



(REVIEW ARTICLE)



RIBOSOME COLLISIONS AT THE HOST-PATHOGEN INTERFACE: EVIDENCE LINKING TRANSLATIONAL STRESS TO INNATE IMMUNE OUTCOMES

Ban D.H. Al Khayali *

Department of Biotechnology, College of Science, University of Baghdad, Baghdad, Iraq.

* Corresponding Author

International Journal of Science and Research Archive, 2026, 21(01), 561–583

Publication history: Received on 10 September 2026; revised on 17 October 2026; accepted on 19 October 2026

Article DOI: <https://doi.org/10.30574/ijrsra.2026.21.1.1912>

Copyright © 2026 Author(s) retain the copyright of this article. This article is published under the terms of the Creative Commons Attribution License 4.0

ABSTRACT

Ribosome collisions are increasingly invoked in infection biology, but translational arrest, physical collision, stress-kinase activation and host protection are often treated as interchangeable observations. This critical narrative review asks when translation damage is causally linked to innate-immune outcome. Targeted PubMed searches updated through 13 October 2026 prioritized primary studies with direct disome or structural evidence, pathway perturbation, genetic epistasis and infection or tissue endpoints. We distinguish pausing, stalling and physical collision, then integrate EDF1-GIGYF2-4EHP initiation control, ZNF598-ASCC/RQT and PELO-HBS1L-ABCE1 rescue, NEMF-LTN1 nascent-chain disposal, and ZAK α - and GCN2-dependent signalling. Antiviral effectors can generate stall- and collision-prone states: OAS-RNase L leaves translated cleavage products that form terminal stalls and, where ribosome density permits, collisions, whereas viperin-derived ddhCTP induces a ZAK-GCN2-dependent translational brake in defined flavivirus models. Pathogens can damage rRNA or eEF2, engineer structured pause sites, suppress ribosome influx or exploit quality-control-mediated recycling. Across these settings, collision is neither intrinsically antiviral nor synonymous with inflammation. We therefore separate causal confidence from biological relevance in a two-axis C0-C4/R0-R3 framework and use a traffic-and-threshold synthesis in which ribosome influx, lesion lifetime, rescue capacity, cell lineage and coincident pattern-recognition signals shape repair, adaptation, inflammatory alarm or cell death. Current evidence establishes collision-dependent immune signalling in only a limited set of systems; in many others, collision remains inferred from stall- or stress-associated readouts. Ribosome collision is therefore not a pathogen-specific receptor but one physical state within a broader translation-surveillance network that can couple damage to stress and innate-immune outputs.

Keywords: Ribosome Collision; Innate Immunity; Ribotoxic Stress Response; ZAK α ; RNase L; Viperin; Ribosome-Associated Quality Control; NLRP1; Infection; Translational Surveillance

1 INTRODUCTION

Pattern-recognition receptors identify conserved microbial nucleic acids, lipids and cell-wall products. This ligand-centred view is indispensable, but it is incomplete. Hosts also detect the consequences of virulence: membrane rupture, cytoskeletal disturbance, ion imbalance, organelle injury and interruption of core biosynthetic processes. Such activity-based surveillance has two advantages. It can report pathogenic function even when microbial molecules are concealed,

and it can integrate heterogeneous insults that converge on the same vulnerable host machinery. Its disadvantage is specificity: the same disturbance can arise from sterile stress, drugs or endogenous damage. The biological meaning of the signal must therefore be inferred from magnitude, duration, cell type and coincident receptor inputs rather than from its mere presence.

The translating ribosome sits at this boundary. Viruses depend on host ribosomes, many bacterial toxins target translation, and interferon-stimulated effectors remodel RNA and nucleotide pools. A pause becomes a collision only when a trailing ribosome physically encounters a delayed leader. The composite interface favours recruitment or activation of several surveillance factors, but it is not an absolute exclusivity filter: ZAK α also monitors stalled ribosomes, and ZNF598 can modify non-collided complexes in reconstituted systems. Foundational mammalian studies established collision-responsive small-subunit ubiquitination and stress signalling [1, 2], while structural and biochemical work has resolved how RACK1, ZNF598 and ZAK α recognize partly overlapping stalled and collided states [3–5]. Together, these studies established stalled and collided ribosomes as substrates for quality control and stress signalling.

Table 1: Representative prior reviews and the distinct scope of the present synthesis

Prior review	Principal scope	Coverage boundary	Distinct contribution here
Kim and Zaher [15]	Collisions as platforms for quality control, stress signalling and cell-fate regulation.	Cellular homeostasis is central; infection is not the organising framework.	Applies collision surveillance specifically to host–pathogen biology and separates physical collision from generic translation stress.
Iyer et al. [16]	Eukaryotic and bacterial rescue, degradation and stress-response pathways triggered by collisions.	Immune outputs, authentic infection and paired pathogen–tissue endpoints are not the main focus.	Integrates mammalian immune outputs and cross-kingdom pathogen lesions within one causal framework.
Rozman et al. [17]	Viral control of host translation and host translational defences, with RQC as one component.	Collision geometry, bacterial toxins and collision-specific causal tests are secondary.	Makes collision the organising intermediate and extends analysis to toxins, rescue geometry and inflammasome signalling.
Karasik and Gudyosh [18]	RNase L biology, including cleavage-generated stalling or collision and ZAK α signalling.	One host antiviral pathway is examined in depth.	Compares RNase L with viperin, bacterial toxins, viral recoding and RQC exploitation.
Vind et al. [19]	ZAK α -dependent ribotoxic stress and its links to apoptosis and pyroptosis.	Cell death is central; the wider competition among repair, adaptation and pathogen benefit is not.	Places cell death within competing RQC, GCN2, ZAK α , inflammasome and pathogen-supportive outcomes.
Chang and Choe [20]	Causes of stalling and RQC, ISR and RSR pathways in disease and therapy.	Infection-specific pathway ordering and paired host–pathogen outcomes are not central.	Adds infection-specific evidence calibration and requires both pathogen-control and host-injury endpoints.
Masucci [21]	Antiviral and proviral roles of RQC and viral reprogramming of translational stress.	Virus–RQC interactions are central; host-programmed and bacterial lesions are outside the main scope.	Extends the virus–RQC framework to RNase L, viperin and bacterial effectors, with threshold and intervention logic.

Representative rather than exhaustive comparison. RQC, ribosome-associated quality control; ISR, integrated stress response; RSR, ribotoxic stress response.

Mechanistic links to innate-immune pathways have been established in defined systems. OAS–RNase L activation causes RNA cleavage, terminal stalling and ZAK α -dependent ribotoxic-stress signalling, although the contribution of collisions and the net antiviral value of ZAK α remain context-dependent [6–8]. Viperin-derived ddhCTP restricts Zika and Kunjin viruses through a collision-associated ZAK–GCN2 response [9]. eEF2-inactivating bacterial toxins activate ZAK α –p38–NLRP1 signalling in human epithelial models, but direct collision evidence is unavailable for every toxin model [10–12]. Conversely, vaccinia protein synthesis depends on ZNF598/uS10 ubiquitylation, and programmed viral frameshifting can engage a proviral GCN2 programme [13, 14]. Stall- and collision-associated responses can therefore be antiviral, proviral, inflammatory or neutral depending on context.

Previous reviews have established collided ribosomes as surveillance platforms, defined rescue and stress-signalling branches, or examined particular infection settings such as viral translational control, RNase L activation and the virus–RQC arms race [15–21]. The representative comparisons in Table 1 show a persistent gap: these syntheses do not apply one causal standard across host-programmed RNase L and viperin responses, toxin-induced ribosome injury, viral recoding, viral quality-control dependence, inflammasome activation and therapeutic intervention. They also predate or only partly incorporate several 2024–2026 mechanistic studies that directly connect RNase L cleavage, ZAK α structure, programmed viral frameshifting and macrophage signalling to collision-associated outcomes.

This review addresses that gap by integrating host-built and pathogen-built collision routes within a single evidence-calibrated framework. A fall in translation, phosphorylation of p38 or eIF2 α , or induction of cytokine transcripts does not by itself prove that collided ribosomes were the intermediate. Four questions therefore organize the synthesis: what physical state counts as a collision; how is that state triaged between repair and signalling; when does collision improve pathogen control rather than host injury; and which experimental tests are required before therapeutic targeting is justified? The distinctive contribution is not another catalogue of collision factors, but a host–pathogen decision framework that couples molecular mechanism to paired pathogen and tissue outcomes.

1.1 Scope and evidence strategy

This is a critical narrative review, not a systematic review or meta-analysis. Targeted PubMed searches were updated through 13 October 2026. Search concepts combined terms for collision architecture and rescue, stress signalling, innate immunity and infection, host antiviral effectors, pathogen toxins and translational intervention; the reproducible concept blocks are reported in Table 2. No lower publication-year limit was imposed because the field depends on foundational ribotoxic-stress and rescue studies, although mechanistic coverage was concentrated on 2017–2026. Reference lists of pivotal primary studies and recent reviews were searched backward, and related-article searches were used to identify subsequent mechanistic work.

Primary studies were eligible when they addressed ribosome pausing, stalling, collision, rescue or collision-responsive signalling and reported a mechanistically interpretable readout. Priority was given to direct disome or trisome footprints, toeprinting, structural evidence, collision-dependent ribosomal ubiquitination, defined stall reporters, genetic epistasis and wild-type versus defective rescue. Infection, toxin, primary-cell, organoid and *in vivo* studies were emphasized when they connected the molecular event to pathogen yield, cytokine protein, tissue function or genetically resolved cell fate. Reviews were used to position the field and to locate primary evidence rather than as substitutes for mechanistic data. Studies based only on global translation loss, phospho-p38, phospho-eIF2 α , transcript induction or single-inhibitor experiments were retained as contextual evidence but were not treated as proof of collision causality.

For each central study, the synthesis charted the initiating lesion, experimental system, collision or stall readout, perturbed sensor or rescue factor, downstream immune output, pathogen endpoint, host-injury endpoint and principal limitation. Human and mammalian evidence was emphasized. Yeast, bacterial and purified-system findings were retained when they established conserved principles, but the organism and assay boundary is stated explicitly. Full text was checked when accessible; otherwise, claims were restricted to information supported by the abstract, figures or verified bibliographic record.

Evidence was appraised at the level of each mechanistic claim rather than by assigning one score to an entire paper. Causal confidence and biological relevance were recorded separately. C0 denotes contextual evidence only, such as reduced translation or kinase activation without a collision measurement. C1 denotes one collision-associated signature without selective perturbation. C2 requires direct physical evidence of collision or two orthogonal collision-proximal readouts while distinguishing stalling from collision. C3 requires a manipulation expected to alter collision formation or clearance that changes the proposed downstream output with global-translation and relevant off-target controls. C4 requires temporal ordering plus pathway-specific genetic epistasis and rescue. Biological relevance is annotated independently as R0 for reconstituted or heterologous reporter evidence, R1 for an established cell line or defined non-infectious stressor, R2 for authentic infection or a native toxin or effector in a relevant primary-cell or organoid model, and R3 for *in vivo*, tissue or clinical evidence with a disease-relevant functional outcome. For infection or therapeutic claims, the strongest R3 designation additionally requires paired pathogen-burden and host-injury or host-function endpoints. These categories are an interpretive reporting framework, not a validated risk-of-bias instrument or numerical score (Tables 2–4).

Table 2: Reproducible search concepts and evidence handling

Component	Operational method	Purpose and safeguard
Coverage	Targeted PubMed searches updated through 13 October 2026; no lower year limit. Backward citation chaining and related-article searching supplemented the database search.	Retains foundational RSR and rescue studies while emphasizing 2017–2026 mechanistic evidence.
Collision and rescue	("ribosome collision" OR <i>disome</i> OR <i>trisome</i> OR "ribosome stall") AND (ZNF598 OR ASCC3 OR RQT OR PELO OR HBS1L OR GIGYF2 OR "ribosome quality control")	Captures physical collision, local initiation control, splitting, rescue and downstream RQC.
Stress signalling	("ribosome collision" OR "ribosome stall" OR "translation stress") AND (ZAK OR MAP3K20 OR "ribotoxic stress" OR GCN2 OR EIF2AK4 OR NLRP1 OR cGAS)	Separates collision-linked RSR, ISR, inflammasome and co-sensing claims.
Infection and toxins	("ribosome collision" OR "ribosome stall" OR "ribotoxic stress") AND (virus OR viral OR bacteria OR toxin OR infection OR "innate immun")	Identifies host–pathogen models rather than relying on drug-induced stress alone.
Host effectors	("RNase L" OR RNASEL OR viperin OR RSAD2 OR ddhCTP) AND (ribosome OR translation OR collision OR stall)	Captures host-programmed lesions that convert innate sensing into translational stress.
Translational intervention	(ZAK OR MAP3K20 OR RNASEL OR GCN2 OR EIF2AK4 OR ZNF598 OR ASCC3) AND (inhibitor OR agonist OR therapeut* OR organoid OR "in vivo")	Tests whether a molecular checkpoint has disease-relevant evidence and exposes off-target or timing risks.
Eligibility and charting	Mechanistic primary studies were charted for lesion, model, collision readout, perturbation, pathway order, immune output, pathogen endpoint, host-injury endpoint and main limitation.	Direct evidence and genetic ordering were weighted above translation loss, phosphoproteins or single-drug associations.

