

Compartment-Resolved ERK Signaling in Oxidant-Linked Mitochondrial Outcomes

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Abstract

Background: Oxidative-stress studies often compress reactive oxygen species, ERK phosphorylation, mitochondrial dysfunction, mitophagy and cell fate into one injury narrative. That compression leaves unresolved whether compartment-localized ERK activity helps set mitochondrial endpoint identity or merely marks generic upstream damage.

Objective: This working paper retains the selected question: Under matched oxidant or mitochondrial-stress conditions, does compartment-localized ERK activity set mitochondrial respiration, mitophagy, ATP-maintenance or permeability-transition endpoints rather than merely marking generic ROS injury?

Methods: PubMed, PubMed Central, prior Mito Map bundles, the reviewed candidate-selection dossier and six complete public primary PMC texts were checked on 2026-10-10. Endpoint classes were renal complex 1 respiration and ATP production, neuronal ERK2-linked mitophagy, macrophage ATP and membrane-potential maintenance, redox-compartment ERK phosphorylation, ERK5/NRF2 antioxidant response, and tumor-cell permeability-transition sensitivity. The analysis was a source-coded competing-model synthesis and measurement-rule analysis; no pooled effect size, raw-image reanalysis, clinical inference, treatment recommendation or wet-lab validation was performed.

Results: In oxidant-injured renal proximal tubular cells, MEK/ERK inhibition restored respiration, complex 1 activity and ATP production, while constitutively active MEK1 suppressed respiration in noninjured cells [1]. In SH-SY5Y neuronal cells, mitochondrial ERK2 activity was sufficient to recapitulate 6-OHDA-linked mitophagy and autophagic cell death [2]. Boundary evidence prevented a universal ERK-injury claim: ERK activity maintained ATP and mitochondrial membrane potential in human alveolar macrophages [3], redox state altered ERK phosphorylation by compartment in lung cells [4], complex 1 activity induced an ERK5/NRF2 antioxidant response without requiring de novo ROS [5], and mitochondrial ERK desensitized tumor-cell permeability transition through GSK-3 and cyclophilin D [6].

Conclusions: Current primary evidence supports a bounded discriminator rather than a new biological proof. Where compartment ERK is measurable, oxidant-linked mitochondrial endpoints should be interpreted with ERK localization, perturbation, viability, mitochondrial-content and endpoint-specific controls before being assigned to generic ROS injury. The claim would weaken if matched experiments showed no ERK-perturbation effect on respiration, mitophagy, ATP maintenance or pore sensitivity after ROS burden, viability, mitochondrial mass and off-target kinase controls were applied.

Keywords: ERK; mitochondria; oxidative stress; mitophagy; complex 1; permeability transition; source-coded synthesis

1 Introduction

Oxidant-stress mitochondrial studies often assign several events to the same biological bin: ROS exposure, ERK phosphorylation, impaired respiration, mitophagy, ATP loss, membrane-potential change and death signaling. That shorthand is useful for orientation, but it becomes a bottleneck when the research decision is whether ERK should be perturbed, localized or merely recorded as a stress marker. The selected question was therefore fixed before drafting: Under matched oxidant or mitochondrial-stress conditions, does compartment-localized ERK activity set mitochondrial respiration, mitophagy, ATP-maintenance or permeability-transition endpoints rather than merely marking generic ROS injury?

The central relationship evaluated here is deliberately narrow. In oxidant- or mitochondrial-stress cell systems where ERK compartment localization is measurable, mitochondrial or compartment-localized ERK activity changes mitochondrial respiration, mitophagy, ATP maintenance, or permeability-transition sensitivity independently enough that bulk ROS exposure alone cannot identify the mitochondrial outcome. The strongest competing account is ordinary and credible: bulk oxidative injury, cell type, viability state, or upstream stress severity explains the endpoint, while ERK phosphorylation or localization is only a downstream marker under matched conditions.

The contribution is not a claim that ERK is universally harmful or protective. The closest primary works each resolve one context: renal respiration after TBHP, neuronal ERK2 mitophagy, macrophage ATP maintenance, redox-compartment ERK phosphorylation, ERK5 antioxidant response, or tumor-cell PTP desensitization [1-6]. This paper asks what comparison those sources jointly force: whether future oxidant-linked mitochondrial studies need compartment-resolved ERK and endpoint-specific controls before assigning an outcome to generic ROS injury.

