

Ferroptosis

Iron-dependent regulated cell death

Membrane lipid peroxidation • oxidative injury • cell-death mechanisms

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Learning objectives

Define ferroptosis and distinguish it from other forms of cell death.

Trace the roles of iron, oxidants, membrane lipids and antioxidant defenses.

Interpret ultrastructural findings and disease associations critically.

Use mechanistic evidence to evaluate whether ferroptosis occurred.

Ferroptosis in the cell-death landscape



Regulated injury pathways are mechanistically distinct even if tissue injury appears necrotic.

Definition: Iron-dependent lipid injury

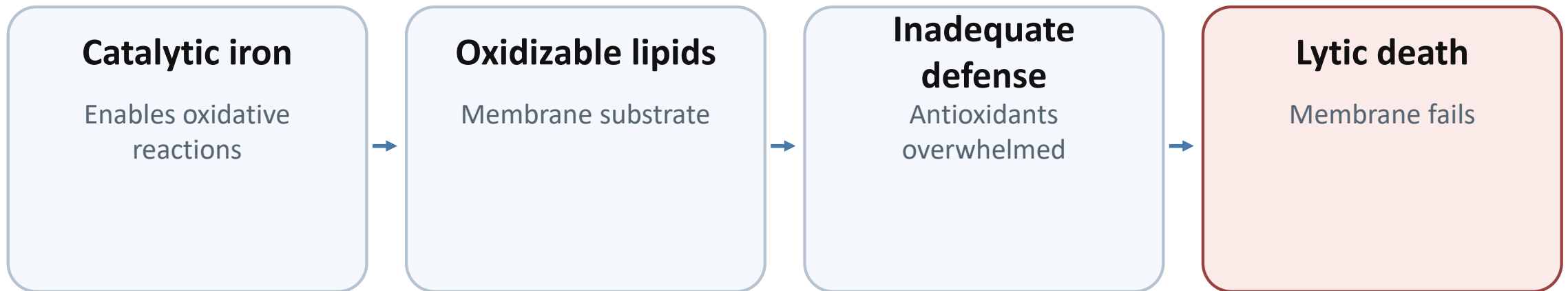
A distinct, regulated pathway of cell death.

Excess iron and/or oxidant burden defeats glutathione-dependent protection.

Unchecked membrane lipid peroxidation is central to the fatal process.

Ultimately, selective plasma-membrane barrier function fails.

Three converging requirements



The diagram is a conceptual convergence, not a universal quantitative threshold.

Regulated mechanism, necrosis-like end point

Regulated process

Specific biochemical vulnerabilities and controls.

Can be suppressed by limiting iron.

Distinct from unregulated chemical membrane destruction.

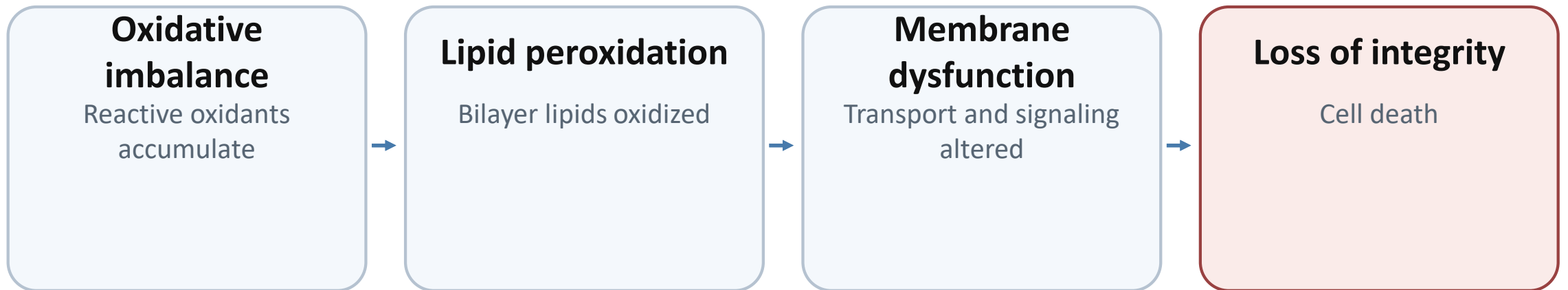
Necrosis-like result

Barrier breakdown and eventual cell death.

Cell death may be lytic rather than apoptotic.

Final morphology alone does not specify mechanism.

Where the lethal damage occurs



Distinguish a mechanistic sequence from a histologic diagnostic criterion.

Mechanism versus microscopic appearance

Mechanism

Iron-dependent oxidation of phospholipids.

Defective antioxidant control.

A regulated cell-death pathway.

Appearance

Loss of membrane integrity, resembling necrosis.

Characteristic mitochondrial alterations reported.

Conventional H&E is not pathway-specific.

Reactive oxygen species have normal roles

Low levels arise during ordinary cellular metabolism.

Scavenging pathways normally constrain oxidant accumulation.

Physiologic signaling can use controlled oxidant production.

Ferroptosis concerns destructive oxidation, not ROS presence alone.

Oxidative stress is a balance problem

ROS production

Normal respiration, cellular enzymes and stress

Versus

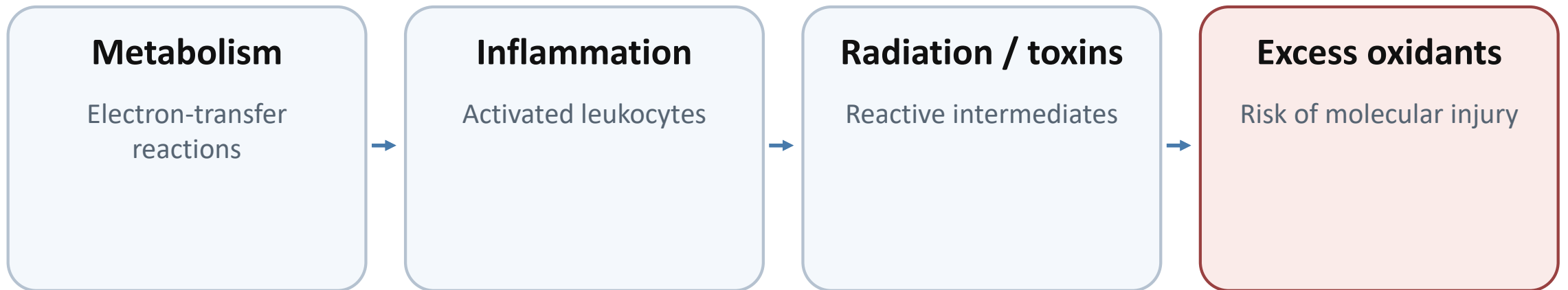
ROS removal

Antioxidants and detoxifying enzymes

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graph TD; A[ROS production<br/>Normal respiration, cellular enzymes and stress] --> D[Oxidant load exceeds protection]; B[ROS removal<br/>Antioxidants and detoxifying enzymes] --> D; A -- Versus --> B;
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Oxidant load exceeds protection