The searches support a critical narrative review. They are not presented as an exhaustive systematic search and no PRISMA flow or pooled effect estimate is claimed.

Table 3: Two-axis framework for claim-level evidence calibration

Code	Minimum evidence	Permitted interpretation
C0	Translation change, kinase activation or another contextual signal without a collision measurement.	Context only; do not attribute the output to collision.
C1	One stall- or collision-associated signature without route-selective perturbation.	Consistent with a stall/collision mechanism; hypothesis-generating.
C2	Direct physical collision evidence or two orthogonal collision-proximal readouts, with stall distinguished from collision.	The claimed ribosome state occurred in the tested model.
C3	Manipulating ribosome influx, lesion formation or rescue changes the output, with global-translation and relevant off-target controls.	The ribosome state is causally required for the nominated output.
C4	Temporal ordering plus pathway-specific genetic epistasis and wild-type versus sensor- or catalytic-defective rescue.	An ordered collision-to-pathway mechanism is supported.

Biological relevance is reported separately: R0, reconstituted or heterologous reporter; R1, established cell line or defined non-infectious stressor; R2, authentic infection or native toxin/effector in a relevant primary-cell or organoid model; R3, in vivo, tissue or clinical evidence with a disease-relevant outcome. High relevance cannot compensate for weak collision causality.

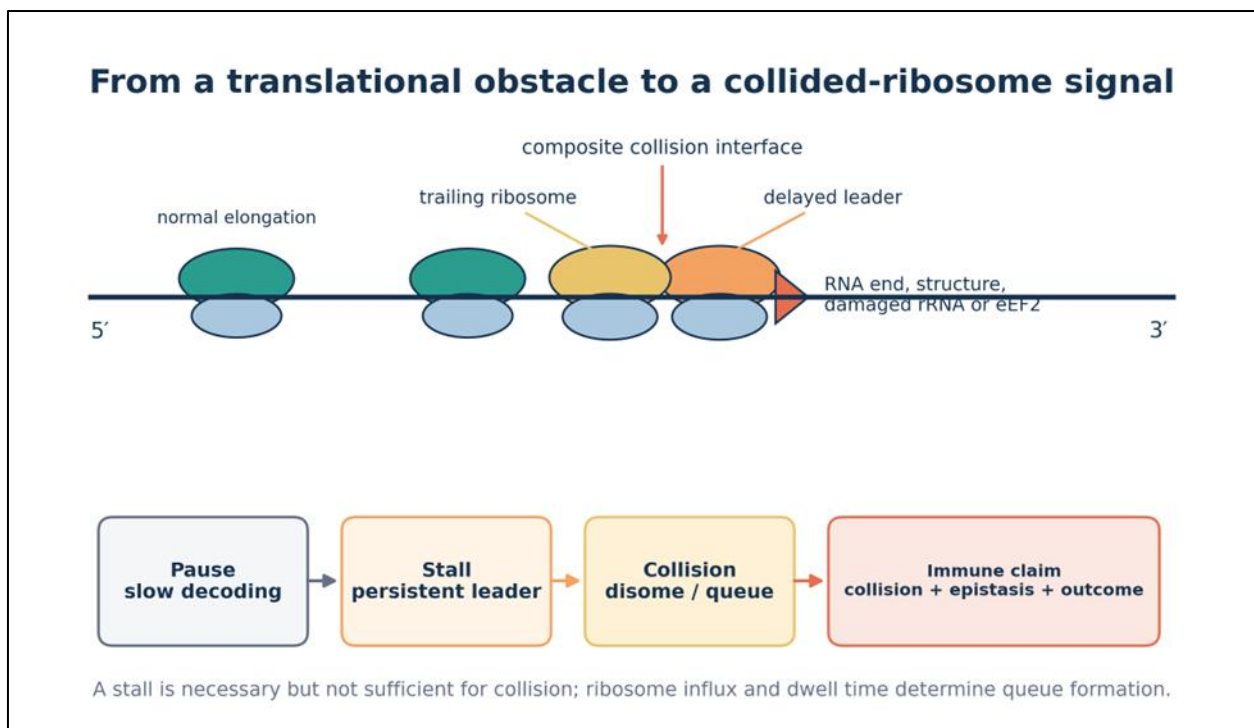


Figure 1: From a translational obstacle to a collided-ribosome signal. A pause becomes a stall when the leading ribosome remains delayed; a collision requires a trailing ribosome to reach that leader. An immune claim additionally requires pathway ordering and a biologically meaningful outcome.

Table 4: Operational definitions and assays for collision-centred immune claims

State	Operational definition	Preferred evidence	Common inference	false
Pause	Transiently reduced elongation rate at a codon or sequence context.	Monosome profiling with dwell-time or toeprinting support.	A local footprint peak is assumed to be a stable stall.	
Stall	A sustained translational delay detectable over the relevant experimental timescale; if unresolved, it may recruit rescue machinery or become a terminal complex.	Defined reporter, cleavage-end mapping, toeprinting or structural evidence.	Every stall is assumed to recruit a trailing ribosome.	
Collision	A trailing ribosome physically contacts a delayed leader to form a disome or longer queue.	Disome/trisome profiling, cryo-EM or two orthogonal collision markers.	Reduced puromycin incorporation or polysome compression is treated as direct proof.	
RQC activation	Ubiquitin-dependent recognition and splitting of a problematic ribosome, followed where applicable by 60S-associated nascent-chain disposal.	ZNF598-dependent eS10/uS10 ubiquitin plus ASCC/RQC epistasis; demonstrate collision separately if claimed.	Ribosomal ubiquitin is assigned to collisions without linkage, site or genetic controls.	
Ribotoxic stress response	ZAK α -dependent p38/JNK signalling from damaged or collided ribosomes.	ZAK loss and rescue with kinase- or ribosome-binding-defective variants.	p38/JNK activation alone is labelled collision-specific.	
Integrated stress response	GCN2 or another eIF2 α kinase reduces initiation and permits selective stress translation.	GCN2/eIF2 α genetics, ATF4 output and collision manipulation.	Phospho-eIF2 α is attributed to collisions	

			despite uncharged-tRNA or other inputs.
Immune outcome	Collision-dependent change in pathogen control, cytokine protein or genetically resolved cell fate.	Temporal ordering, epistasis and pathogen plus tissue endpoints.	Cytokine transcripts or viability loss are treated as sufficient.

Abbreviations: ISR, integrated stress response; RQC, ribosome-associated quality control; RSR, ribotoxic stress response.

1.2 Collision architecture coordinates translational quality control and ribosome-associated stress signalling

A ribosome collision is not simply a slow ribosome. It is a higher-order state produced when a trailing ribosome physically catches a leading ribosome whose elongation, termination, or recycling has become sufficiently slow. Collision probability therefore depends on both the duration of the delay and the initiation-dependent spacing of ribosomes on the message. Modelling and reporter experiments in mammalian systems indicate that a modest pause can become collision-prone on a densely loaded transcript, whereas the same pause may be tolerated when initiation is sparse [1]. Causal lesions include damaged or endonucleolytically cleaved RNA, premature polyadenylation, difficult nascent-chain sequences, inhibitory metabolites and drugs, defective termination or recycling, and pathogen-derived toxins that disable elongation factors [22, 23]. Two adjacent ribosomes form a disome; recruitment of another trailing ribosome produces a trisome, and continued loading generates a queue. Nuclease digestion preserves approximately two- or three-ribosome footprints, permitting disome- and trisome-profiling. Mammalian profiling has mapped collisions at inefficient elongation, termination, and recycling contexts and has distinguished discrete elongation states [24–26], whereas genome-wide trisome profiling and much of the genetic logic linking queue length to quality control were established in *Saccharomyces cerevisiae* [27]. Thus, trisomes are a useful measure of collision persistence, but mammalian mechanistic claims should not be inferred from yeast profiling without qualification.

Cryo-electron microscopy shows why a collision is a qualitatively new signal. In rabbit and human systems, the two 40S subunits create a reproducible composite interface that is absent from isolated translating ribosomes. RACK1 on the leading ribosome lies near small-subunit proteins of the trailing ribosome, while eS10/RPS10 and uS10/RPS20 are positioned at or close to this interface [1, 3]. Human and yeast disomes are not structurally identical, although both expose a geometry that can be decoded by the ZNF598/Hel2 ubiquitin ligase system; the yeast-specific structural and genetic evidence therefore supports conservation of a principle rather than identity of every contact [28]. In situ cryo-electron tomography of mammalian cells under persistent collision stress further showed that native interfaces can extend beyond the 40S–40S contacts captured in reconstituted systems and can include the L1 stalk and eEF2 [29]. A disome peak should consequently be interpreted as a structural ensemble, not a single immutable conformation.

In mammalian cellular reporter systems, ZNF598 is the best-established collision-responsive 40S ubiquitin ligase. Loss-of-function studies, stalling reporters and structural analysis show preferential action at collided rather than merely slow ribosomes and modification of eS10/RPS10 and uS10/RPS20 [1, 30–32]. Yeast collision genetics and ubiquitin mapping established the same organizing principle through Hel2-dependent quality control [33, 34]. RACK1 is integral to the collision surface and supports quality-control signalling, but it is a ribosomal scaffold and geometry determinant rather than the catalytic sensor. Early mammalian studies emphasized mono-ubiquitylation of eS10 and uS10; later reconstitution with human collision complexes demonstrated K63-linked polyubiquitylation of uS10 as a signal for human RQT-mediated subunit dissociation [3]. Single-molecule imaging further indicates that ZNF598 abundance can be rate limiting for resolving collided ribosomes [35]. The substrate, ubiquitin-chain architecture and extent of modification vary with reporter, collision persistence and assay. Purified systems also show that a preassembled collision is not strictly required for ZNF598-mediated ubiquitination or ASCC-mediated disassembly under every artificial condition [36]. Collision geometry is therefore a strong selectivity and efficiency determinant, not an absolute biochemical prerequisite.

Ubiquitylated collided ribosomes must be separated before the incomplete polypeptide can be eliminated. In mammalian cell-free reconstitution, the ASC-1 complex—ASCC3 helicase with ASCC2 and associated subunits—binds ubiquitin-marked complexes and uses ATP to split the leading ribosome. This reaction requires ZNF598-dependent 40S ubiquitination but does not require PELO–HBS1L [37]. Human RQT reconstitution similarly supports an ASCC3-centred splitting mechanism responsive to K63-linked uS10 chains [3]. PELO, HBS1L, and ABCE1 perform a related but separable rescue reaction. Mammalian in vitro experiments showed that PELO–HBS1L recognizes vacant or stalled 80S complexes, particularly those with little mRNA downstream of the P site, and that ABCE1 supplies the ATP-dependent splitting activity [38]. This geometry is expected at truncated or cleaved messages and can arise after no-go decay, but PELO–HBS1L should not be presented as the universal first splitter of an intact collided disome. The strongest evidence supports partially overlapping entry routes: ASCC/RQT resolves ubiquitin-marked collision complexes, whereas PELO–HBS1L–ABCE1 is especially suited to ribosomes with an accessible A site and a short or absent downstream RNA segment.

Splitting yields a 60S subunit that retains peptidyl-tRNA. This is the substrate for ribosome-associated quality control (RQC). Mammalian structural and biochemical reconstitution established that NEMF binds the obstructed 60S and the P-site tRNA, prevents premature subunit rejoining, and stabilizes LTN1/Listerin beside the polypeptide exit tunnel. LTN1 then ubiquitylates accessible lysines in the nascent chain, committing it to extraction and proteasomal degradation [39, 40]. The peptidyl-tRNA linkage must also be hydrolysed: mammalian experiments support ANKZF1-mediated release of ubiquitinated products and PTRH1 activity on non-ubiquitinated substrates [41]. A second NEMF activity requires careful species annotation. Yeast Rqc2 can extend nascent chains with non-templated alanine and threonine residues ("CAT tails"); mammalian NEMF also supports C-terminal tailing, but mammalian products are often predominantly alanine and can be recognized by C-end-rule ubiquitin ligases [42, 43]. It is therefore misleading to treat the detailed composition and fate of yeast CAT tails as automatically conserved in human cells.

