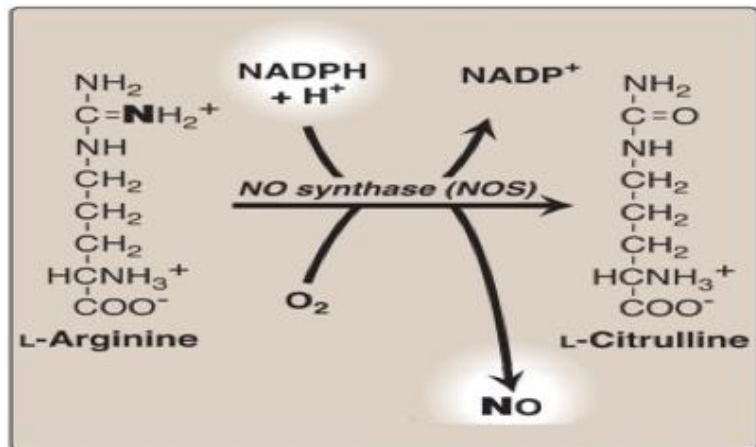
- ❑ DNA damage
- ❑ Lipid peroxidation of *unsaturated fatty acids*
- ❑ Protein denaturation
- ❑ Inactivation of **enzymes**
- ❑ Cell **signaling** effects (e.g., release of Ca^{2+} from intracellular stores)
- ❑ **Cytoskeletal** damage
- ❑ **Chemotaxis**

■ Vascular effects:

- ❑ Altered vascular tone and cerebral blood flow
- ❑ Increased platelet aggregability
- ❑ Increased endothelial cell permeability

The role of NO in the pathophysiology of cerebral ischemia

- Ischemia → abnormal NO production
 - This may be both **beneficial** and **detrimental**, depending upon when and where NO is released
 - NO produced by endothelial NOS (**eNOS**) → improving vascular dilation and perfusion (i.e. **beneficial**).
 - In contrast, NO production by neuronal NOS (**nNOS**) or by the inducible form of NOS (**iNOS**) has **detrimental** (harmful) effects.
 - Increased iNOS activity generally occurs in a delayed fashion after brain ischemia and trauma and is associated with inflammatory processes
-



Metabolic stress

Biochemical changes in The brain during ischemia

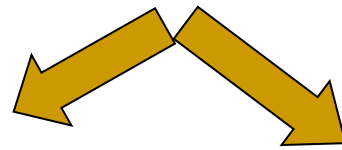
Ischemia → interruption or severe reduction of blood flow, O₂ & nutrients in cerebral arteries → **energy depletion** (depletion of ATP & creatine phosphate)

- Inhibition of ATP-dependent ion pumps
 - Membranes depolarization
 - Perturbance of transmembrane ion gradients



Ca²⁺ Influx (translocation from extracellular to intracellular spaces) → activation of cellular proteases (Calpains) & lipases → breakdown of cerebral tissue