Sources of intracellular oxidants



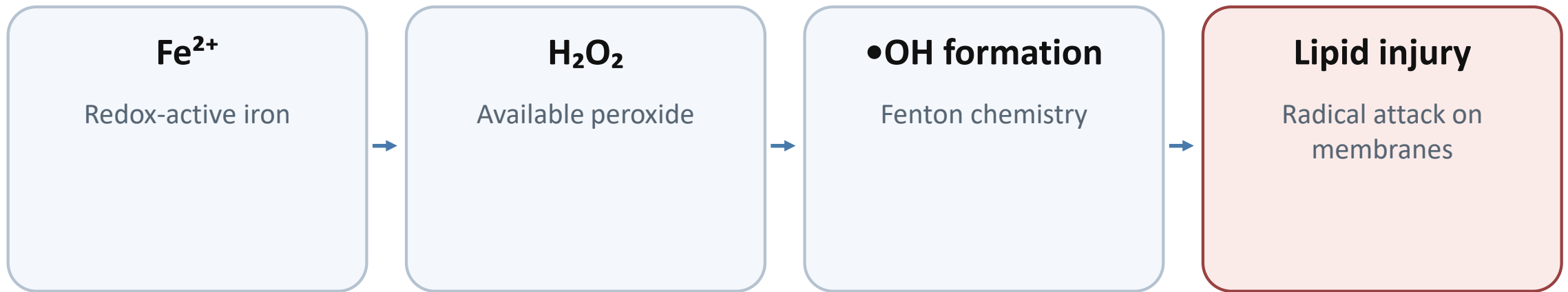
Sources of ROS in cell injury; Not every source specifically proves ferroptosis.

Major reactive oxygen intermediates

Species	Biochemical significance
Superoxide ($\text{O}_2\bullet^-$)	Partially reduced oxygen; Precursor of other ROS.
Hydrogen peroxide (H_2O_2)	Nonradical ROS; Substrate for iron-driven chemistry.
Hydroxyl radical ($\bullet\text{OH}$)	Highly reactive species capable of molecular injury.

H_2O_2 is a reactive oxygen species but not a free radical.

Iron accelerates oxidant chemistry



Fenton reaction is an important general model of iron-catalyzed injury.

Fenton reaction: A causal example

Ferrous iron (Fe^{2+}) reacts with hydrogen peroxide.

The reaction produces ferric iron (Fe^{3+}), hydroxyl radical ($\bullet\text{OH}$) and hydroxide.

Hydroxyl radicals are highly reactive toward cellular molecules.

Do not equate all ferroptotic lipid oxidation with one chemical reaction.

Iron binding limits free-radical chemistry

Restricted iron

Transferrin and ferritin bind iron in transport and storage.

Metal-binding proteins limit free-ion reactivity.

This reduces oxidant-generating chemistry.

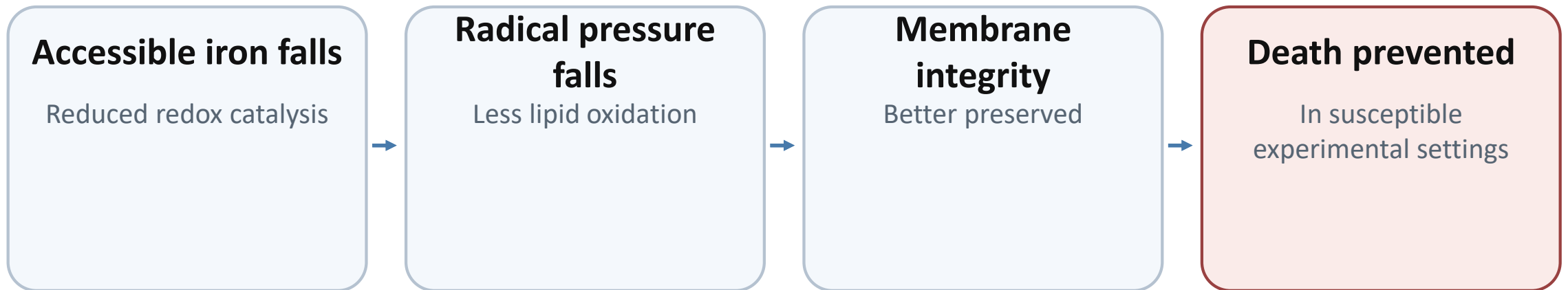
Available redox-active iron

Greater catalytic capacity under permissive conditions.

Can cooperate with ROS to amplify injury.

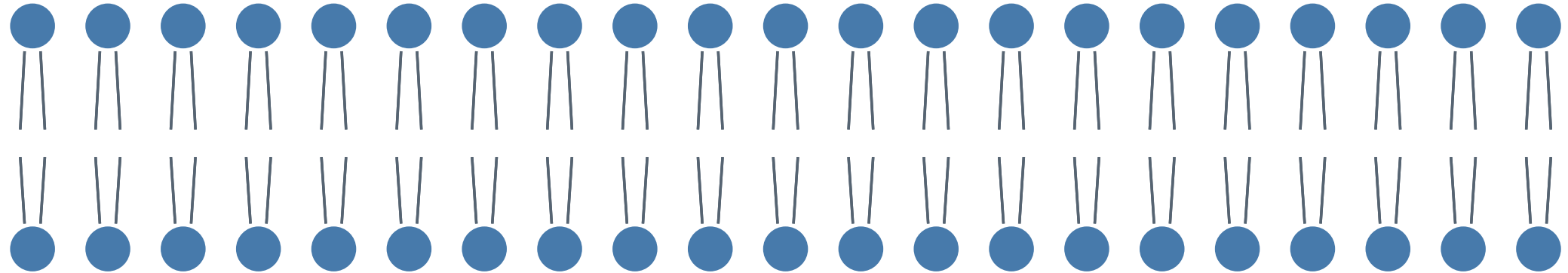
Specific cellular pools matter more than iron abundance alone.