The distinct contribution is the direction-reversal comparison across ERK mitochondrial endpoint classes. Prior Mito Map working papers tested exercise redox ceilings, vesicle triage, UPRmt identity or lactate routing; this analysis instead compares ERK perturbation or localization against four endpoint classes in which ERK can suppress respiration, promote mitophagy, maintain ATP, prime antioxidant response or resist PTP opening. The resulting decision rule is therefore not a topic rotation of a generic caution framework. It is a falsifiable same-condition test: ERK adds endpoint information only if perturbation or compartment evidence moves a named mitochondrial endpoint after oxidant burden, viability and mitochondrial content are controlled.

2 Methods

The research question was fixed before drafting, and six public PMC full texts were archived for source verification. For manuscript drafting, each archived text was read against its public primary-source URL, DOI, PMID, PMCID and SHA-256 value. Current PubMed/PMC and web searches on 2026-10-10 were used to confirm that the selected sources remained public primary records; review articles were used only for orientation and were not counted as evidence records.

Eligibility required a primary source with ERK compartment localization, ERK activity, or ERK-pathway perturbation linked to a mitochondrial endpoint. The evidence set included six primary records and six endpoint classes: renal complex 1 respiration and ATP production, neuronal ERK2-linked mitophagy, macrophage ATP and membrane-potential maintenance, redox-compartment ERK phosphorylation, ERK5/NRF2 antioxidant response, and tumor-cell PTP sensitivity. No patient-level dataset, raw images, unpublished correspondence, proprietary archive or clinical-treatment dataset was used.

Each source was coded for tissue or cell model, ERK isoform or pathway, stress condition, perturbation or localization

measurement, mitochondrial endpoint, direction of ERK-endpoint relationship, role in the concept and interpretation limit. Table 1 is the native evidence matrix. Figure 1 summarizes the analysis process, and Figure 2 summarizes the competing-model discriminator.

Sensitivity analyses were labelled by role: robustness for direct perturbation sources [1,2], decision-changing for protective boundary sources [3,5,6], and exploratory for compartment measurement without endpoint perturbation [4]. These checks tested whether endpoint-specific ERK movement remained informative after generic redox-stress explanations were retained.

The analysis is a public full-text source-coded synthesis. It is not a pooled meta-analysis, raw-image reanalysis, animal-to-human translation, clinical validation, therapeutic recommendation or proof of biological truth. A machine-readable evidence and assumption ledger is provided in Supplementary File S1, with a readable companion in Supplementary File S2. The ledger records quotation checks, source-version hashes and unresolved premises; it does not establish biological or clinical validity.

The biological audit helper independently queried the local public-paper corpus for the PubMed source identifiers. The retained audit reports UNKNOWN source-retrieval status where that corpus did not return source text, and it reports UNREVIEWED interpretation status for every claim. Those statuses are preserved as limitations rather than converted into artificial support; the manuscript's scientific support comes from the archived public PMC primary texts, the source-coded evidence table and the cited primary articles.

3 Results

3.1 Evidence synthesis

The renal proximal tubular-cell source provides the strongest respiration endpoint. TBHP exposure increased ERK1/2 phosphorylation and was followed by decreases in basal and uncoupled respiration, complex 1-coupled state 3 respiration, ATP production and complex 1 activity [1]. MEK/ERK inhibition restored basal and state 3 respiration, ATP production and complex 1 activity, while inactive analog control and other pathway controls narrowed the interpretation [1]. Constitutively active MEK1 suppressed respiration even without oxidant injury [1]. This is the most direct support for ERK as endpoint-setting in one oxidant-injured epithelial context.

The same source also limits the claim. ERK inhibition did not restore aconitase activity, and the model was a rabbit renal epithelial culture with sublethal TBHP exposure [1]. Therefore, the result supports a complex 1-linked mitochondrial endpoint under a defined oxidant and cell system, not a generic rule that every ROS-linked mitochondrial defect is ERK mediated.

The neuronal source tested whether elevated ERK activity was sufficient to induce mitochondrial turnover in SH-SY5Y cells exposed to 6-OHDA [2]. Wild-type, constitutively active and kinase-deficient GFP-ERK2 constructs showed that the level of mitophagy was more tightly correlated with ERK activity and mitochondrial localization than macroautophagy alone [2]. The abstract conclusion that mitochondrial ERK2 activity was sufficient to recapitulate 6-OHDA effects on mitophagy and autophagic cell death supplies a second direct endpoint class [2].