Collision control begins before irreversible RQC. Two mammalian studies converge on a local negative-feedback circuit in which GIGYF2 recruits the cap-binding repressor 4EHP/eIF4E2 to reduce further initiation on the defective mRNA. Hickey et al. [44] found that ZNF598 can recruit GIGYF2–4EHP, coupling collision recognition to shut-off of new ribosome loading. In parallel, collision proteomics identified EDF1 as an early, ZNF598-independent collision binder that stabilizes GIGYF2 at the affected message; the EDF1–GIGYF2 arm and ZNF598 arm became more prominent when the other was absent [45]. A parsimonious model is tiered triage: EDF1–GIGYF2–4EHP rapidly throttles initiation and may allow a transient queue to clear, whereas persistent collisions engage ZNF598, ubiquitin-dependent splitting, and nascent-chain destruction. However, the relative contribution of direct ZNF598 recruitment versus EDF1-mediated recruitment is reporter- and context-dependent. This distinction matters at the host–pathogen interface because GIGYF2–4EHP also regulates antiviral transcripts and can be co-opted by viral proteins—for example, SARS-CoV-2 NSP2 enhanced GIGYF2–4EHP-dependent repression of *Ifnb1* translation in one experimental study [46]. Such immune regulation does not, by itself, demonstrate that the targeted transcript first underwent a collision.

The same physical lesion can also be converted into kinase signalling. ZAK α , the long ribosome-binding isoform of MAP3K20, is the proximal sensor of the ribotoxic stress response (RSR). Human-cell experiments established that its C-terminal ribosome-binding modules contact 18S rRNA and that partially redundant sensing regions are required for activation after perturbation of the E site, peptidyl-transferase centre, or rRNA by ribotoxins [47]. More recent structural and cell-biological work refines this model. ZAK α dynamically monitors the mRNA-exit region through contacts involving RACK1, RPS27, and 18S rRNA; prolonged residence on stalled or collided ribosomes permits sterile- α -motif dimerization and activation-loop trans-autophosphorylation [4, 5]. Cryo-EM further places RACK1 at the collision-dependent activation interface and identifies SERBP1 as a competitor that restrains inappropriate ZAK activation [4]. These findings explain how RACK1 can support two distinct outputs—ZNF598-dependent quality control and ZAK α -dependent signalling—without implying that the pathways are identical.

Activated ZAK α drives MKK-dependent p38 and JNK signalling. The output is dose-, duration- and cell-type-dependent: transient signalling can reprogramme translation and inflammatory gene expression, whereas sustained ribotoxic stress can promote apoptosis or inflammatory cell death. In human keratinocytes, ZAK α and p38 phosphorylate a human-specific disordered region of NLRP1, licensing inflammasome activation and pyroptosis after ultraviolet or toxin-induced ribotoxic stress [10]. At a direct host–pathogen boundary, *Corynebacterium diphtheriae* toxin and related elongation-factor-targeting exotoxins activate the ZAK α –p38–NLRP1 axis in primary human keratinocytes [11]. During antiviral defence, OAS-generated 2–5A activates RNase L; RNA cleavage creates translation-defective templates with terminal stalls and, where ribosome density permits, collisions, alongside ZAK α -dependent p38/JNK activation, pro-inflammatory transcription and apoptosis [6, 8]. This connects a canonical nucleic-acid-sensing effector to ribosome-associated danger signalling without making ZAK α or cleavage-end footprints collision exclusive.

Translational lesions that generate stalls and collisions feed parallel, partially overlapping pathways rather than a single linear routing hub. EDF1–GIGYF2–4EHP and ZNF598–ASCC are most directly linked to collision recognition; PELO–HBS1L–ABCE1 is particularly relevant to terminal stalls with little downstream RNA; NEMF–LTN1 acts after 60S splitting; and ZAK α and GCN2 can also respond to ribosome states not established as collisions. Which pathway dominates depends on lesion geometry, ribosome density, persistence, cell lineage and surveillance-factor availability. Mechanistic inference therefore requires a validated readout for the ribosome state being claimed, route-selective perturbation and genetic rescue. Polysome compression, p38 phosphorylation or reduced translation alone cannot establish collision causality.

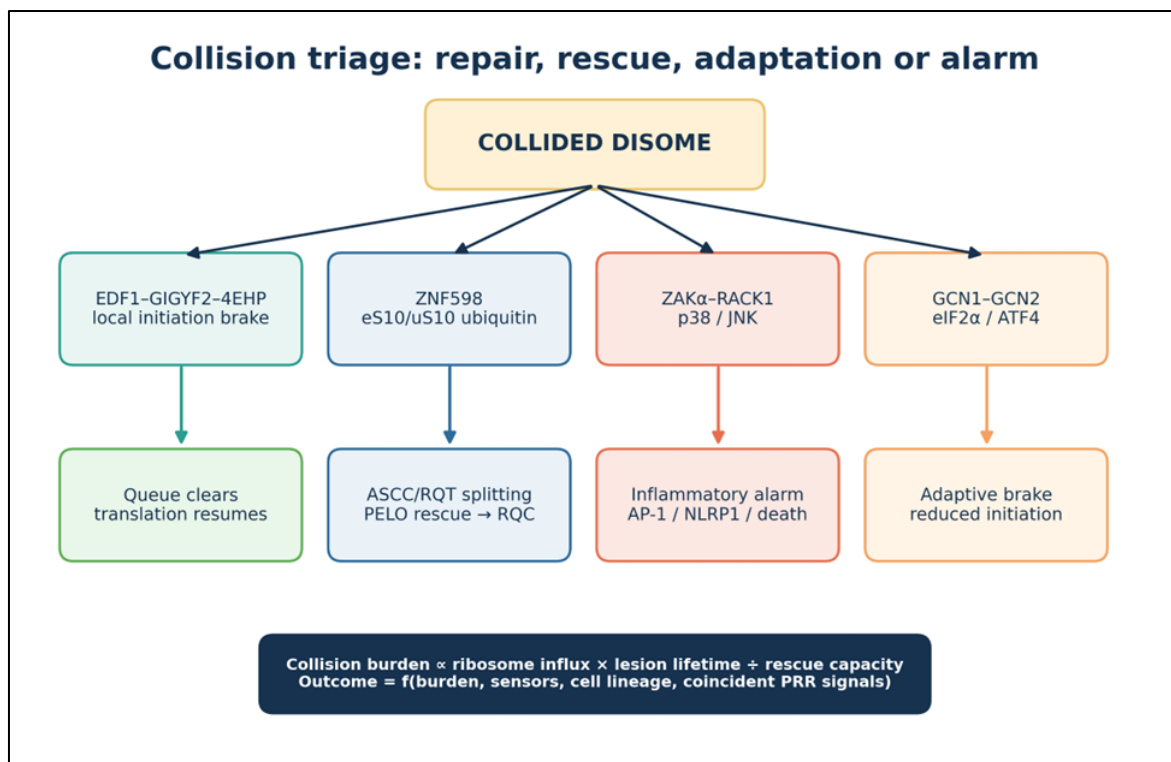


Figure 2: Collision triage. The collided disome can recruit an initiation brake, ubiquitin-dependent rescue machinery, ZAKα-dependent inflammatory signalling or GCN2-dependent translational adaptation. These branches overlap but are not interchangeable.

Table 5: Collision-responsive branches and their evidentiary boundaries

Branch	Immediate action	Best-supported evidence	Boundary
EDF1-GIGYF2-4EHP	Reduces new initiation on the affected mRNA.	Human collision proteomics and reporters [45, 48].	Recruitment route and persistence are reporter dependent.
ZNF598-eS10/uS10	Marks defective small subunits with ubiquitin.	Mammalian genetics, reconstitution and cryo-EM [1, 3].	Collision increases selectivity but is not an absolute prerequisite in every purified system.
ASCC/RQT	ATP-dependent splitting of ubiquitin-marked collided complexes.	Mammalian cell-free and human reconstitution [3, 37].	Not equivalent to PELO-mediated rescue.
PELO-HBS1L-ABCE1	Rescues accessible stalled 80S complexes, especially short or absent downstream RNA.	Purified mammalian systems [38].	Should not be presented as the universal first splitter of intact disomes.
NEMF-LTN1	Recognizes peptidyl-tRNA-60S and ubiquitinates the nascent chain.	Mammalian structural and biochemical reconstitution [39, 40].	Mammalian tailing cannot be equated uncritically with yeast CAT tails.
ZAKα-p38/JNK	Broadcasts persistent translation damage as RSR signalling.	Human cells, structures and primary epithelia [4, 5, 47].	Kinase output alone is not collision specific.
GCN2-eIF2α	Lowers initiation and permits ATF4-family stress translation.	Collision reporters and pathway genetics [2, 49].	GCN2 also responds to uncharged tRNA and ribosomal P-stalk signals.

cGAS co-sensing	Collided ribosomes enhance DNA-dependent cGAS activity.	RQC-factor depletion, ribosome association and in vitro activity [50].	Not a proven universal ZAK branch; DNA remains required.
-----------------	---	--	--

2 HOST-PROGRAMMED AND PATHOGEN-GENERATED STALL AND COLLISION STATES

2.1 RNase L links viral RNA sensing to ribosome-associated stress

The OAS–RNase L pathway provides the clearest connection between canonical viral sensing and collision-driven signalling. Double-stranded RNA activates oligoadenylate synthetases, which produce 2'–5'-linked oligoadenylates that activate RNase L [51]; OAS3 is a major RNase L activator across diverse viral infections [52]. RNase L then cleaves host and viral single-stranded RNAs. RNA destruction directly limits replication, but recent studies show that cleavage products also create abnormal templates on which translating ribosomes stall.

Two independent 2024 studies established the signalling arm of this response. Xi et al. [6] activated RNase L with 2–5A and found ZAK α -dependent activation of MAP2Ks, p38 and JNK, accompanied by inflammatory transcription and apoptosis. Karasik et al. [7] likewise found that RNase L- or ectopic RNase A-mediated fragmentation activated ZAK α –p38/JNK, supporting RNA cleavage as a sufficient upstream lesion. That study did not by itself distinguish terminally stalled monosomes from collisions. RNase L also suppressed translation of induced stress-response transcripts, including IFIT-family mRNAs and GADD34, showing that the pathway does not simply amplify all antiviral gene expression.

A subsequent study used nanopore sequencing and ribosome profiling to detect short 15–18-nucleotide footprints characteristic of ribosomes stalled at RNase L-generated RNA ends. Loss of the rescue factor PELO increased these footprints and augmented ribotoxic signalling [8]. Earlier work had shown that RNase L reprograms the translome, exposes non-coding reading frames and generates small immunostimulatory or tRNA-derived RNAs [53–56]. A supported sequence is therefore RNA cleavage → persistence of translation-engaged fragments → terminal stalling and, where ribosome density permits, collision → ZAK α -dependent signalling. The 2026 cleavage-end footprints establish stalling, not a disome in every case.

RNase L therefore couples direct RNA destruction to a secondary ZAK α -dependent stress response. Whether that second layer improves pathogen control or mainly contributes to cell death and tissue injury has not been established across infections. Recurrent viral acquisition of 2–5A phosphodiesterases demonstrates selection against the OAS–RNase L pathway as a whole, not specifically against its stall/collision-to-ZAK branch [57, 58].

2.2 Viperin imposes a collision-dependent antiviral brake

Viperin offers a mechanistically distinct interferon-induced route into ribosome stress. Its radical-SAM activity converts CTP into 3'-deoxy-3',4'-didehydro-CTP (ddhCTP), a naturally occurring antiviral ribonucleotide [59]. Hsu et al. [9] showed that viperin expression or delivery of the ddhCTP precursor ddhC increased collided ribosomes, required ZAK and GCN2 for eIF2 α phosphorylation and translation inhibition, and restricted Kunjin and Zika virus replication. Catalytically inactive viperin did not reproduce these effects, linking the response to ddhCTP rather than to viperin abundance alone.

ddhCTP activated a ZAK-dependent GCN2/eIF2 α response without detectable p38/JNK activation in the tested system, demonstrating branch selectivity but not its cause. Collision geometry, abundance or kinetics could favour GCN2 over SAPK activation, but the study did not establish a universal moderate-collision threshold or show that viperin limits host-cell death. Because ddhCTP also affects nucleotide metabolism and can act on viral polymerases, the ZAK–GCN2 route should be presented as one experimentally supported viperin mechanism rather than a universal explanation of viperin antiviral activity. In addition, ddhCTP can be misincorporated by mitochondrial RNA polymerase and suppress mitochondrial transcripts, a collision-independent effect that reinforces this qualification [60].

2.3 Bacterial toxins and epithelial stress signalling

Bacterial virulence factors generate lesions at several distinct points in elongation. Shiga toxins from *Shigella dysenteriae* and Shiga-toxin-producing *Escherichia coli* depurinate a conserved adenine in the sarcin–ricin loop of 28S rRNA, compromising elongation-factor engagement. Early work showed that ZAK inhibition or knockdown reduced Shiga toxin 2-induced p38/JNK signalling and IL-8 expression [61]. Because that study predated direct collision assays, it establishes a ZAK-dependent RSR after ribotoxic damage but not a collided-ribosome intermediate.