Why iron reduction can prevent ferroptosis



Membrane phospholipids are the substrate

Schematic phospholipid bilayer



Head groups

Water-facing

Fatty-acid tails

Membrane core

Intact barrier

Selective transport

Original editable schematic — not a micrograph

Unsaturated lipid bonds are vulnerable

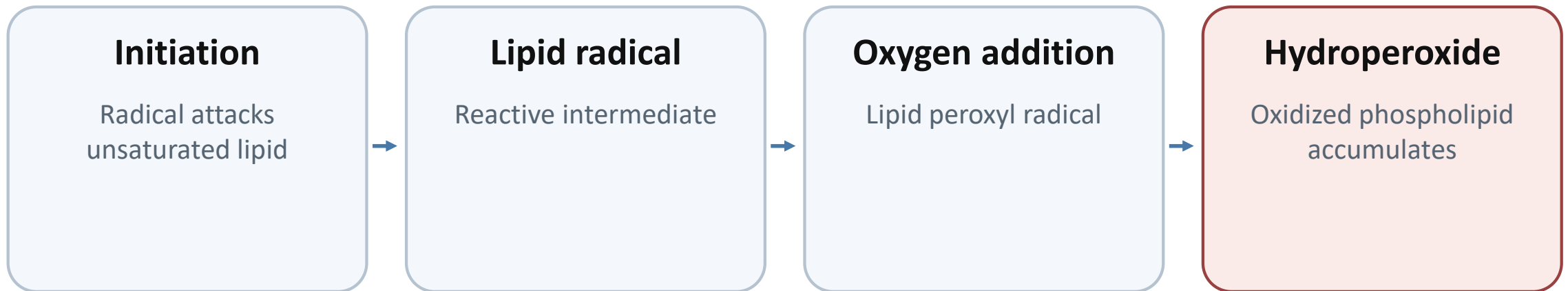
Double bonds in unsaturated membrane fatty acids are sites of radical attack.

The initial oxidative event creates reactive lipid intermediates.

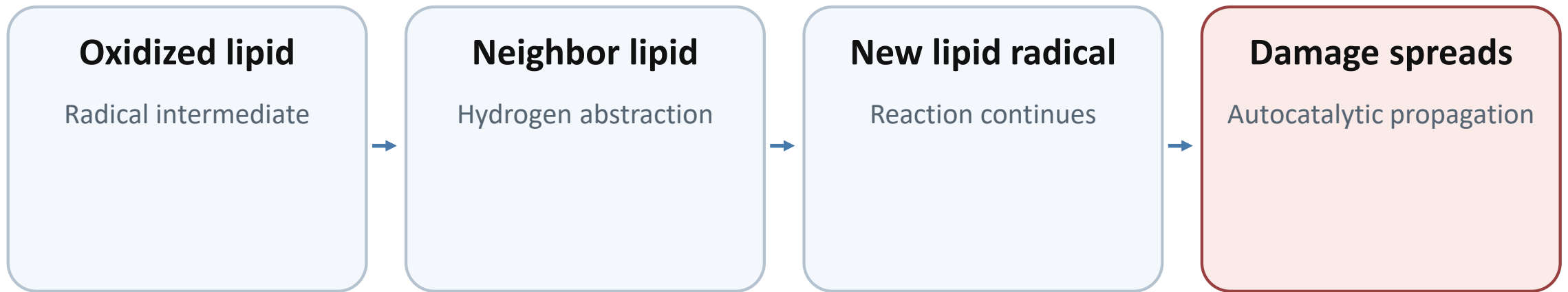
Susceptibility depends on membrane lipid composition.

The specific phospholipid landscape is an important biological modifier.

Lipid peroxidation begins with radical attack



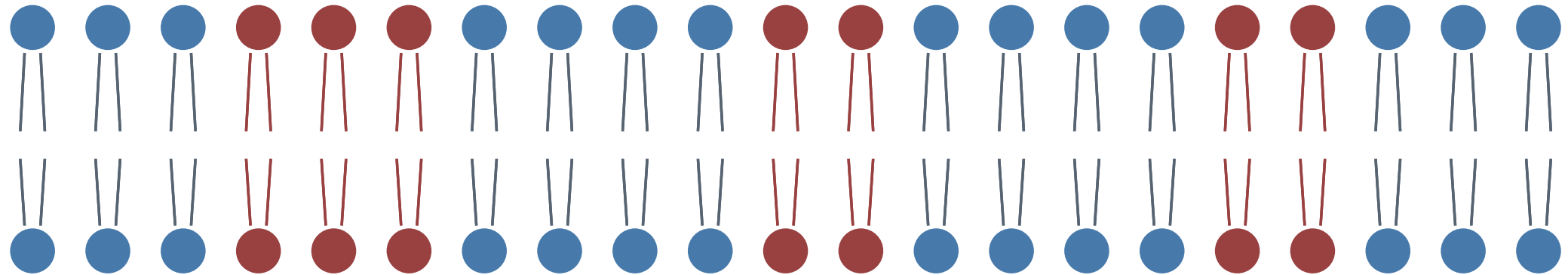
Peroxidation propagates as a chain



Schematic chain propagation; Reaction rates and stoichiometry not quantified.

Unchecked oxidation destabilizes the bilayer

Schematic phospholipid bilayer



Oxidation

Altered membrane lipids

Protein context

Normal interactions perturbed

Functional loss

Barrier weakened

Original editable schematic — not a micrograph

Membrane fluidity changes

Lipid composition and organization govern membrane fluidity.

Extensive oxidation perturbs lipid packing and membrane behavior.

Impaired fluidity alters the physical environment of integral proteins.

This is one route from chemical damage to cellular dysfunction.

Lipid–protein interactions are disturbed

Intact membrane

Lipids support normal organization of membrane proteins.

Protein activities depend partly on local lipid environment.

After extensive lipid oxidation

Altered bilayer may disturb protein interactions.

Transporters and receptors may no longer function normally.

Ion and nutrient transport become abnormal

Lipid oxidation

Membrane physical properties
change



Transport failure

Ion/nutrient homeostasis disturbed



Cell dysfunction

Metabolism and gradients destabilize

Membrane signaling is not spared

Plasma membranes organize receptors and signaling proteins.

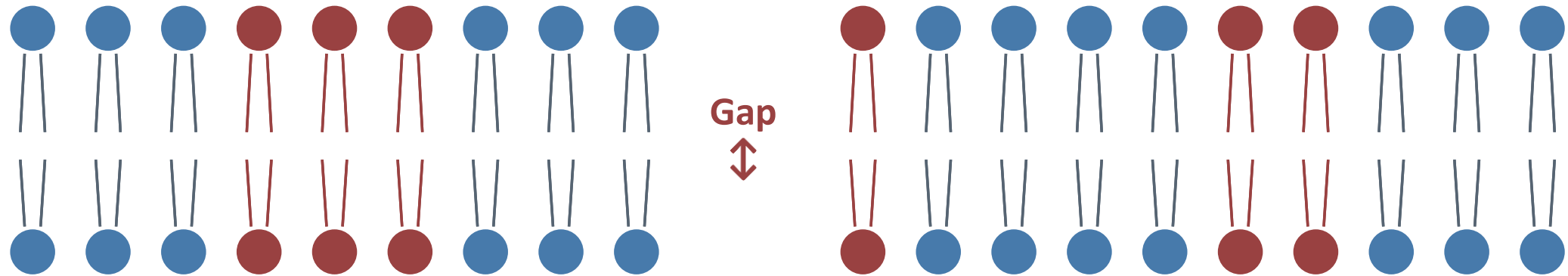
Lipid oxidation can alter lipid–protein relationships.