This source does not decide whether ERK2 localization also governs complex 1 respiration, macrophage ATP maintenance or PTP resistance. It instead makes the measurement rule more precise: mitophagy cannot be assigned from ERK phosphorylation and ROS exposure alone when ERK activity and mitochondrial localization are separable.

Three boundary records materially changed interpretation. In primary human alveolar macrophages, MEK/ERK localized to

mitochondria and ERK inhibition caused early ATP depletion and mitochondrial membrane-potential loss [3]. That result contradicts the simple claim that ERK activation only damages mitochondria.

In tumor-cell models, mitochondrial active ERK desensitized the permeability transition pore through a GSK-3 and cyclophilin-D axis, protecting transformed cells from PTP-dependent death stimuli [6]. In an ERK5 source, complex 1 activity induced ERK5 expression and NRF2 antioxidant response through fumarate; this pathway was described as directly linked to OXPHOS and not requiring de novo ROS [5]. These sources do not refute the renal or neuronal findings. They define failure conditions for overgeneralization: ERK compartment, isoform, transformation state, endpoint and time window must be specified before endpoint direction is inferred.

The lung-cell redox source mapped ERK tyrosine and threonine phosphorylation across cytosol, mitochondria and nucleus under low and high H2O2 [4]. At low H2O2, cytosolic pTyr increased rapidly and mitochondrial pTyr was sustained; at high H2O2, double-phosphorylated ERK was retained in mitochondria [4]. This evidence supports the feasibility and importance of compartment measurement.

However, that source did not measure respiration, ATP production, mitophagy flux or PTP sensitivity as causal endpoints. In the sensitivity analysis, it therefore served as an exploratory

measurement boundary. It strengthens the case that bulk ERK phosphorylation is too coarse but cannot by itself validate mitochondrial endpoint causality.

3.2 Evidence-coded sensitivity analysis

Under the proposed concept, matched oxidant or mitochondrial stress should show endpoint-specific movement after ERK perturbation or compartment-localization evidence: renal respiration and complex 1 activity improve with MEK/ERK inhibition; neuronal mitophagy tracks mitochondrial ERK2 activity; macrophage ATP maintenance can require ERK; tumor-cell PTP sensitivity can decrease with mitochondrial ERK; and ERK5 can support antioxidant priming [1-6]. Under the alternative, those endpoints should track ROS burden, cell injury, cell type or mitochondrial content after ERK perturbation and localization controls are applied.

The six-source synthesis supports the research decision to require compartment-resolved ERK and mitochondrial endpoint controls before assigning oxidant-linked mitochondrial change to ROS damage, mitophagy activation or mitochondrial protection. A null or contrary result would be informative: if matched experiments showed no ERK-perturbation effect after ROS, viability, mitochondrial content and off-target kinase controls, ERK should be treated as a context marker rather than a mitochondrial fate gate.

Table 1. Primary-source evidence matrix for ERK-linked mitochondrial endpoint identity.

ID	Model and perturbation	Key finding	Interpretive limitation
[1]	RPTC + TBHP; ERK inhibitor or active MEK1.; Respiration, ATP, complex I.	Inhibition restored; active MEK1 suppressed.	Renal oxidant model only.
[2]	SH-SY5Y + 6-OHDA; ERK2 constructs.; Mitochondrial ERK2 and mitophagy.	Mitochondrial ERK2 drove mitophagy stress.	Mitophagy endpoint only.
[3]	Human alveolar macrophages; ERK inhibition.; ATP and membrane potential.	Inhibition depleted ATP and potential.	Maintenance boundary only.
[4]	Murine lung cells/tissue; low and high H2O2.; Compartment ERK phosphorylation.	Redox dose changed compartment ERK traffic.	Measurement source only.
[5]	OXPHOS models; complex-I/fumarate axis.; ERK5 and NRF2 response.	Complex I induced ERK5/NRF2 without new ROS.	ERK5 boundary only.
[6]	Tumor cells; mitochondrial active ERK.; PTP, GSK-3 and cyclophilin D.	Mitochondrial ERK desensitized PTP.	Tumor-survival boundary.