Diphtheria toxin, *Pseudomonas aeruginosa* exotoxin A and *Vibrio cholerae* cholix toxin act one step away from the ribosome by ADP-ribosylating the diphthamide residue of eEF2, irreversibly blocking translocation. Companion studies

in 2023 showed that this lesion activates ZAK α and p38, followed by phosphorylation and activation of the human NLRP1 inflammasome. Toxigenic *Corynebacterium diphtheriae* induced ZAK α /p38-dependent NLRP1 activation and pyroptosis in primary keratinocytes and reconstructed human skin. In airway and corneal epithelial models, *P. aeruginosa* exotoxin A was required for NLRP1 activation, IL-1-family cytokine release and barrier injury; exotoxin-deficient bacteria and ZAK α -deficient cells strongly reduced these outcomes [11, 12]. These infection and bacterial-mutant experiments provide stronger physiological support than toxin treatment alone.

Legionella pneumophila uses yet another strategy. Its SidI effector structurally mimics tRNA, binds and glycosylates the ribosome, and potentially inhibits elongation. Cryo-electron microscopy, biochemical assays and collision-sensitive profiling showed SidI-dependent stalling and collisions. During infection, SidI activated ZAK α and p38 and induced ATF3. ATF3 mRNA preferentially escaped the translational blockade, allowing ATF3 protein to enter the nucleus and activate a cell-death programme [62]. Whether this death response primarily protects the host or facilitates bacterial dissemination remains unresolved.

Together, these examples show that lesions in rRNA, eEF2 or the ribosome converge on translational arrest and ZAK α -dependent signalling, while SidI provides direct stall/collision evidence. It is not yet justified to assign a collided-ribosome intermediate to every toxin. Recent work further resolved Shiga-toxin-induced human cell death into NLRP1- and gasdermin-E-associated components [63]. The downstream NLRP1 connection is especially relevant to human barrier cells and should not be generalized uncritically to rodents, in which the equivalent human NLRP1 regulatory linker is not conserved.

2.4 Viruses exploit and avoid collision surveillance

Collision signalling is not intrinsically antiviral. Programmed –1 ribosomal frameshifting (PRF), used by many RNA viruses to expand coding capacity, deliberately slows ribosomes at a slippery sequence and structured RNA element. Xavier et al. [14] showed that a minimal SARS-CoV-2 PRF element generated collisions and activated GCN2 through IGF2BP3 and DRG1, independently of ZAK α . During infection, this PRF–GCN2 axis acted as an early switch that suppressed host translation and supported viral propagation in cells and human airway organoids. Similar dependence was demonstrated for HIV-1 and West Nile virus. Thus, viruses can convert a nominal stress response into a resource-allocation programme.

Disome profiling during HIV-1 infection provides complementary physical evidence. Kibe et al. [64] detected ribosome collisions in Gag–Pol upstream of the frameshift site and linked them to an RNA structural fold. That study localizes collision formation on authentic viral RNA, whereas the 2026 work supplies the causal GCN2 mechanism and replication phenotype.

Vaccinia virus illustrates exploitation of the repair branch. Infection increases ZNF598-dependent ubiquitination of uS10. ZNF598 loss or non-ubiquitylatable uS10 reduces viral protein production without a comparable reduction in viral transcripts, showing that vaccinia protein synthesis and replication depend on ZNF598 and ubiquitylatable uS10 [13]. Maintenance of ribosome availability through recycling is a plausible explanation, but the causal recycling step was not measured directly. RACK1 also supports poxvirus translation through collision-independent functions [65]. Conversely, host SAMD9 can be activated by poxvirus infection to cleave tRNA^{Phe}, causing phenylalanine-codon-specific pausing and translation arrest. Vaccinia K1 and C7 host-range proteins antagonize SAMD9/9L, illustrating viral evasion of a stall-generating defence [17, 66].

SARS-CoV-2 Nsp1 suggests a second avoidance principle: lower ribosome influx before queues form. Nsp1 occupies the 40S mRNA-entry channel and suppresses host initiation. SARS-CoV-2 RNAs show relative resistance in some experimental systems involving 5'-leader features, although the magnitude and mechanism of escape are context-dependent. In heterologous *Drosophila* and mammalian stress models, Nsp1 reduced collision-associated JNK signalling and rescued collision-driven toxicity [67]. It is plausible that initiation shutdown also limits trailing-ribosome arrival on damaged host mRNAs, but direct demonstration that Nsp1 suppresses ZAK α during authentic infection remains incomplete.

The collective evidence supports a traffic-based model rather than a simple host-versus-pathogen dichotomy. Cleavage, toxin damage, tRNA depletion and structured viral RNA all create slow or terminal ribosomes. Outcome is then determined by initiation flux, lesion persistence, rescue capacity, cell type and which sensor captures the resulting ribosome state. Future infection studies should measure monosome stalls, disomes, ZAK α , GCN2, ZNF598 and pathogen burden in the same time course; phosphorylation alone cannot establish that collisions were the causal intermediate.

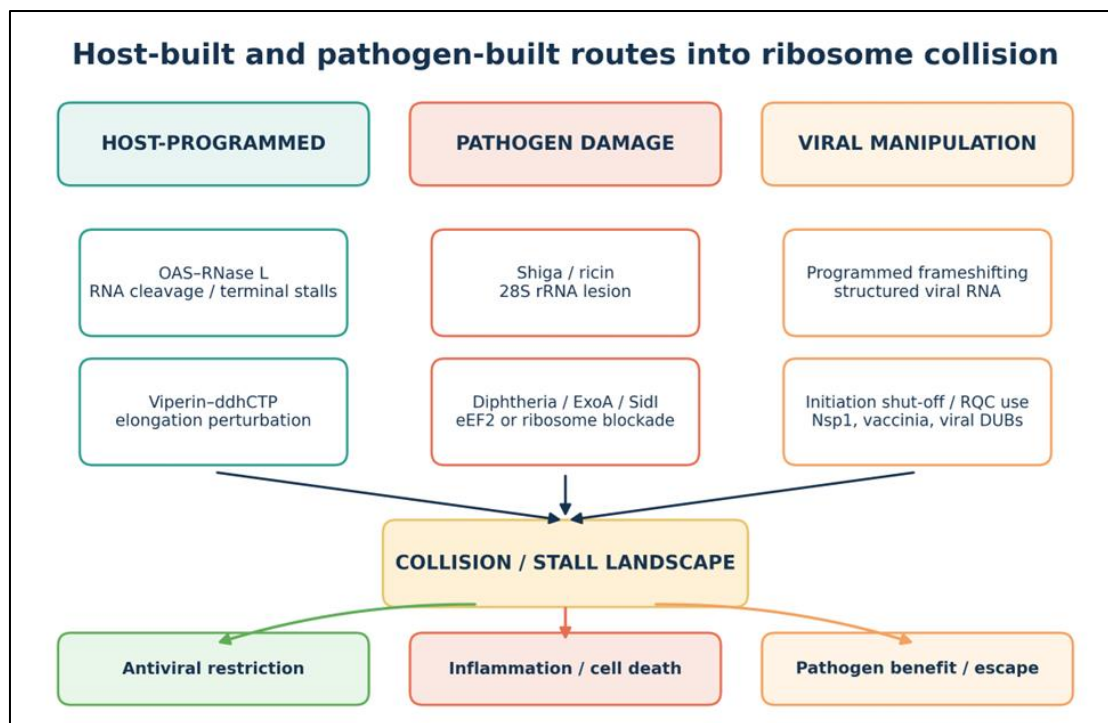


Figure 3: Host- and pathogen-derived routes to translational stalling and collision-prone states. Direct evidence for physical collision differs among routes; Table 6 summarizes supporting assays and limitations. Outcomes depend on lesion type, ribosome influx, rescue capacity and cellular context.

Table 6: Claim-level evidence matrix for representative host, pathogen and toxin routes

Trigger	Collision evidence	R	Observed output	Bounded interpretation
OAS–RNase L	C3: cleavage-end terminal stalls are direct; collision is density-dependent.	R1	ZAKα–p38/JNK, inflammatory transcription and apoptosis.	Strong stall-to-RSR chain; net antiviral value of ZAKα remains unresolved.
Viperin–ddhCTP	C3: increased collisions with catalytic and ZAK/GCN2 dependence.	R2	eIF2α phosphorylation, translation repression and flavivirus restriction.	Direct antiviral support in defined models; not a universal viperin mechanism.
Shiga toxin / ricin	C1: ribotoxic arrest and ZAK signalling; early studies predate disome profiling.	R1	p38/JNK and IL-8.	RSR demonstrated; collision intermediate remains inferred.
Diphtheria toxin / ExoA / cholix	C1: elongation arrest; collision not directly shown in every model.	R2	ZAKα–p38–NLRP1 and pyroptosis.	Strong human epithelial pathway ordering without universal collision proof.
<i>Legionella</i> Sidi	C2: direct structural, stalling and collision-sensitive assays.	R2	ZAKα–p38–ATF3 and cell death.	Collision supported; net host benefit versus dissemination unresolved.
Viral programmed frameshifting	C3: authentic viral RNA and reporter collisions with route perturbation.	R2	IGF2BP3/DRG1–GCN2 ISR and host translation suppression.	Collision-associated signalling can support replication.
Vaccinia virus	C1: ZNF598-dependent uS10 ubiquitylation; physical collision untested.	R2	RQC dependence of viral protein synthesis.	Proviral RQC use supported; recycling mechanism remains plausible, not measured.

SAMD9 during poxvirus infection	C1: codon-specific pausing; collision untested.	R2	Translation arrest opposed by K1/C7 proteins.	Host stall defence; collision extent requires measurement.
SARS-CoV-2 Nsp1	C1: reduced collision-associated toxicity in heterologous models.	R0–R1	Initiation shut-off and reduced JNK/toxicity.	Authentic-infection suppression of ZAK α remains unproven.

C0–C4 grade causal confidence; R0–R3 grade biological relevance. Grades apply to the specific claim summarized here, not to an entire paper or pathway.

3 COLLISION SENSING AND IMMUNE OUTCOME

Ribosome collision is not itself an immune outcome. It is a molecular state that can be interpreted by several partly overlapping pathways, and the balance among those pathways determines whether the cell adapts, enters an antiviral state, releases inflammatory mediators or dies. The best-established collision-proximal branch is the ribotoxic stress response (RSR), historically defined by stress-kinase activation after peptidyl-transferase inhibition or sarcin–ricin-loop damage [68], in which ribosome-associated ZAK α (MAP3K20) activates p38 and JNK. A second branch engages the GCN2–eIF2 α integrated stress response (ISR), which initially reduces new initiation events and therefore limits further collisions. A third, less mature connection links unresolved collided ribosomes to DNA-dependent cGAS–STING signalling. During infection, all three operate in cells that are simultaneously receiving signals from conventional pattern-recognition receptors. Consequently, a cytokine or interferon phenotype should not be assigned to “collision sensing” unless collision formation and the relevant collision sensor have both been causally tested.

3.1 ZAK α –p38/JNK signalling

ZAK α provides a mechanistic bridge between aberrant translation and stress-kinase signalling. Biochemical and selective ribosome-profiling experiments showed that ZAK α associates broadly with translating ribosomes but becomes strongly activated on stalled or collided complexes; ZAK deletion or disruption of its ribosome-sensing region abolishes stress-induced p38 and JNK phosphorylation in the tested systems [2, 47]. Time-resolved studies support stimulus- and dose-dependent partitioning between adaptive and death-associated outputs, but a universal sequence in which p38 is reparative and JNK is destructive has not been established. The safer conclusion is that p38 and JNK differ in kinetics and downstream programmes, with sustained JNK frequently associated with apoptosis in the tested models. Analysis of UV-damaged RNA defined regimes ranging from homeostasis through tolerance to apoptosis and showed that GCN2-mediated reduction of initiation and activation-coupled ZAK degradation provide negative feedback [69]. UV is not an infection model, but this work demonstrates that ribosome-state burden and duration can encode cell fate.

The inflammatory output of this arm is mediated most convincingly through p38/JNK-dependent activation of AP-1-family transcription factors and post-transcriptional control, including p38–MK2 effects on cytokine-mRNA stability. Classic ribotoxin experiments established that ZAK is required for Shiga toxin- and ricin-induced stress-kinase activation and pro-inflammatory cytokine expression [61]. Selective activation of RNase L with 2–5A gives this pathway direct relevance to signalling initiated by an antiviral effector: it produces ZAK α -dependent p38/JNK phosphorylation, AP-1-associated inflammatory transcription and apoptosis [6]. Ribosome profiling after RNase L or ectopic RNase A activation supported a cleavage-induced ribotoxic mechanism, while a later study directly mapped 15–18-nucleotide terminal-stall footprints at RNase L cleavage sites and showed PELO-sensitive rescue [7, 8]. The 2024 experiments did not alone prove collision as the intermediate or show that ZAK α improves viral clearance; the 2026 study strengthens the stall arm, while disome-resolved infection experiments remain necessary to establish the collision arm.