Consequent disruption of signaling contributes to cellular failure.

Signaling changes should not be mistaken for a ferroptosis-specific test.

Loss of selective permeability is lethal

Schematic phospholipid bilayer



Before injury

Selective membrane permeability

During injury

Peroxidized phospholipids

After injury

Loss of barrier function

Original editable schematic — not a micrograph

Different membranes, shared vulnerability

Plasma membrane

Selective permeability supports cellular homeostasis.

Barrier failure produces necrosis-like loss of integrity.

Organelle membranes

ROS can damage membranes throughout the cell.

Mitochondrial alterations may accompany ferroptosis.

Ultrastructural findings reported in ferroptosis

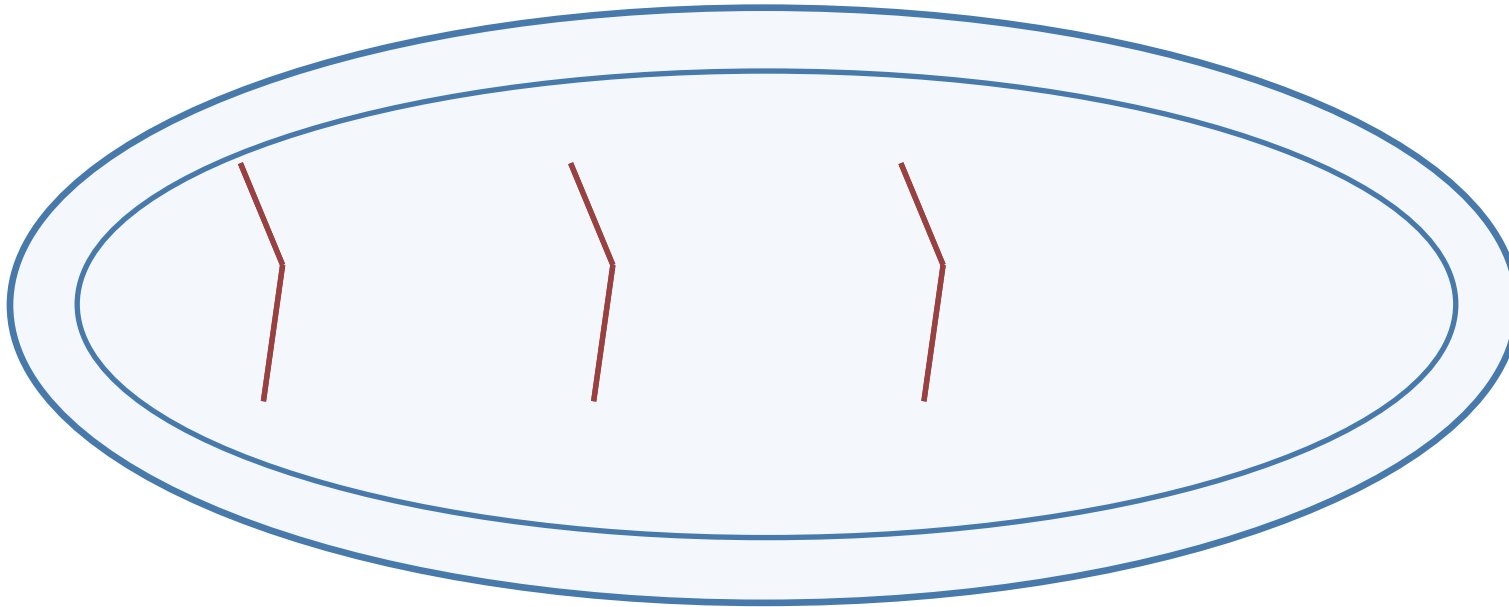
Prominent mitochondrial cristae loss.

Disruption or rupture of the outer mitochondrial membrane.

These abnormalities have been described ultrastructurally.

Interpretation requires experimental context and appropriate methods.

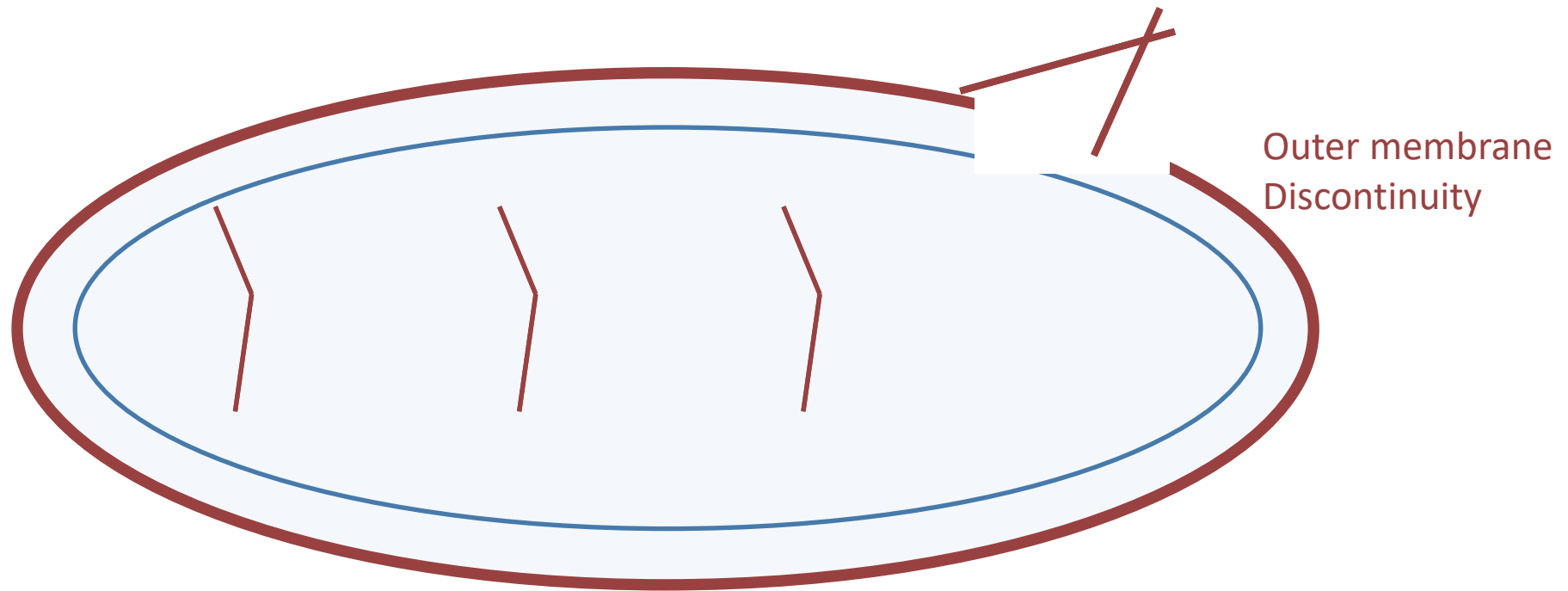
Cristae loss: Conceptual ultrastructure



Fewer cristae / disrupted outer boundary in the source description

Native PowerPoint schematic; Not diagnostic electron microscopy

Outer mitochondrial membrane disruption



Fewer cristae / disrupted outer boundary in the source description

Native PowerPoint schematic; Not diagnostic electron microscopy

Morphology provides clues, not proof

What morphology contributes

Can show mitochondria with diminished cristae.