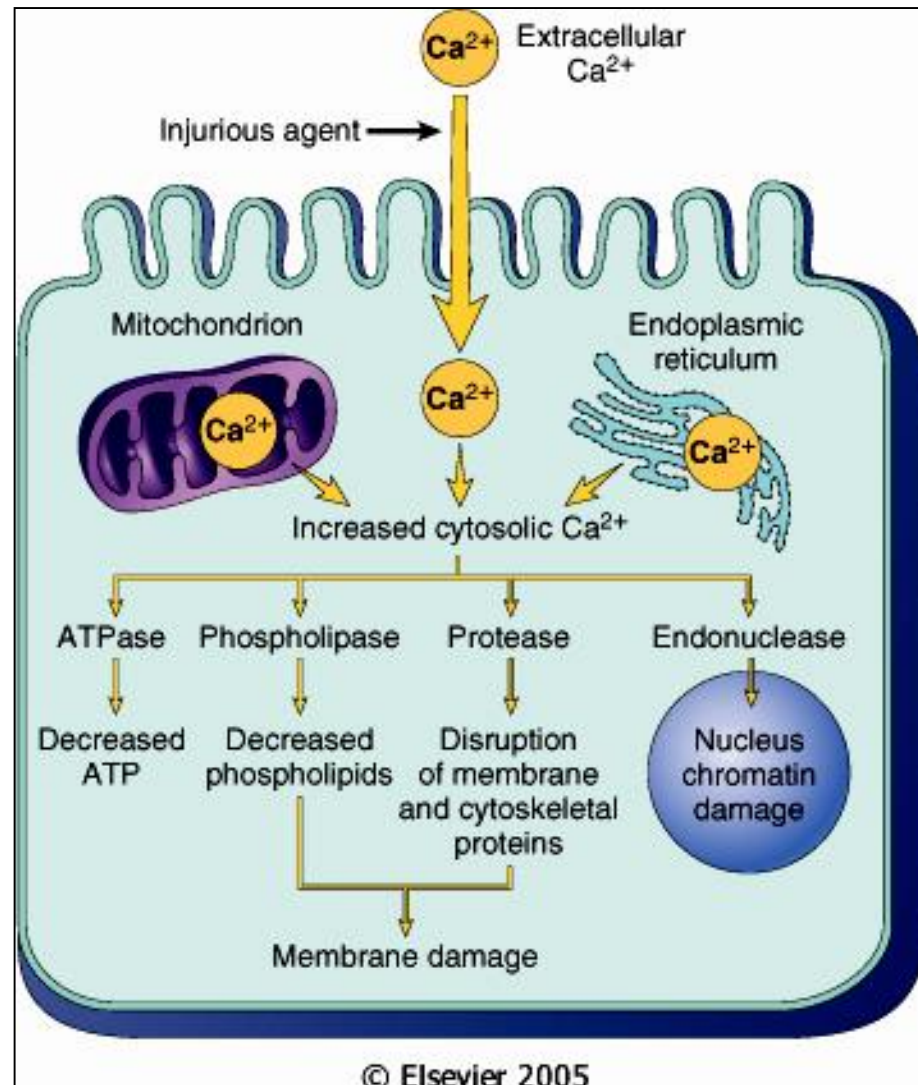
- **Na⁺ influx**
- **K⁺ efflux**



Lactic acid in neurons → acidosis → promotes the pro-oxidant effect → ↑ the rate of conversion of O₂^{·-} to H₂O₂ or to hydroxyperoxyl radical

• K⁺-induced **release of excitatory** amino acids

Sources & consequences of increased cytosolic Calcium in cell injury



Neurochemical response

The neurochemical response to cerebral ischemia

- Following cerebral ischemia, extracellular levels of various neurotransmitters are increased e.g.,
 - Glutamate
 - Glycine
 - GABA
 - Dopamine
-

The Blood tests in patients with brain ischemia or hemorrhage

- Complete blood count, including hemoglobin, hematocrit, white blood cell count, and platelet count
- Prothrombin time, international normalized ratio (INR), and activated partial thromboplastin time
- Thrombin time and/or ecarin clotting time if patient is known or suspected to be taking a direct thrombin inhibitor or a direct factor Xa inhibitor
- Blood lipids, including total, high-density lipoprotein (HDL), and low-density lipoprotein (LDL) cholesterol, and triglycerides.
- Cardiac enzymes and troponin

Biochemical basis of pharmacological intervention

Examples of Potential Biochemical Intervention in Cerebral Ischemia

- Inhibitors of glutamate release
 - Ca^{2+} channel blockers
 - Nitric oxide synthase inhibitors & free radical inhibition
 - Calpain inhibitors
-



To Summarize:



Ischemic cascade

Lack of oxygen supply to ischemic neurones



ATP depletion



Malfunctioning of membrane ion system



Depolarisation of neurones



Influx of calcium



Release of neurotransmitters, activation of proteases

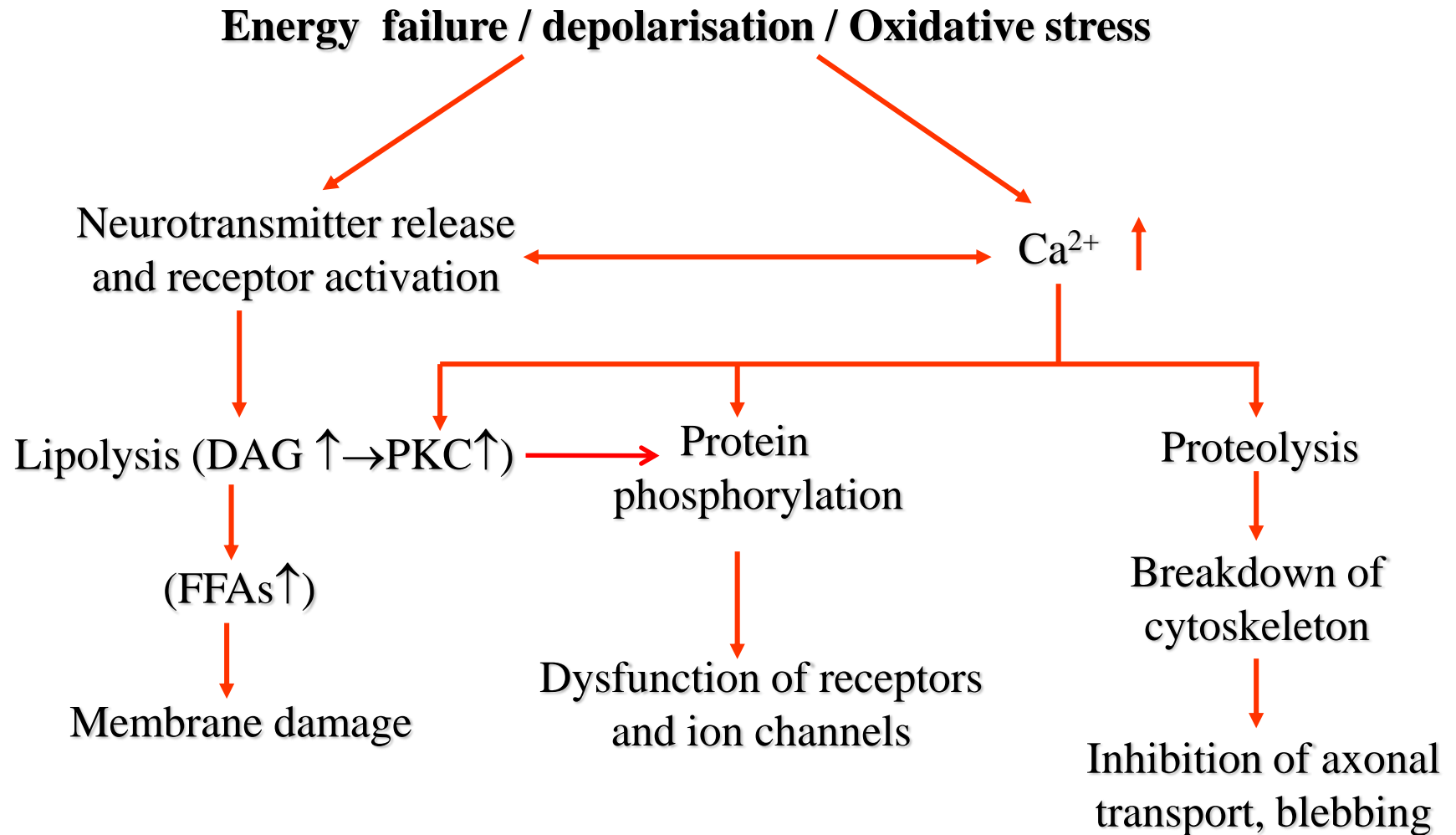


Further depolarisation of cells



Further calcium influx

Cosequences of brain ischemia



Take Home Message

Severe cerebral ischemic insults lead to a complex cascade of biochemical and molecular events, including:

1. Cell death
 2. Oxidative stress
 3. Metabolic stress and neurochemical changes
-

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