NF- κ B requires more cautious treatment. Viral dsRNA can activate RIG-I/MDA5–MAVS and other pathways that converge on NF- κ B, while STING can also engage an NF- κ B output. However, current collision-specific evidence is strongest for ZAK α –p38/JNK–AP-1, not for a universal direct ZAK α -to-NF- κ B route. During poly(I:C) treatment or infection, NF- κ B activation therefore cannot be attributed to collided ribosomes merely because ZAK, p38 or eIF2 α is phosphorylated. Such a claim requires loss of the NF- κ B readout after collision-selective manipulation and genetic ZAK α disruption, while intact signalling from the conventional PRR is shown in parallel.

3.2 GCN2–eIF2 α –ATF4 adaptation with antiviral potential

Collisions can engage the ISR. GCN2 phosphorylates eIF2 α , limiting eIF2B-dependent ternary-complex regeneration and suppressing bulk initiation; fewer initiating ribosomes reduce the probability that trailing ribosomes reach a stalled leader. Upstream open reading frames simultaneously permit preferential translation of selected stress mRNAs, particularly ATF4. GCN2 can respond to stalled translation, uncharged tRNA and the ribosomal P stalk, and it has antiviral activity in selected RNA-virus systems [70–73]. Recent structural and biochemical work showed that GCN1 senses di-ribosome states and couples GCN2 to activating ribosomal contacts, strengthening the physical basis for

collision-associated activation [74]. Wu et al. [2] found ZAK to be required for collision-induced GCN2 activation in their system, whereas RQC can antagonize this branch by resolving the collided state [75]. The relationship remains stimulus-specific: Misra et al. [49] showed collision dependence after elongation inhibition, whereas amino-acid depletion and UV can activate GCN2 through other routes. Phospho-eIF2 α is therefore not, by itself, evidence of collision sensing.

Viperin supplies a compelling antiviral example. Its radical-SAM product ddhCTP promotes ribosome collision, followed by ZAK- and GCN2-dependent ISR activation and inhibition of both host and viral translation; disrupting this pathway weakens restriction of flaviviruses including Zika and Kunjin viruses [9]. Yet translational repression can also compromise the immune programme it is meant to support. RNase L induces many stress and inflammatory transcripts but simultaneously reduces translation of selected antiviral IFIT mRNAs and of GADD34/PPP1R15A, a feedback phosphatase regulator that normally helps terminate the ISR [7]. Transcript induction therefore cannot be equated with cytokine or effector production. A rigorous analysis should pair RNA-seq with ribosome profiling or nascent-protein synthesis and direct measurement of secreted cytokines.

3.3 Inflammasome and cell death outputs

In human barrier epithelia, the strongest direct link from the RSR to an inflammasome is the ZAK α -p38-NLRP1 axis. Ribotoxic stress causes phosphorylation of the human-specific disordered NLRP1 linker by ZAK α and p38; mutation of critical linker residues prevents inflammasome assembly, gasdermin-D processing, IL-1 β /IL-18 release and pyroptosis [10]. p38-mediated phosphorylation of NLRP1 at Ser107 was independently placed upstream of N-terminal ubiquitination and degradation, which frees the UPA-CARD fragment to nucleate the inflammasome [76]. Portimine A further linked translation impairment to ZAK α -p38-NLRP1 signalling in keratinocytes and organotypic skin, although direct disome evidence was not reported [77]. Semliki Forest and Chikungunya virus activate the pathway in human keratinocyte models, although alphavirus-induced p38 is only partly ZAK α -dependent. F1L-deficient modified vaccinia Ankara produces greater translational disruption and ZAK α -dependent NLRP1 activation, consistent with F1L acting upstream of ZAK α ; the study did not identify a direct F1L target or directly measure collided ribosomes [78].

This branch is highly context restricted. Human NLRP1 contains regulatory features not conserved in rodent paralogues, and expression is enriched in skin and airway epithelia. It should not be generalized to all immune cells or all species. RNase L has also been linked to NLRP3 activation during viral infection [79], but that observation should not be conflated with the collision-ordered ZAK α -p38-NLRP1 mechanism. Moreover, apoptosis and pyroptosis must be separated experimentally: JNK-dependent apoptosis, NLRP1-caspase-1-gasdermin-D pyroptosis and nonspecific lysis can coexist after severe translational damage. Cleaved caspases, gasdermins, ASC specks, cytokine maturation and genetic deletion of the nominated death machinery are required to distinguish them.

3.4 cGAS-STING as collision co-sensing

In cells with chronic early-RQC deficiency or experimentally induced translation stress, collided ribosomes can potentiate DNA-dependent cGAS-STING signalling. Depletion of ASCC3, ZNF598 or RACK1 caused cytosolic redistribution of cGAS, cGAS-STING-dependent ISG expression and preferential association of cGAS with collided ribosomes. Purified ribosomes enhanced cGAS activity *in vitro*, but only in the presence of DNA [50]. This is best described as coincidence sensing: collided ribosomes alter cGAS localization and increase its DNA-dependent activity. It is not evidence that RNA or ribosomes replace DNA as the cGAS ligand, nor that cGAS lies downstream of ZAK α . No later primary study has yet established this route during authentic infection, so infection relevance remains incomplete.

3.5 Protection and immunopathology

Direct evidence that a collision-associated pathway improves pathogen control is strongest for viperin-ddhCTP-ZAK-GCN2 in the tested flavivirus systems. By contrast, RNase L-ZAK studies establish signalling and death after nuclease activation but generally do not isolate the contribution of ZAK α to viral clearance, and NLRP1 activation by toxins or viruses does not yet establish net benefit to the host or tissue. Viral antagonism by F1L is consistent with immune pressure but is not, by itself, proof of host protection. Antiviral, inflammatory and pathological outcomes should therefore be assigned separately and only when pathogen burden and host injury are measured in the same model.

3.6 Causal evidence standards and recurrent pitfalls

A defensible collision-to-immunity claim should satisfy four linked tests. First, demonstrate the ribosome state being claimed: a stall-resolved assay establishes stalling, whereas validated disome/trisome profiling, structural evidence or a direct collision reporter is required for collision. ZNF598-dependent uS10/eS10 ubiquitylation is supportive but not sufficient by itself because non-collided complexes can be modified in reconstituted systems. Second, alter collisions independently of global translation where possible. Third, establish pathway order with genetic epistasis and wild-type versus sensor- or catalytic-defective rescue. Fourth, measure the terminal phenotype at the correct level: infectious titre rather than viral RNA alone, secreted cytokine protein rather than transcript alone, and genetically resolved apoptosis or pyroptosis rather than nonspecific viability loss.

Several common designs fail these standards. UV, anisomycin, ricin and viral infection each create additional damage besides collision; single-stressor studies therefore require orthogonal collision generators. GCN2 can respond to uncharged tRNA and ribosomal P-stalk signals, so phospho-eIF2 α is non-specific. RQC-gene depletion may alter innate pathways independently of collision. Kinase inhibitors can be misleading: GCN2iB was recently shown to inhibit ZAK directly, which can falsely make GCN2 blockade appear protective by simultaneously suppressing the death-promoting RSR [80]. Finally, bulk measurements obscure the threshold behaviour revealed by single-cell and time-resolved studies. A strong host–pathogen experiment should therefore combine collision-resolved profiling, pathway-specific genetics, temporal measurements and both antiviral and tissue-injury endpoints.

3.7 Traffic-and-threshold framework for protection, pathology and pathogen benefit

A traffic-and-threshold framework can reconcile the literature by treating collision burden as an emergent property of ribosome traffic rather than a binary lesion. This is a hypothesis-generating synthesis, not an experimentally established universal ordering; thresholds and branch sequence may differ by lesion and cell type. Collision burden rises with initiation-dependent ribosome influx and lesion lifetime and falls with local initiation suppression and rescue capacity, including EDF1–GIGYF2–4EHP, ZNF598–ASCC, PELO–HBS1L–ABCE1 and downstream RQC. Sensor and effector availability differs among epithelial cells, immune cells and infected tissues, while coincident pattern-recognition signals change how the same translational state is interpreted (Fig. 4).

At lower burdens, local initiation suppression and rescue may dominate and limit proteotoxicity. In some stimuli, GCN2-dependent initiation control may reduce further trailing-ribosome arrival. Persistent or widespread damage can engage ZAK α –p38/JNK and, in susceptible human epithelia, NLRP1; severe unresolved damage can culminate in apoptosis or pyroptosis. Viperin and RNase L illustrate different portions of this spectrum, but they do not establish four universal, sequential thresholds [6–9].

Pathogens can benefit at any boundary. A virus that reduces initiation may prevent queues while suppressing host protein synthesis. A virus that depends on intense translation may exploit RQC-mediated ribosome recycling. Programmed frameshifting can create a pause whose stress output reallocates translation in favour of viral propagation rather than host defence [14]. Thus, the same collision-associated pathway can be protective early and harmful late, or antiviral in one cell type and proviral in another. The decisive experiment must measure pathogen yield and host-tissue injury together; neither an interferon signature nor a viability assay alone captures biological value.

This model also explains why pharmacological results are dose sensitive. Partial elongation inhibition may increase collision frequency by slowing leaders while allowing trailing ribosomes to continue. Near-complete arrest can instead reduce relative movement and prevent collision formation. Likewise, an inhibitor that suppresses p38 phosphorylation may reduce inflammatory injury while leaving unresolved stalled complexes and proteotoxic burden. Therapeutic interpretation therefore requires time-resolved measurements of collision state, pathway output, pathogen load and tissue function.

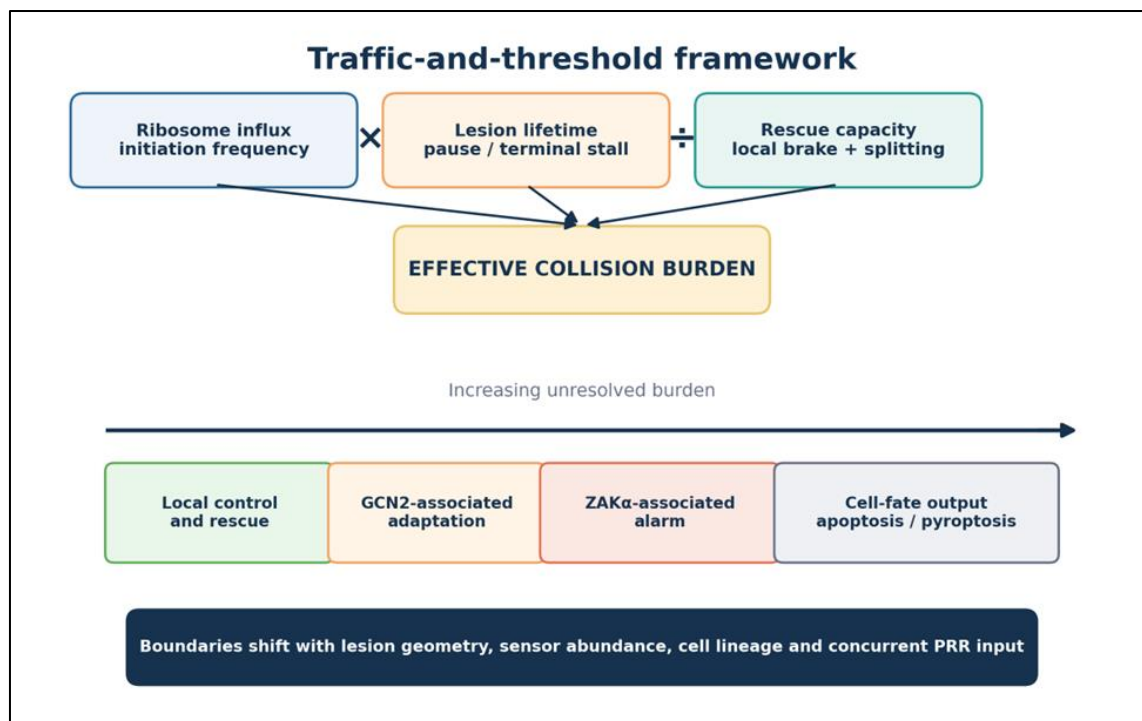


Figure 4: Traffic and threshold framework. Effective collision burden emerges from ribosome influx and lesion lifetime relative to rescue capacity. Increasing burden may favour local repair, GCN2-dependent adaptation, ZAKα-dependent alarm or terminal cell death, but the boundaries shift with cell lineage, sensor competence and coincident pattern-recognition signals. The sequence is a testable synthesis rather than a universal temporal programme.

4 THERAPEUTIC HYPOTHESES AND EVIDENCE REQUIREMENTS

Ribosome-state surveillance exposes several experimental intervention points between lesion and cell fate: the lesion-producing pathway, the ribosome-associated RSR sensor ZAKα, p38/JNK signalling, the GCN2–eIF2α integrated stress response (ISR), and ribosome-associated quality control (RQC). These nodes are not one linear pathway, and no collision-directed treatment or clinical biomarker is validated. Any proposed intervention must therefore be treated as hypothesis-generating and tested for both pathogen control and host-tissue function.