Can show outer membrane disruption.

Supports a mechanism when paired with assays.

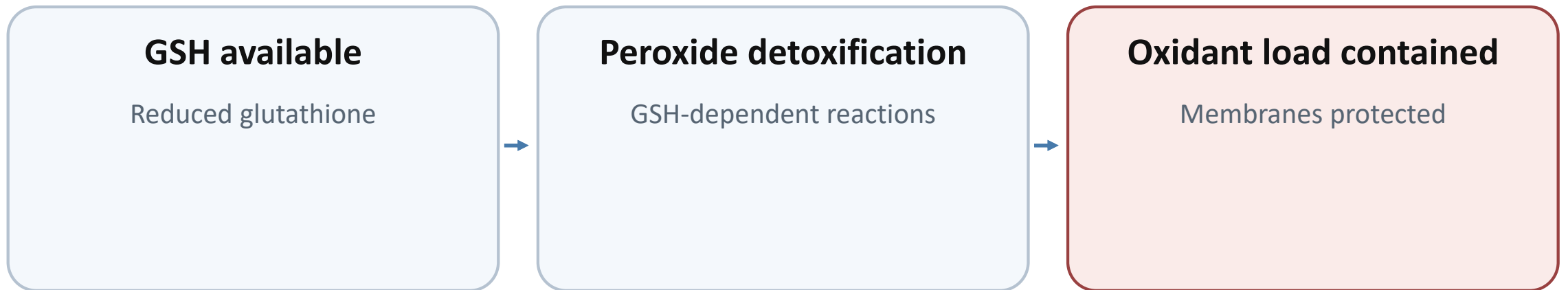
What morphology cannot settle

Cannot determine iron dependence alone.

Cannot identify a unique biochemical cascade.

Does not replace orthogonal rescue experiments.

Glutathione is a central antioxidant reserve



GSH-dependent protection is explicit; Downstream molecular details are supplemental.

Glutathione peroxidase removes peroxides

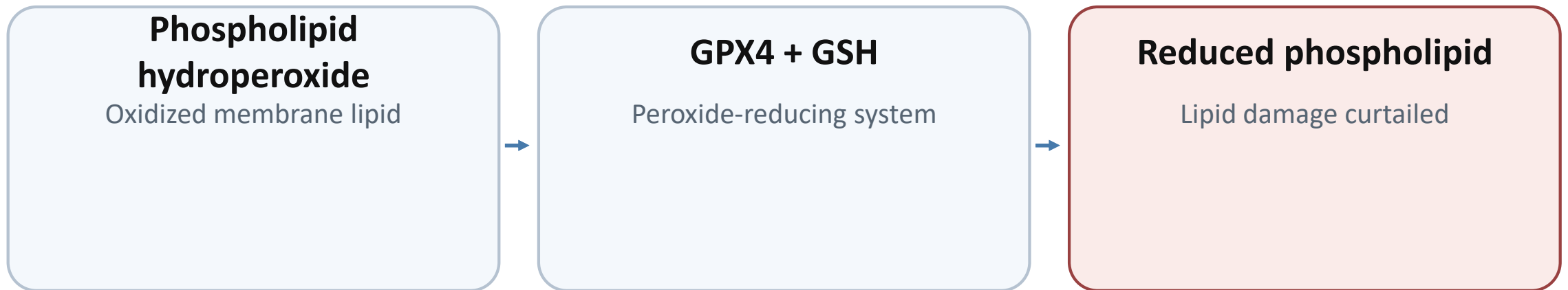
Glutathione peroxidase uses reduced GSH in peroxide-detoxifying reactions.

Oxidized glutathione (GSSG) is generated during this process.

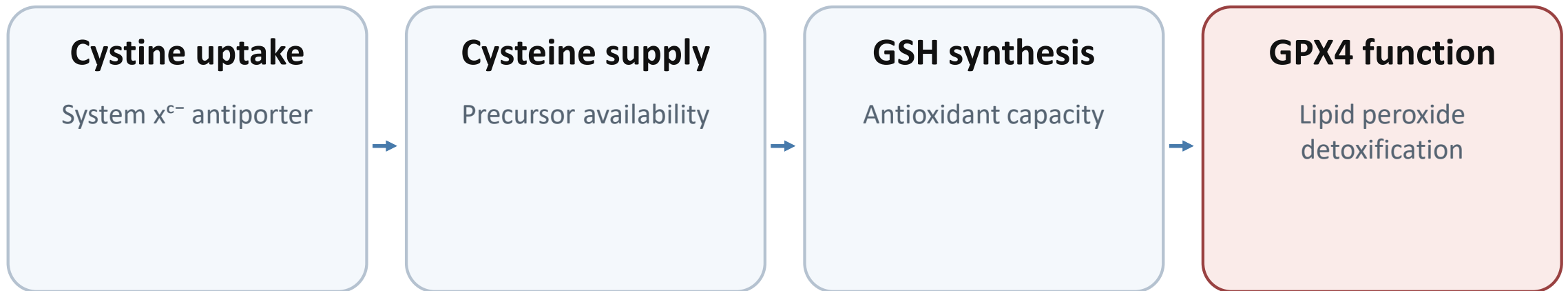
Cellular GSH/GSSG balance reflects redox state.

When defenses are overwhelmed, oxidized lipids can accumulate.