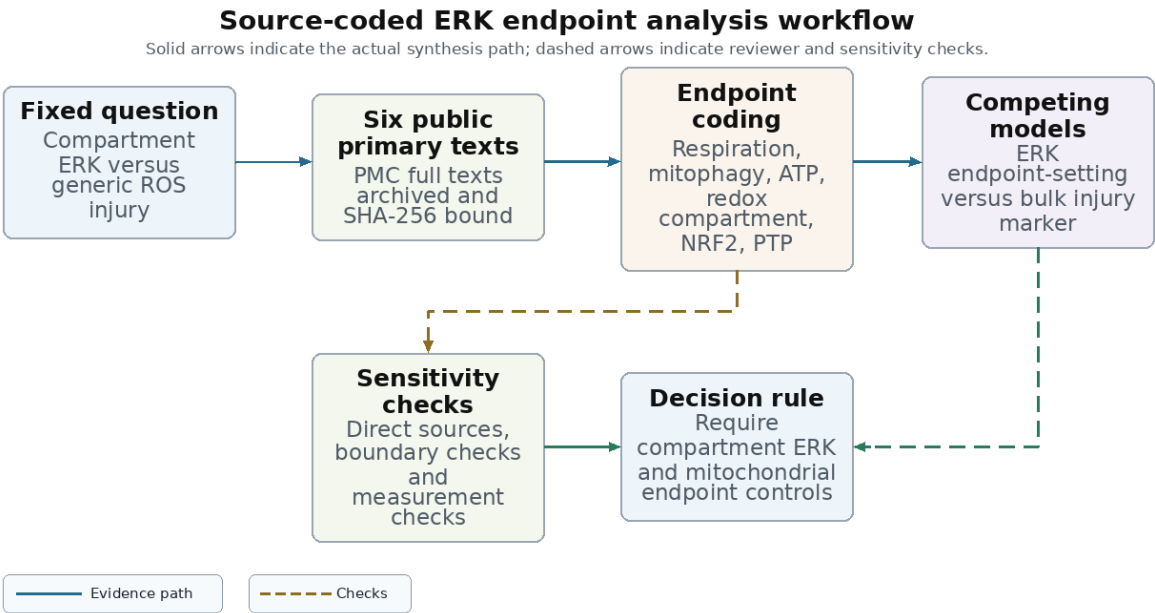


Figure 1. Source-bound ERK endpoint workflow. The workflow preserves the selected question, verifies six primary full texts, codes endpoint classes, contrasts ERK endpoint setting against generic injury, runs sensitivity checks and binds review artifacts.

Compartment ERK separates endpoint identities

Solid links are directly supported contexts; dashed links are proposed matched-condition discriminators.

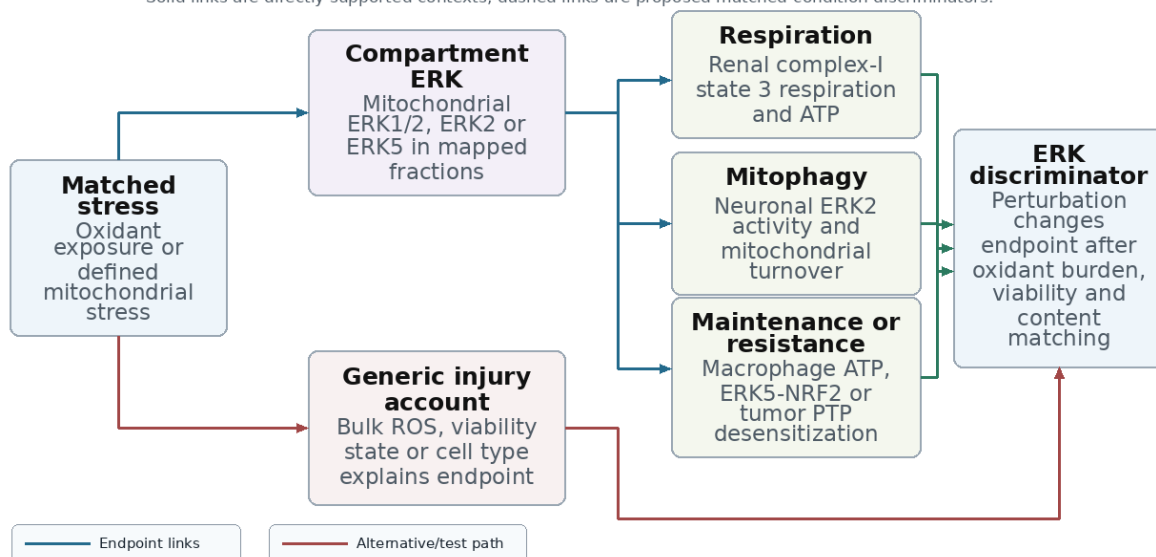


Figure 2. Compartment ERK endpoint discriminator. Solid arrows show endpoint links supported by the source set; dashed arrows show the generic-injury alternative and the proposed matched-condition discriminator.