4.1 ZAKα–p38/JNK as an inflammatory and cell-death axis

ZAKα is a proximal, pharmacologically tractable experimental signalling node, although available inhibitors remain research tools and no selective ZAKα-directed therapy is clinically validated. Genetic and structural studies place ZAKα on stalled or collided ribosomes and show that its kinase activity is required for p38/JNK activation in the tested systems [2, 4, 47]. Preclinical evidence in sterile injury motivates short, tissue-restricted testing where persistent ribotoxic signalling is demonstrably pathogenic, but extrapolation to infection requires simultaneous measurement of pathogen burden and tissue function [81, 82].

The same biology imposes a major safety boundary. ZAKα–p38 can activate human NLRP1 and thereby couple translation damage to pyroptosis [10]; suppressing this arm during infection may preserve an infected cell that should be eliminated. Downstream p38 or JNK inhibitors are still less discriminating and may alter cytokine, differentiation and stress pathways unrelated to collisions. Candidate studies should therefore stratify by trigger and timing, quantify pathogen burden where relevant, and distinguish reduction of damaging host inflammation from loss of protective infected-cell death. At present, ZAKα inhibitors used in mechanistic studies remain research tools rather than clinically validated therapies.

4.2 RNase L as an antiviral amplifier with a narrow therapeutic window

RNase L is an upstream innate-immune entry point. Following OAS-dependent generation of 2–5A, RNase L fragments viral and host RNA. Two 2024 studies showed that RNase L- or RNase A-mediated fragmentation activates ZAKα–p38/JNK; those studies supported, but did not fully resolve, a stall/collision mechanism [6, 7]. A 2026 study directly mapped 15–18-nucleotide terminal-stall footprints at RNase L cleavage sites and showed PELO-sensitive rescue. It strengthened the stall-to-RSR chain but did not establish that every cleavage event produces a collision [8]. Human loss-

and gain-of-function genetics further position RNase L as a determinant of postviral inflammation and autoinflammation, underscoring that its clinical effect is not reducible to early cell-autonomous antiviral restriction [83].

Therapeutically, RNase L activation could amplify antiviral restriction, whereas partial inhibition could reduce bystander RNA destruction, inflammatory transcription and cell death. Neither direction is intrinsically beneficial. Agonism risks widespread host-RNA cleavage and tissue injury; inhibition risks prolonged viral replication. The appropriate unit of evaluation is therefore a defined virus–cell–time combination, not “antiviral activity” in the abstract. Experiments should pair viral titre or replication-competent output with host-cell survival, inflammatory mediators, RNA-cleavage products and collision measurements. A fall in cytokines alone cannot establish benefit if it reflects failure to clear infection.

4.3 ISR tuning is not collision suppression

The GCN2–eIF2 α arm decreases initiation and can reduce the arrival of trailing ribosomes while preferentially translating stress-adaptive transcripts. ISRIB restores eIF2B activity downstream of phospho-eIF2 α and is useful for probing whether persistent ISR output is causal [84], yet restoring initiation while elongation remains impaired could increase traffic into an unresolved stall. Conversely, blocking GCN2 may remove an initiation brake: in activated macrophages, GCN2 constrained ZAK α -dependent TNF hypersecretion, although that study did not directly measure ribosome collisions [85]. GCN2iB can also inhibit ZAK directly, so ISRIB, GCN2 inhibitors and eIF2 α -phosphatase modulators require distinct genetic and target-engagement controls [80].

4.4 RQC as a finite capacity clearance pathway

Collision-responsive quality control can shorten the lifetime of some queues by marking and splitting problematic ribosomes; downstream NEMF–LTN1 clears 60S-associated nascent chains. These are separable steps, and enhancing one need not increase overall rescue capacity. ZNF598-dependent ubiquitination is necessary for collision-responsive quality control in mammalian reporters, K63-linked uS10/RPS20 polyubiquitylation can recruit human RQT, and live-cell imaging indicates that ZNF598 availability can limit queue clearance [1, 3, 35]. No evidence supports systemic RQC enhancement or inhibition as a safe therapy. A testable alternative is context-specific modulation with direct evidence that queues are resolved rather than converted into a different stress state.

Table 7: Candidate intervention points and evidence required for evaluation

Intervention	Hypothesized benefit	Principal liability	Evidence required before translation
ZAK α inhibition	Reduce collision-proximal p38/JNK inflammation and cell death.	May preserve infected cells and blunt human NLRP1 defence.	Trigger-specific target engagement, pathogen burden, tissue function and timing.
p38/JNK inhibition	Limit inflammatory transcription or apoptosis.	Broad non-collision effects and impaired repair or antimicrobial responses.	Demonstrate a ZAK α -ordered collision mechanism rather than general stress.
RNase L activation	Increase viral-RNA destruction and secondary alarm signalling.	Host-RNA loss, translational collapse and tissue injury.	Replication-competent virus, RNA cleavage, collision burden and viability together.
Partial RNase L inhibition	Reduce bystander RNA damage and inflammatory death.	Prolonged viral replication and transmission.	Late-stage benefit after microbial control, ideally with combination antiviral therapy.
GCN2 / ISR tuning	Restore translation or limit persistent stress output.	Removing the initiation brake may increase ribosome influx into unresolved stalls.	Genetic validation; note that GCN2iB can inhibit ZAK directly [80].
RQC enhancement	Shorten collision lifetime and clear incomplete polypeptides.	Could support pathogen translation or alter normal regulatory pausing.	Show queue resolution, not merely pathway switching, in the relevant infection.
Pathogen-specific effector blockade	Neutralize a toxin, viral DUB or collision-hijacking protein.	Escape mutations and narrow pathogen coverage.	Effector dependence in authentic infection plus preserved host quality control.

4.5 Pathogen escape and pathway hijacking

Pathogens illustrate why treatment direction cannot be chosen from pathway diagrams alone. EBV BPLF1 counteracts ZNF598-dependent 40S ubiquitylation and impairs disposal of stalled reporter products. Its activity correlates with a GCN2-dependent ISR and preferential translation of uORF- or IRES-containing transcripts; during productive EBV infection, active BPLF1 increased EBNA1 translation and progeny release [86]. Thus, the proviral outcome is demonstrated for BPLF1 in EBV, whereas extension to HCMV and KSHV deubiquitinases rests mainly on ectopic-expression reporter assays. Viperin provides a contrasting collision-associated restriction route, and *Legionella pneumophila* SidI modifies the ribosome and activates ZAK α -p38-ATF3 signalling with an unresolved net effect on host defence [9, 62]. These examples require pathogen-specific mapping of who creates the lesion, which branch benefits the pathogen and whether intervention should restore quality control, preserve host restriction or prevent maladaptive cell death.

4.6 Experimental readouts and assay technologies

No single marker is sufficient to diagnose a biologically consequential collision. Conventional ribosome profiling (Ribo-seq) maps monosome-protected fragments and reveals pause sites, initiation changes and translation efficiency, but a high local footprint can reflect slow decoding without a stable collision. Disome or trisome profiling sequences longer nuclease-protected fragments and more directly localizes queued ribosomes [27, 87]. Its limitations include nuclease and fractionation sensitivity, loss of spatial information, and the possibility that physiological disomes support regulated folding rather than damage.

An experimental readout panel should combine: (i) a physical collision measure such as nuclease-resistant disomes, disome-seq or structural imaging; (ii) an RQC mark, such as ZNF598-dependent ubiquitylated uS10/eS10, with linkage and site information; (iii) pathway outputs including ZAK α activation, phospho-p38/JNK and GCN2-phospho-eIF2 α -ATF4; and (iv) a functional consequence such as nascent-chain clearance, genetically resolved cell fate or infectious yield. Reporter protein, reporter RNA and global translation should be measured in parallel, and the ribosome state must precede sensor activation. These are research readouts, not clinically validated biomarkers.

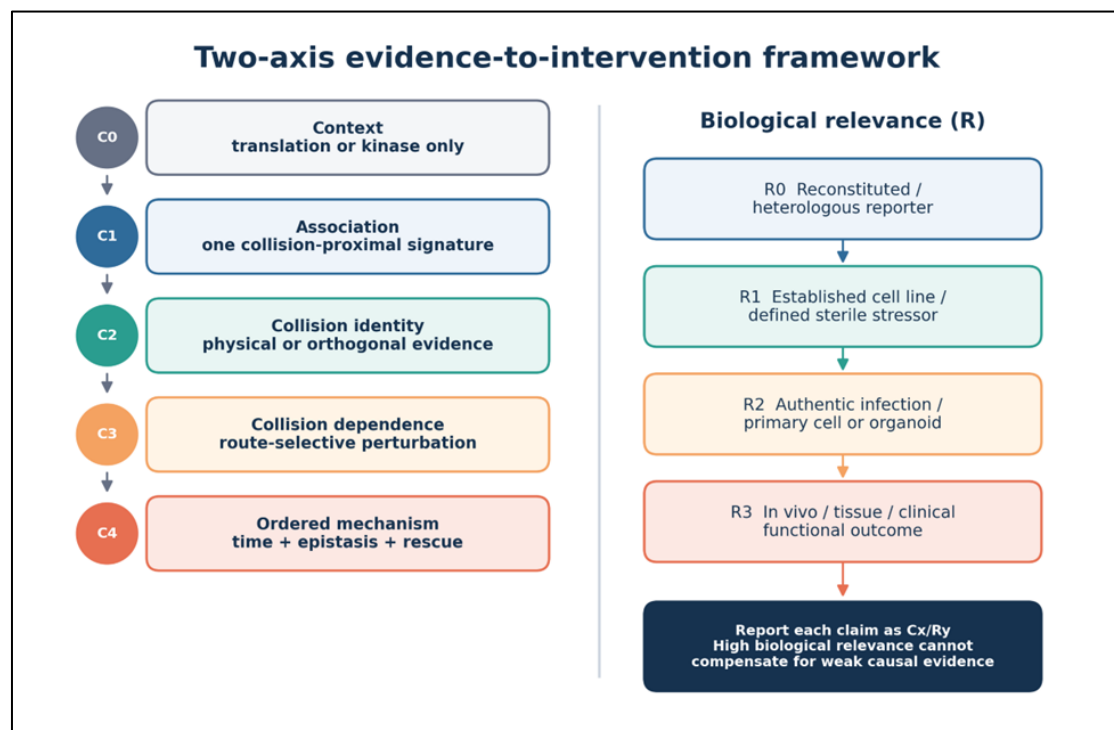


Figure 5: Two-axis evidence-to-intervention framework. Claim-level causal confidence (C0–C4) and biological relevance (R0–R3) must be reported separately. Intervention should target a checkpoint demonstrated by route perturbation and genetic ordering and should preserve pathogen control while improving tissue function.

4.7 Strengths and limitations of the current evidence base

The central mechanistic case is supported by unusually convergent evidence. Structural studies define composite disome interfaces; biochemical reconstitution identifies collision-responsive ubiquitination and splitting reactions; cell genetics orders ZNF598, ASCC, ZAK α and GCN2 relative to downstream outputs; and selected infection or toxin models

connect those events to pathogen control or cell fate. Agreement across these different experimental modes makes the existence of collision-responsive surveillance more secure than any single reporter assay would suggest.

The biological interpretation is less mature. Many decisive experiments use engineered stall reporters, anisomycin, UV, ricin or high toxin doses rather than authentic infection. Disome preparation, nuclease digestion and footprint-length definitions differ between laboratories, and ribosomal ubiquitination or kinase phosphorylation is sometimes used as a collision surrogate without direct physical confirmation. Most infection-linked evidence comes from transformed cell lines or a limited set of epithelial models. Human NLRP1 biology is not reproduced faithfully in rodents, whereas yeast findings define conserved principles but cannot establish mammalian immune function. Publication bias may also favour strong stress phenotypes over adaptive or null responses.

This review has corresponding limitations. Its targeted narrative search was not registered, did not use duplicate independent screening, and did not apply a formal risk-of-bias tool or meta-analysis. The two-axis framework calibrates mechanistic confidence and biological relevance but is not a validated scoring system. The synthesis can therefore identify convergent pathways and decisive experimental gaps, but it cannot estimate pooled effect sizes or comparative treatment benefit. Conclusions are strongest for the existence and ordering of collision-responsive pathways, narrower for their net value during authentic infection, and preliminary for clinical intervention.

4.8 Outstanding questions and research priorities

Several questions now determine whether collision biology will mature into an infection framework rather than remain a collection of stress paradigms. First, authentic-infection maps are scarce. Disome profiling should be paired with matched monosome profiling, RNA cleavage maps and quantitative proteomics across a time course, rather than applied only after toxin or drug treatment. Second, collision identity may be route specific. A cleaved RNA end, a structured frameshift site, damaged rRNA and an eEF2 lesion present different downstream mRNA lengths, nascent chains and ribosomal conformations; whether they preferentially recruit ZAK α , ZNF598, GCN2 or PELO remains unresolved.