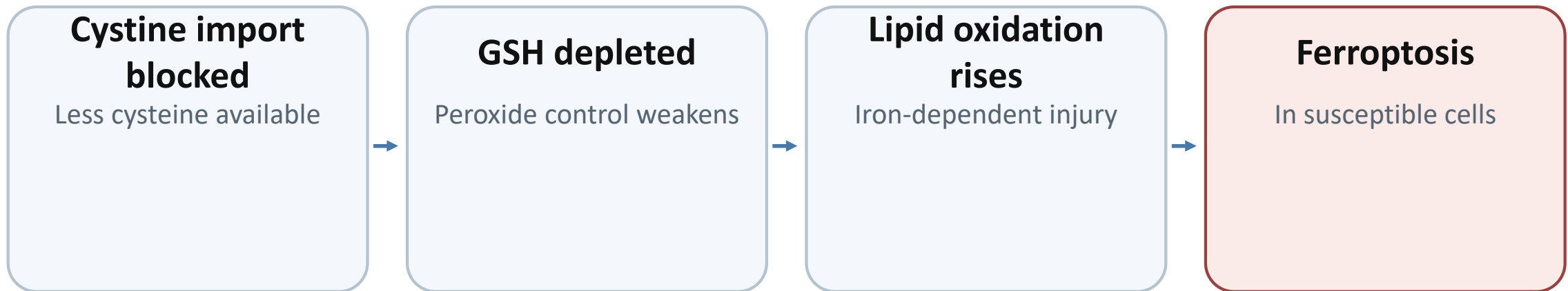
GPX4 protects membrane phospholipids



System x^{c-} supports GSH synthesis

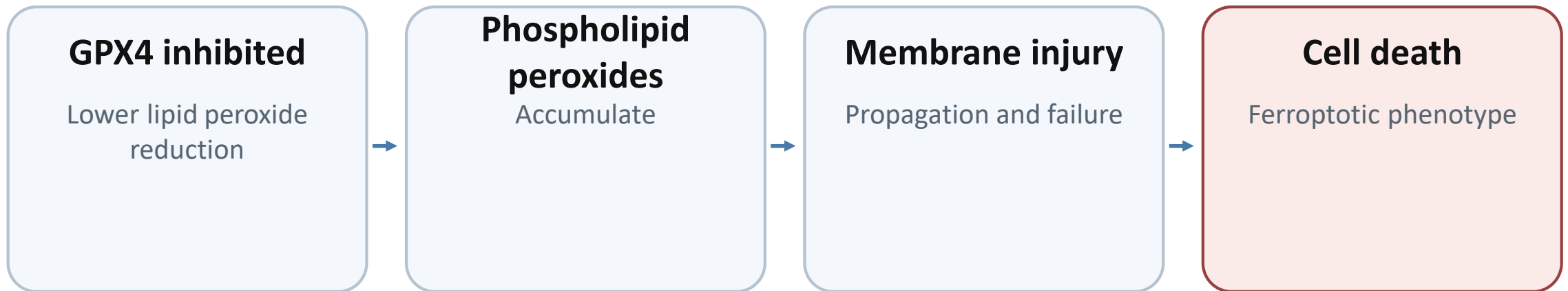


Inhibiting cystine import creates vulnerability

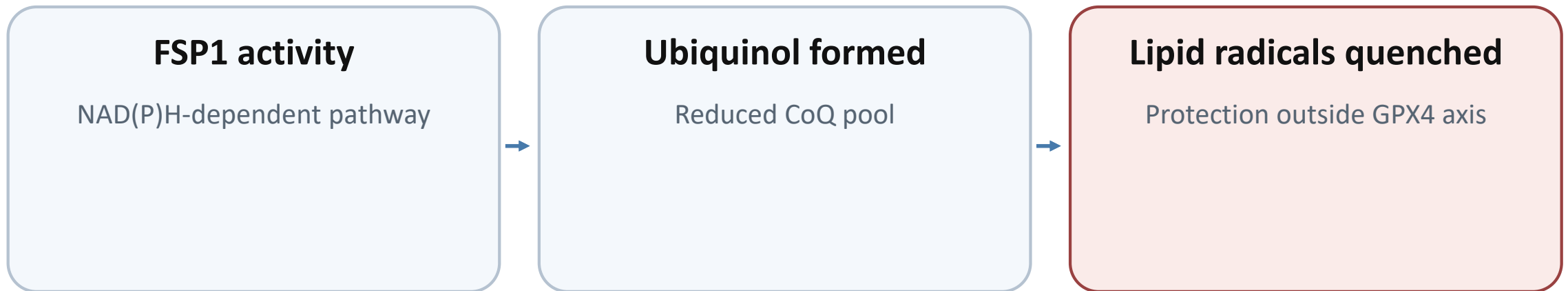


Experimental example, not a universal response in every cell type.

Direct impairment of GPX4 also sensitizes cells



A parallel antioxidant defense: FSP1



Supplementary pathway; Represented schematically without claiming precise flux or location.

Multiple protections converge on lipids

Protective arm	Principal role
Iron-binding proteins	Restrict redox-active metal chemistry.
GSH–GPX4	Reduce membrane lipid hydroperoxides.
FSP1–CoQ	Radical-trapping antioxidant protection.
Vitamin E and other antioxidants	Interrupt free-radical injury under suitable conditions.

Injury occurs when protection is surpassed

Pro-oxidant drive

Redox-active iron + reactive lipid intermediates

Versus

Antioxidant defenses

GSH–GPX4, FSP1 and metal sequestration

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graph TD; A[Pro-oxidant drive<br/>Redox-active iron + reactive lipid intermediates] --> D[Lipid peroxidation escapes control]; B[Antioxidant defenses<br/>GSH–GPX4, FSP1 and metal sequestration] --> D; A --- C[Versus] --- B;
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Lipid peroxidation escapes control

Three mechanistic levers to investigate

Promote ferroptotic injury

Increase accessible redox-active iron.

Increase oxidizable membrane substrate.

Compromise lipid-peroxide detoxification.

Oppose ferroptotic injury

Restrict accessible iron.

Interrupt radical chain propagation.

Preserve antioxidant defense function.

Lipid peroxidation is not every membrane injury

Route of injury	Primary disturbance
Ferroptotic lipid damage	Iron-dependent oxidative peroxidation.
ATP depletion	Reduced phospholipid synthesis and pump function.
Calcium-activated phospholipases	Increased phospholipid breakdown.
Cytoskeletal proteolysis	Loss of membrane anchorage and stability.

Ferroptosis and passive necrosis differ

Ferroptosis

Biochemically regulated lipid oxidation.

Depends on iron and antioxidant status.

May culminate in lytic morphology.

Classical accidental necrosis

Severe injury produces membrane failure.

Multiple upstream causes are possible.

A similar end point does not imply the same mechanism.