4 Discussion

The evidence supports a discriminator rather than a universal mechanism. Direct support comes from two perturbation-rich contexts: ERK1/2 activation suppressed renal respiration and ERK2 activity/localization promoted neuronal mitophagy [1,2]. Boundary evidence is equally important because ERK activity can maintain macrophage ATP and membrane potential, support ERK5/NRF2 antioxidant response, or desensitize tumor-cell PTP [3,5,6]. A redox-compartment source shows that ERK phosphorylation traffic can differ by H₂O₂ condition and subcellular fraction [4].

This synthesis changes study design. Bulk ROS exposure, total phospho-ERK and a downstream mitochondrial endpoint are insufficient for endpoint assignment. A stronger design would match oxidant dose, time window, viability range, mitochondrial mass, substrate availability and assay conditions, then compare MEK/ERK inhibition, ERK activation, kinase-dead ERK2, compartment localization and negative kinase controls. The endpoint must be named: complex 1 respiration, ATP maintenance, mitophagy flux, antioxidant transcription, or PTP sensitivity.

The closest Mito Map papers address exercise redox adaptation, MDV cargo triage and ATF5-associated UPRmt identity rather than compartment ERK endpoint discrimination. This paper extends that portfolio by asking whether ERK localization and perturbation classify mitochondrial outcome under oxidant or mitochondrial stress, using a source set that does not overlap the prior selected mechanisms except at the broad redox and mitochondrial-quality-control level.

5 Limitations

This is a reanalysis-stage source-coded synthesis of public primary texts. It cannot reanalyze raw images, compute pooled effects, prove cell-type transfer, establish clinical efficacy, or validate the proposed discriminator experimentally. The source set spans renal epithelial cells, neuronal cells, alveolar macrophages, murine lung cells, hematopoietic/hepatic systems and tumor cells, so cross-context inference is intentionally bounded.

ERK1/2, ERK2 and ERK5 are not interchangeable. MEK inhibitors can have off-target or pathway-context limits, and source methods differ in stressor, timing, culture conditions, mitochondrial assays and viability state. The absence of a direct contradiction in one endpoint class is not support for universality. Contradictory and

protective contexts were retained because they prevent an overgeneral ERK-injury conclusion.

6 Conclusions

Compartment-resolved ERK signaling should be treated as an endpoint-identity discriminator in oxidant-linked mitochondrial studies when ERK localization, activity or perturbation can be connected to respiration, mitophagy, ATP maintenance, antioxidant response or PTP sensitivity. The current public primary evidence supports a bounded measurement and design rule: assign mitochondrial endpoints only after ROS burden, viability, mitochondrial content, compartment ERK and endpoint-specific controls are separated.

The practical research implication is selective rather than expansive. ERK perturbation is most informative when a mitochondrial endpoint can be measured in the same stress window, with the same viability range and with controls for mitochondrial amount and non-ERK kinase pathways. When those controls are unavailable, ERK phosphorylation should be reported as context rather than used to name the mitochondrial mechanism.

Declarations

Data and code availability. The repository deposit includes the manuscript source, evidence table, source data, analysis code, figures, measured figure geometry, biological-audit supplements, layout reports, QA record, Gemini review and checksums needed to replay the table, counts, figures and artifacts. Private replay inputs are retained locally and excluded from public manifests. Manuscript-quality and concept-impact grades are available at <https://precisionmito.com/paper-grades>.

Source verification. The biological audit is a manuscript-integrity ledger, not proof of biological truth or clinical efficacy. Corpus anchors and archived normalized source files were replayed against recorded hashes; UNKNOWN or NOT_RUN biological interpretation statuses remain visible.

Ethics statement. This literature synthesis used no identifiable patient data and required no new human-participant or animal research.

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AI assistance. AI tools assisted literature synthesis, drafting, data visualization, formatting and QA preparation. Gemini independently reviewed the exact rendered PDF when artifact-bound review was available. Andrew Friedl remains solely responsible for the content and is the sole author.

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