Third, cell-type and species boundaries need explicit study. Human NLRP1 provides an epithelial translation-damage sensor, but it cannot be assumed to operate identically in mouse tissues or myeloid cells. Fourth, the field needs quantitative definitions of signal thresholds. Single-cell reporters that simultaneously measure translation, ZAK α activity, ISR output and death commitment could distinguish reversible queues from terminal stress. Fifth, cGAS activation by collided ribosomes needs validation during authentic infection with DNA dependence, cGAMP measurement and cGAS/STING epistasis in the same system [50]. Finally, therapeutic studies must resolve timing: early inhibition of a collision alarm may increase pathogen replication, whereas later inhibition could protect tissue after microbial control has been established.

Table 8: Priority questions for collision-centred infection research

Priority	Decisive experiment	Why it matters
Authentic infection maps	Matched monosome/disome profiling, RNA cleavage maps and proteomics across time.	Distinguishes drug/toxin paradigms from genuine host–pathogen biology.
Route-specific geometry	Compare cleaved RNA ends, frameshift sites, rRNA lesions and eEF2 blockade structurally.	May explain selective recruitment of ZAK α , ZNF598, GCN2 or PELO.
Cell and species boundaries	Primary human barrier and immune cells with matched organoid or in vivo validation.	Prevents overgeneralization of human NLRP1 and mouse phenotypes.
Subcellular location	Spatial translation and collision mapping in viral replication compartments and organelle surfaces.	Tests whether local sensor availability sets pathway choice.
Thresholds and reversibility	Single-cell reporters of translation, collision, ZAK α /GCN2 and fate during washout.	Separates reversible queues from irreversible inflammatory death.
Tissue-linked readouts	Disome or ubiquitin signatures correlated with pathogen and injury endpoints.	Tests whether blood or bulk translation markers report the diseased organ.

5 CONCLUSIONS

Ribosome collisions are physical consequences of persistent translational delay, not pathogen-specific receptors. They can nevertheless connect damage to the translation apparatus with local initiation control, ubiquitin-dependent rescue, nascent-chain disposal, ZAK α - and GCN2-dependent signalling and, in defined human epithelial contexts, NLRP1 activation. Antiviral pathways such as OAS–RNase L and viperin can secondarily generate stall- and collision-prone

states, but their net host benefit must be demonstrated experimentally. Bacterial toxins and viral recoding elements can generate or exploit related states, with direct collision evidence varying among examples.

The strongest general conclusion is methodological as well as biological: translational arrest is not evidence of collision, and collision is not evidence of a beneficial immune response. Causal claims require direct or orthogonal collision measurements, pathway-specific genetics and terminal outcomes at both pathogen and tissue levels. Progress now requires time-resolved infection models that pair collision-resolved measurements and pathway-specific rescue with infectious yield and tissue-function endpoints. Therapeutic modulation should target a demonstrated causal checkpoint and be judged by both microbial control and host injury.

STATEMENTS ON COMPLIANCE WITH ETHICAL STANDARDS

Disclosure of conflict of interest

The author declares no competing interests.

Author contributions

Ban D.H. Al Khayali conceived the review, evaluated the literature, developed the conceptual framework and prepared the manuscript.

REFERENCES

- [1] Juskiewicz S, Chandrasekaran V, Lin Z, Kraatz S, Ramakrishnan V, Hegde RS. ZNF598 Is a Quality Control Sensor of Collided Ribosomes. *Mol Cell*. 2018;72(3):469-481.e7. <https://doi.org/10.1016/j.molcel.2018.08.037>
- [2] Wu CC, Peterson A, Zinshteyn B, Regot S, Green R. Ribosome Collisions Trigger General Stress Responses to Regulate Cell Fate. *Cell*. 2020;182(2):404-416.e14. <https://doi.org/10.1016/j.cell.2020.06.006>
- [3] Narita M, Denk T, Matsuo Y, Sugiyama T, Kikuguchi C, Ito S, et al. A distinct mammalian disome collision interface harbors K63-linked polyubiquitination of uS10 to trigger hRQT-mediated subunit dissociation. *Nat Commun*. 2022;13(1):6411. <https://doi.org/10.1038/s41467-022-34097-9>
- [4] Huso VL, Niu S, Catipovic MA, Saba JA, Denk T, Park E, et al. ZAK activation at the collided ribosome. *Nature*. 2026;649(8098):1051-1060. <https://doi.org/10.1038/s41586-025-09772-8>
- [5] Vind AC, Martínez JF, Wu Z, Bugai A, Mordente K, Abis G, et al. Mechanism of RACK1-dependent ZAK α activation at stalled and collided ribosomes. *Mol Cell*. 2026;86(12):2341-2357.e10. <https://doi.org/10.1016/j.molcel.2026.04.034>
- [6] Xi J, Snieckute G, Martínez JF, Arendrup FSW, Asthana A, Gaughan C, et al. Initiation of a ZAK α -dependent ribotoxic stress response by the innate immunity endoribonuclease RNase L. *Cell Rep*. 2024;43(4):113998. <https://doi.org/10.1016/j.celrep.2024.113998>
- [7] Karasik A, Lorenzi HA, DePass AV, Guydosh NR. Endonucleolytic RNA cleavage drives changes in gene expression during the innate immune response. *Cell Rep*. 2024;43(6):114287. <https://doi.org/10.1016/j.celrep.2024.114287>
- [8] Karasik A, Jones GD, Guydosh NR. Stalled translation on transcripts cleaved by RNase L activates signaling important for innate immunity. *RNA*. 2026;32(6):945-961. <https://doi.org/10.1261/rna.080699.125>
- [9] Hsu JC, Laurent-Rolle M, Pawlak JB, Xia H, Kunte A, Hee JS, et al. Viperin triggers ribosome collision-dependent translation inhibition to restrict viral replication. *Mol Cell*. 2022;82(9):1631-1642.e6. <https://doi.org/10.1016/j.molcel.2022.02.031>
- [10] Robinson KS, Toh GA, Rozario P, Chua R, Bauernfried S, Sun Z, et al. ZAK α -driven ribotoxic stress response activates the human NLRP1 inflammasome. *Science*. 2022;377(6603):328-335. <https://doi.org/10.1126/science.abl6324>
- [11] Robinson KS, Toh GA, Firdaus MJ, Tham KC, Rozario P, Lim CK, et al. Diphtheria toxin activates ribotoxic stress and NLRP1 inflammasome-driven pyroptosis. *J Exp Med*. 2023;220(10):e20230105. <https://doi.org/10.1084/jem.20230105>
- [12] Pinilla M, Mazars R, Vergé R, Gorse L, Paradis M, Suire B, et al. EEF2-inactivating toxins engage the NLRP1 inflammasome and promote epithelial barrier disruption. *J Exp Med*. 2023;220(10):e20230104. <https://doi.org/10.1084/jem.20230104>

- [13] Sundaramoorthy E, Ryan AP, Fulzele A, Leonard M, Daugherty MD, Bennett EJ. Ribosome quality control activity potentiates vaccinia virus protein synthesis during infection. *J Cell Sci.* 2021;134(8):jcs257188. <https://doi.org/10.1242/jcs.257188>
- [14] Xavier A, Kolling M, Bourke AM, Mosler T, Prieto-Garcia C, Karaulanov E, et al. Programmed ribosomal frameshifting triggers translational stress to promote viral replication. *Cell.* 2026;S0092-8674(26)01004-4. <https://doi.org/10.1016/j.cell.2026.08.031>
- [15] Kim KQ, Zaher HS. Canary in a coal mine: collided ribosomes as sensors of cellular conditions. *Trends Biochem Sci.* 2022;47(1):82-97. <https://doi.org/10.1016/j.tibs.2021.09.001>
- [16] Iyer KV, Müller M, Tittel LS, Winz ML. Molecular Highway Patrol for Ribosome Collisions. *Chembiochem.* 2023;24(20):e202300264. <https://doi.org/10.1002/cbic.202300264>
- [17] Rozman B, Fisher T, Stern-Ginossar N. Translation-A tug of war during viral infection. *Mol Cell.* 2023;83(3):481-495. <https://doi.org/10.1016/j.molcel.2022.10.012>
- [18] Karasik A, Guydosh NR. The Unusual Role of Ribonuclease L in Innate Immunity. *Wiley Interdiscip Rev RNA.* 2024;15(6):e1878. <https://doi.org/10.1002/wrna.1878>
- [19] Vind AC, Zhong FL, Bekker-Jensen S. Death by ribosome. *Trends Cell Biol.* 2025;35(7):615-626. <https://doi.org/10.1016/j.tcb.2024.10.013>
- [20] Chang WD, Choe YJ. Quality control and signaling pathways at stalled ribosomes. *Exp Mol Med.* 2026;58(1):82-93. <https://doi.org/10.1038/s12276-025-01623-w>
- [21] Masucci MG. Viruses and RQC: a molecular arms race at the heart of translation. *Trends Microbiol.* 2026;34(6):650-664. <https://doi.org/10.1016/j.tim.2026.03.002>
- [22] D'Orazio KN, Wu CC, Sinha N, Loll-Krippelbein R, Brown GW, Green R. The endonuclease Cue2 cleaves mRNAs at stalled ribosomes during No Go Decay. *Elife.* 2019;8:e49117. <https://doi.org/10.7554/eLife.49117>
- [23] Chandrasekaran V, Juszkievicz S, Choi J, Puglisi JD, Brown A, Shao S, et al. Mechanism of ribosome stalling during translation of a poly(A) tail. *Nat Struct Mol Biol.* 2019;26(12):1132-1140. <https://doi.org/10.1038/s41594-019-0331-x>
- [24] Wu CC, Zinshteyn B, Wehner KA, Green R. High-Resolution Ribosome Profiling Defines Discrete Ribosome Elongation States and Translational Regulation during Cellular Stress. *Mol Cell.* 2019;73(5):959-970.e5. <https://doi.org/10.1016/j.molcel.2018.12.009>
- [25] Han P, Shichino Y, Schneider-Poetsch T, Mito M, Hashimoto S, Udagawa T, et al. Genome-wide Survey of Ribosome Collision. *Cell Rep.* 2020;31(5):107610. <https://doi.org/10.1016/j.celrep.2020.107610>
- [26] Arpat AB, Liechti A, De Matos M, Dreos R, Janich P, Gatfield D. Transcriptome-wide sites of collided ribosomes reveal principles of translational pausing. *Genome Res.* 2020;30(7):985-999. <https://doi.org/10.1101/gr.257741.119>
- [27] Meydan S, Guydosh NR. Disome and Trisome Profiling Reveal Genome-wide Targets of Ribosome Quality Control. *Mol Cell.* 2020;79(4):588-602.e6. <https://doi.org/10.1016/j.molcel.2020.06.010>
- [28] Ikeuchi K, Tesina P, Matsuo Y, Sugiyama T, Cheng J, Saeki Y, et al. Collided ribosomes form a unique structural interface to induce Hel2-driven quality control pathways. *EMBO J.* 2019;38(5):e100276. <https://doi.org/10.15252/embj.2018100276>
- [29] Fedry J, Silva J, Vanevic M, Fronik S, Mechulam Y, Schmitt E, et al. Visualization of translation reorganization upon persistent ribosome collision stress in mammalian cells. *Mol Cell.* 2024;84(6):1078-1089.e4. <https://doi.org/10.1016/j.molcel.2024.01.015>
- [30] Juszkievicz S, Hegde RS. Initiation of Quality Control during Poly(A) Translation Requires Site-Specific Ribosome Ubiquitination. *Mol Cell.* 2017;65(4):743-750.e4. <https://doi.org/10.1016/j.molcel.2016.11.039>
- [31] Sundaramoorthy E, Leonard M, Mak R, Liao J, Fulzele A, Bennett EJ. ZNF598 and RACK1 Regulate Mammalian Ribosome-Associated Quality Control Function by Mediating Regulatory 40S Ribosomal Ubiquitylation. *Mol Cell.* 2017;65(4):751-760.e4. <https://doi.org/10.1016/j.molcel.2016.12.026>
- [32] Garzia A, Jafarnejad SM, Meyer C, Chapat C, Gogakos T, Morozov P, et al. The E3 ubiquitin ligase and RNA-binding protein ZNF598 orchestrates ribosome quality control of premature polyadenylated mRNAs. *Nat Commun.* 2017;8:16056. <https://doi.org/10.1038/ncomms16056>
- [33] Simms CL, Yan LL, Zaher HS. Ribosome Collision Is Critical for Quality Control during No-Go Decay. *Mol Cell.* 2017;68(2):361-373.e5. <https://doi.org/10.1016/j.molcel.2017.08.019>