Ferroptosis versus apoptosis

Characteristic	Ferroptosis	Apoptosis
Central machinery	Iron-dependent lipid oxidation	Caspase-mediated proteolysis
Membrane fate	Eventual loss of integrity	Often intact until phagocytic removal
Characteristic morphology	Mitochondrial cristae alteration	Cell shrinkage; Apoptotic bodies
Inflammatory context	Lytic end point possible	Usually limited by rapid clearance

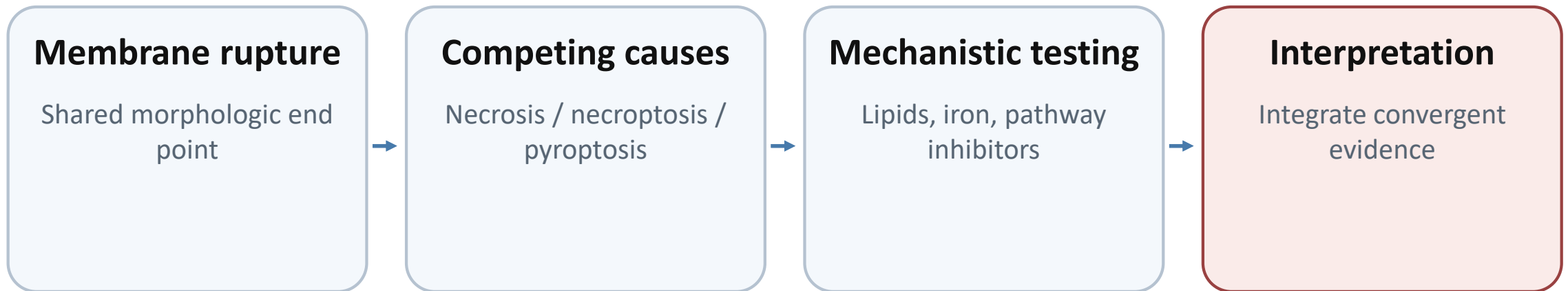
Ferroptosis versus necroptosis

Feature	Ferroptosis	Necroptosis
Principal mechanism	Iron-dependent lipid peroxidation	RIPK1/RIPK3–MLKL signaling
Membrane disruption	Oxidative lipid damage	MLKL-mediated membrane disruption
Diagnostic inference	Requires biochemical/rescue evidence	Requires pathway-specific evidence
Shared feature	Necrosis-like cell death	Necrosis-like cell death

Ferroptosis versus pyroptosis

Feature	Ferroptosis	Pyroptosis
Key trigger / context	Oxidative lipid damage	Inflammasome or cytosolic LPS pathways
Key effector	Phospholipid peroxidation	Gasdermin D pore formation
Inflammatory mediators	Not defined by IL-1 β processing	Associated with IL-1 β release
Outcome	Membrane failure	Osmotic disruption and lysis

Do not classify cell death by lysis alone



Ferroptosis diagnosis is mechanistic, not merely morphologic.

A ferroptosis claim needs converging evidence

Show an iron- and lipid-peroxidation–linked injury phenotype.

Demonstrate biochemical evidence consistent with oxidized membrane lipids.

Test protection by interventions with relevant mechanisms.

Exclude plausible alternative regulated cell-death routes.

Oxidized lipid measurement: Use caution

Informative evidence

Biochemical assays of lipid peroxidation.

Appropriate temporal relation to loss of viability.

Orthogonal lipid readouts strengthen the inference.

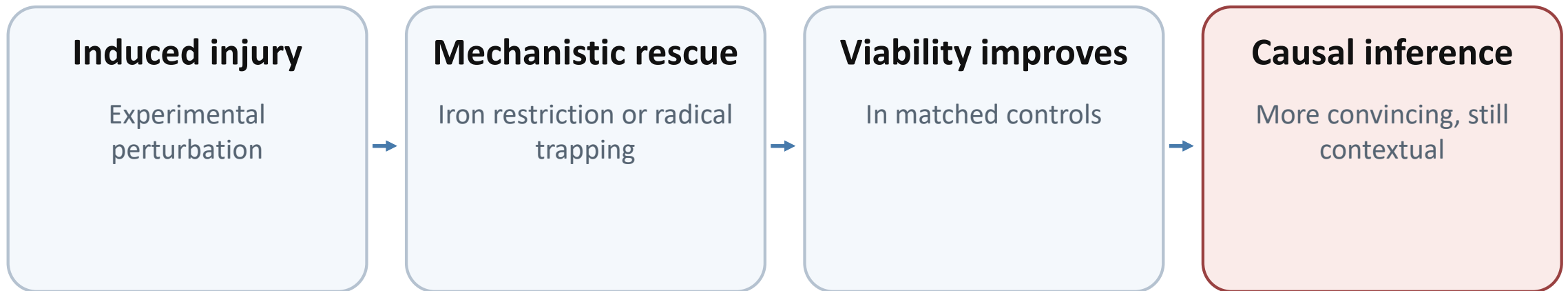
Limitations

Oxidative stress also occurs in other types of injury.

Single assays have technical and biological confounders.

A positive lipid signal is not a stand-alone diagnosis.

Mechanism-based rescue strengthens attribution



Do not over-interpret nonselective compounds as pathognomonic.

GPX4 status alone is insufficient

GPX4 participates in a major suppressive defense system.

Changes in protein level or expression are not equivalent to functional activity.

Parallel ferroptosis-suppressive systems also exist.

Interpret pathway markers together with lipid injury and rescue data.

Experimental inference is not clinical diagnosis

In experimental models

Perturbation and rescue can interrogate causality.

Lipid oxidation and survival can be quantified.

Genetic manipulation can test mechanism.

In clinical tissue

Routine H&E cannot establish ferroptosis.

No single universally accepted clinical diagnostic stain.

An association does not prove an individual lesion was ferroptotic.

Cancer: Vulnerability and therapeutic hypothesis

Cancer among pathologies linked with ferroptosis.

Some cancer models exhibit sensitivity to GPX4-related perturbation.

Inducing ferroptosis is an investigated antitumor strategy.

Preclinical susceptibility must not be presented as an established clinical indication.

Neurodegeneration: Proposed injury mechanism

Ferroptosis with neurodegenerative disorders.

Neural tissue may be affected by altered iron and oxidative lipid injury.

Experimental studies motivate ferroptosis-inhibition strategies.

These links do not establish one uniform mechanism in every patient or disease.

Stroke: Ischemic injury and oxidative context

Stroke is explicitly identified among ferroptosis-linked conditions.

Oxidative stress also occurs in ischemia-reperfusion injury.

Multiple distinct cell-death pathways may coexist in injured brain.

A clinical neuroprotective benefit from ferroptosis targeting cannot be assumed.

Disease association $\neq$ validated therapy

What can be stated

Ferroptosis is biologically connected with selected pathologies.

Drug-target ideas follow from preclinical mechanisms.

The field has plausible therapeutic directions.

What should not be asserted

A pathology association proves causation in each patient.

A specific drug is clinically effective without trial data.

A single molecular marker validates patient-level diagnosis.

Case exercise: A cell-culture death phenotype

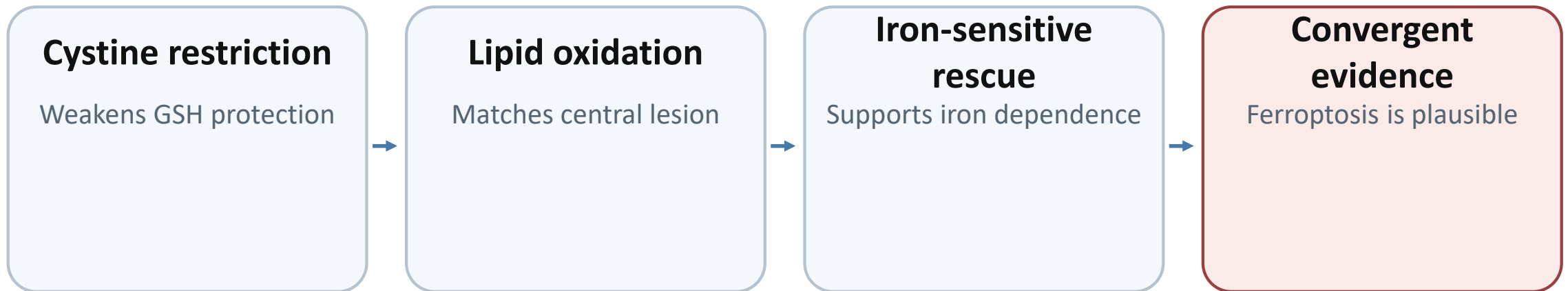
A cell-culture perturbation reduces cystine uptake. Reduced glutathione falls, lipid oxidation rises, and viability declines. Iron reduction and a radical-trapping agent improve survival.

Discussion questions

Which observations support ferroptosis?

What alternative mechanisms and controls require examination?

Case interpretation: Triangulate the mechanism



Rescue is evidence of mechanism, not absolute proof; Controls and specificity remain required.

Case exercise: Lytic cell death in infection

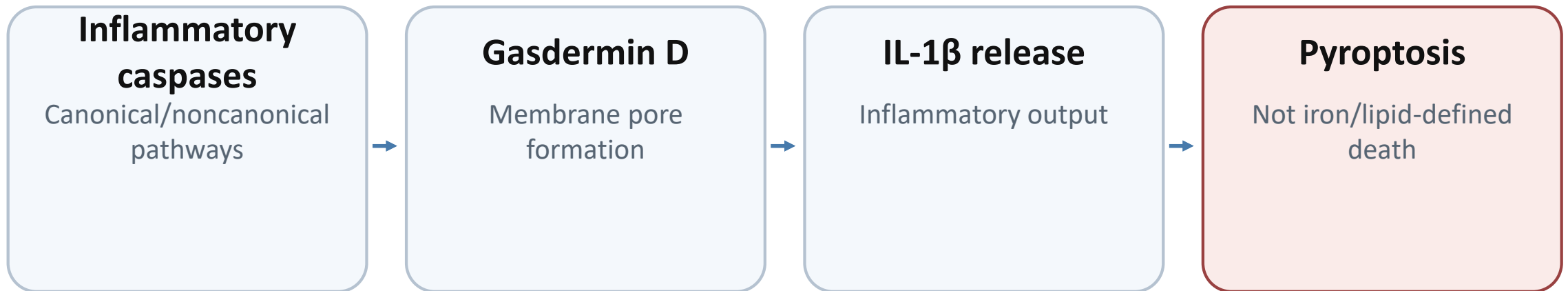
Infected immune cells activate inflammatory caspases, cleave gasdermin D and release IL-1 β through membrane pores. The cells ultimately rupture.

Discussion questions

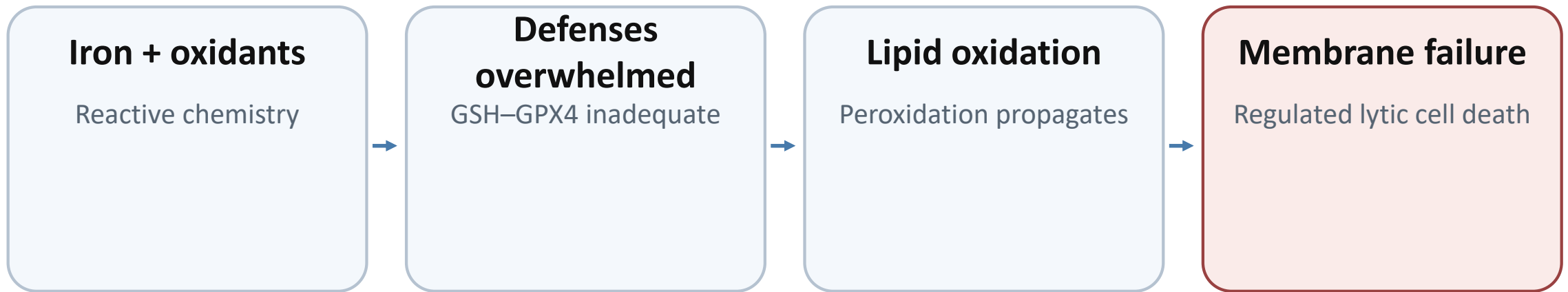
Which form of regulated cell death is more directly supported?

Why is membrane rupture alone insufficient to infer ferroptosis?

Case interpretation: Mechanism determines the label



An integrated causal framework



Simplified logic: Antioxidant failure is permissive and can occur upstream or alongside oxidation.

Ferroptosis: Five defensible conclusions

Its defining lesion is iron-dependent membrane lipid peroxidation.

The lethal transition reflects oxidant burden exceeding antioxidant defenses.

It can culminate in necrosis-like plasma-membrane failure.

Mitochondrial cristae loss and outer-membrane disruption are reported ultrastructural findings.

Disease links are biologically important but not equivalent to clinical diagnostic or treatment standards.

Source references

Primary textbook and contextual sections

Robbins, Cotran & Kumar: Pathologic Basis of Disease, 11th edition (2026), Chapter 2.

Ferroptosis and related key concepts in Chapter 2.

Related sections: Membrane injury, oxidative stress, free radical generation and removal, and cell-death pathways.

Supplementary mechanistic references

Dixon SJ et al. Ferroptosis: an iron-dependent form of nonapoptotic cell death. *Cell*. 2012;149:1060–1072. doi:10.1016/j.cell.2012.03.042.

Yang WS et al. Regulation of ferroptotic cancer cell death by GPX4. *Cell*. 2014;156:317–331. doi:10.1016/j.cell.2013.12.010.

Jiang X, Stockwell BR, Conrad M. Ferroptosis: mechanisms, biology and role in disease. *Nat Rev Mol Cell Biol*. 2021;22:266–282. doi:10.1038/s41580-020-00324-8.