- [34] Matsuo Y, Ikeuchi K, Saeki Y, Iwasaki S, Schmidt C, Udagawa T, et al. Ubiquitination of stalled ribosome triggers ribosome-associated quality control. *Nat Commun.* 2017;8(1):159. <https://doi.org/10.1038/s41467-017-00188-1>
- [35] De La Cruz AC, Tisdale G, Nakayama E, Huang Z, Sinha NK, Green R, et al. Single-protein/RNA imaging reveals ZNF598 as a limiting factor in resolving collided ribosomes. *EMBO J.* 2025;44(18):5215-5232. <https://doi.org/10.1038/s44318-025-00523-z>
- [36] Miścicka A, Bulakhov AG, Kuroha K, Zinoviev A, Hellen CUT, Pestova TV. Ribosomal collision is not a prerequisite for ZNF598-mediated ribosome ubiquitination and disassembly of ribosomal complexes by ASCC. *Nucleic Acids Res.* 2024;52(8):4627-4643. <https://doi.org/10.1093/nar/gkae087>
- [37] Juszkiwicz S, Speldewinde SH, Wan L, Svejstrup JQ, Hegde RS. The ASC-1 Complex Disassembles Collided Ribosomes. *Mol Cell.* 2020;79(4):603-614.e8. <https://doi.org/10.1016/j.molcel.2020.06.006>
- [38] Pisareva VP, Skabkin MA, Hellen CU, Pestova TV, Pisarev AV. Dissociation by Pelota, Hbs1 and ABCE1 of mammalian vacant 80S ribosomes and stalled elongation complexes. *EMBO J.* 2011;30(9):1804-17. <https://doi.org/10.1038/emboj.2011.93>
- [39] Shao S, Hegde RS. Reconstitution of a minimal ribosome-associated ubiquitination pathway with purified factors. *Mol Cell.* 2014;55(6):880-890. <https://doi.org/10.1016/j.molcel.2014.07.006>
- [40] Shao S, Brown A, Santhanam B, Hegde RS. Structure and assembly pathway of the ribosome quality control complex. *Mol Cell.* 2015;57(3):433-44. <https://doi.org/10.1016/j.molcel.2014.12.015>
- [41] Kuroha K, Zinoviev A, Hellen CUT, Pestova TV. Release of Ubiquitinated and Non-ubiquitinated Nascent Chains from Stalled Mammalian Ribosomal Complexes by ANKZF1 and Ptrh1. *Mol Cell.* 2018;72(2):286-302.e8. <https://doi.org/10.1016/j.molcel.2018.08.022>
- [42] Udagawa T, Seki M, Okuyama T, Adachi S, Natsume T, Noguchi T, et al. Failure to Degrade CAT-Tailed Proteins Disrupts Neuronal Morphogenesis and Cell Survival. *Cell Rep.* 2021;34(1):108599. <https://doi.org/10.1016/j.celrep.2020.108599>
- [43] Thrun A, Garzia A, Kigoshi-Tansho Y, Patil PR, Umbaugh CS, Dallinger T, et al. Convergence of mammalian RQC and C-end rule proteolytic pathways via alanine tailing. *Mol Cell.* 2021;81(10):2112-2122.e7. <https://doi.org/10.1016/j.molcel.2021.03.004>
- [44] Hickey KL, Dickson K, Cogan JZ, Replogle JM, Schoof M, D'Orazio KN, et al. GIGYF2 and 4EHP Inhibit Translation Initiation of Defective Messenger RNAs to Assist Ribosome-Associated Quality Control. *Mol Cell.* 2020;79(6):950-962.e6. <https://doi.org/10.1016/j.molcel.2020.07.007>
- [45] Juszkiwicz S, Slodkowicz G, Lin Z, Freire-Pritchett P, Peak-Chew SY, Hegde RS. Ribosome collisions trigger cis-acting feedback inhibition of translation initiation. *Elife.* 2020;9:e60038. <https://doi.org/10.7554/eLife.60038>
- [46] Xu Z, Choi JH, Dai DL, Luo J, Ladak RJ, Li Q, et al. SARS-CoV-2 impairs interferon production via NSP2-induced repression of mRNA translation. *Proc Natl Acad Sci U S A.* 2022;119(32):e2204539119. <https://doi.org/10.1073/pnas.2204539119>
- [47] Vind AC, Snieckute G, Blasius M, Tiedje C, Krogh N, Bekker-Jensen DB, et al. ZAK α Recognizes Stalled Ribosomes through Partially Redundant Sensor Domains. *Mol Cell.* 2020;78(4):700-713.e7. <https://doi.org/10.1016/j.molcel.2020.03.021>
- [48] Sinha NK, Ordureau A, Best K, Saba JA, Zinshteyn B, Sundaramoorthy E, et al. EDF1 coordinates cellular responses to ribosome collisions. *Elife.* 2020;9:e58828. <https://doi.org/10.7554/eLife.58828>
- [49] Misra J, Carlson KR, Spandau DF, Wek RC. Multiple mechanisms activate GCN2 eIF2 kinase in response to diverse stress conditions. *Nucleic Acids Res.* 2024;52(4):1830-1846. <https://doi.org/10.1093/nar/gkae006>
- [50] Wan L, Juszkiwicz S, Blears D, Bajpe PK, Han Z, Faull P, et al. Translation stress and collided ribosomes are co-activators of cGAS. *Mol Cell.* 2021;81(13):2808-2822.e10. <https://doi.org/10.1016/j.molcel.2021.05.018>
- [51] Silverman RH. Viral encounters with 2',5'-oligoadenylate synthetase and RNase L during the interferon antiviral response. *J Virol.* 2007;81(23):12720-9. <https://doi.org/10.1128/JVI.01471-07>
- [52] Li Y, Banerjee S, Wang Y, Goldstein SA, Dong B, Gaughan C, et al. Activation of RNase L is dependent on OAS3 expression during infection with diverse human viruses. *Proc Natl Acad Sci U S A.* 2016;113(8):2241-6. <https://doi.org/10.1073/pnas.1519657113>
- [53] Malathi K, Dong B, Gale M, Silverman RH. Small self-RNA generated by RNase L amplifies antiviral innate immunity. *Nature.* 2007;448(7155):816-9. <https://doi.org/10.1038/nature06042>

- [54] Burke JM, Moon SL, Matheny T, Parker R. RNase L Reprograms Translation by Widespread mRNA Turnover Escaped by Antiviral mRNAs. *Mol Cell*. 2019;75(6):1203-1217.e5. <https://doi.org/10.1016/j.molcel.2019.07.029>
- [55] Karasik A, Jones GD, DePass AV, Guydosh NR. Activation of the antiviral factor RNase L triggers translation of non-coding mRNA sequences. *Nucleic Acids Res*. 2021;49(11):6007-6026. <https://doi.org/10.1093/nar/gkab036>
- [56] Takenaka Y, Yamada A, Tomioka Y, Akiyama Y, Ivanov P. RNase L produces tRNA-derived RNAs that contribute to translation inhibition. *RNA*. 2025;31(7):961-972. <https://doi.org/10.1261/rna.080419.125>
- [57] Asthana A, Gaughan C, Dong B, Weiss SR, Silverman RH. Specificity and Mechanism of Coronavirus, Rotavirus, and Mammalian Two-Histidine Phosphoesterases That Antagonize Antiviral Innate Immunity. *mBio*. 2021;12(4):e0178121. <https://doi.org/10.1128/mBio.01781-21>
- [58] Goldstein SA, Elde NC. Recurrent viral capture of cellular phosphodiesterases that antagonize OAS-RNase L. *Proc Natl Acad Sci U S A*. 2024;121(5):e2312691121. <https://doi.org/10.1073/pnas.2312691121>
- [59] Gizzi AS, Grove TL, Arnold JJ, Jose J, Jangra RK, Garforth SJ, et al. A naturally occurring antiviral ribonucleotide encoded by the human genome. *Nature*. 2018;558(7711):610-614. <https://doi.org/10.1038/s41586-018-0238-4>
- [60] Majhi S, Roy P, Jo M, Liu J, Hurto R, Freddolino L, et al. Viperin expression leads to downregulation of mitochondrial genes through misincorporation of ddhCTP by mitochondrial RNA polymerase. *J Biol Chem*. 2025;301(4):108359. <https://doi.org/10.1016/j.jbc.2025.108359>
- [61] Jandhyala DM, Ahluwalia A, Obrig T, Thorpe CM. ZAK: a MAP3Kinase that transduces Shiga toxin- and ricin-induced proinflammatory cytokine expression. *Cell Microbiol*. 2008;10(7):1468-77. <https://doi.org/10.1111/j.1462-5822.2008.01139.x>
- [62] Subramanian A, Wang L, Moss T, Voorhies M, Sangwan S, Stevenson E, et al. A Legionella toxin exhibits tRNA mimicry and glycosyl transferase activity to target the translation machinery and trigger a ribotoxic stress response. *Nat Cell Biol*. 2023;25(11):1600-1615. <https://doi.org/10.1038/s41556-023-01248-z>
- [63] Ricci-Azevedo R, Sarkar B, Shivcharan S, Hazra A, Browne M, Carlson EK, et al. Redefining Shiga toxin-induced human cell death as NLRP1- and gasdermin E-mediated pyroptosis. *Proc Natl Acad Sci U S A*. 2026;123(3):e2510274123. <https://doi.org/10.1073/pnas.2510274123>
- [64] Kibe A, Buck S, Gribbling-Burrer AS, Gilmer O, Bohn P, Koch T, et al. The translational landscape of HIV-1 infected cells reveals key gene regulatory principles. *Nat Struct Mol Biol*. 2025;32(5):841-852. <https://doi.org/10.1038/s41594-024-01468-3>
- [65] Park C, Walsh D. RACK1 Regulates Poxvirus Protein Synthesis Independently of Its Role in Ribosome-Based Stress Signaling. *J Virol*. 2022;96(18):e0109322. <https://doi.org/10.1128/jvi.01093-22>
- [66] Zhang F, Ji Q, Chaturvedi J, Morales M, Mao Y, Meng X, et al. Human SAMD9 is a poxvirus-activatable anticodon nuclease inhibiting codon-specific protein synthesis. *Sci Adv*. 2023;9(23):eadh8502. <https://doi.org/10.1126/sciadv.adh8502>
- [67] Wang X, Rimal S, Tantray I, Geng J, Bhurtel S, Khaket TP, et al. Prevention of ribosome collision-induced neuromuscular degeneration by SARS CoV-2-encoded Nsp1. *Proc Natl Acad Sci U S A*. 2022;119(42):e2202322119. <https://doi.org/10.1073/pnas.2202322119>
- [68] Iordanov MS, Pribnow D, Magun JL, Dinh TH, Pearson JA, Chen SL, et al. Ribotoxic stress response: activation of the stress-activated protein kinase JNK1 by inhibitors of the peptidyl transferase reaction and by sequence-specific RNA damage to the alpha-sarcin/ricin loop in the 28S rRNA. *Mol Cell Biol*. 1997;17(6):3373-81. <https://doi.org/10.1128/MCB.17.6.3373>
- [69] Sinha NK, McKenney C, Yeow ZY, Li JJ, Nam KH, Yaron-Barir TM, et al. The ribotoxic stress response drives UV-mediated cell death. *Cell*. 2024;187(14):3652-3670.e40. <https://doi.org/10.1016/j.cell.2024.05.018>
- [70] Berlanga JJ, Ventoso I, Harding HP, Deng J, Ron D, Sonenberg N, et al. Antiviral effect of the mammalian translation initiation factor 2alpha kinase GCN2 against RNA viruses. *EMBO J*. 2006;25(8):1730-40. <https://doi.org/10.1038/sj.emboj.7601073>
- [71] Ishimura R, Nagy G, Dotu I, Chuang JH, Ackerman SL. Activation of GCN2 kinase by ribosome stalling links translation elongation with translation initiation. *Elife*. 2016;5:e14295. <https://doi.org/10.7554/eLife.14295>
- [72] Inglis AJ, Masson GR, Shao S, Perisic O, McLaughlin SH, Hegde RS, et al. Activation of GCN2 by the ribosomal P-stalk. *Proc Natl Acad Sci U S A*. 2019;116(11):4946-4954. <https://doi.org/10.1073/pnas.1813352116>

- [73] Harding HP, Ordonez A, Allen F, Parts L, Inglis AJ, Williams RL, et al. The ribosomal P-stalk couples amino acid starvation to GCN2 activation in mammalian cells. *Elife*. 2019;8:e50149. <https://doi.org/10.7554/eLife.50149>
- [74] Zhou C, Zhang M, Murray J, Paulo J, Gygi S, Shao S, et al. GCN1 couples GCN2 to ribosomal state to initiate amino acid response pathway signaling. *Science*. 2025;390(6768):eads8728. <https://doi.org/10.1126/science.ads8728>
- [75] Yan LL, Zaher HS. Ribosome quality control antagonizes the activation of the integrated stress response on colliding ribosomes. *Mol Cell*. 2021;81(3):614-628.e4. <https://doi.org/10.1016/j.molcel.2020.11.033>
- [76] Jenster LM, Lange KE, Normann S, vom Hemdt A, Wuerth JD, Schiffelers LDJ, et al. P38 kinases mediate NLRP1 inflammasome activation after ribotoxic stress response and virus infection. *J Exp Med*. 2023;220(1):e20220837. <https://doi.org/10.1084/jem.20220837>
- [77] Gorse L, Plessis L, Wearne S, Paradis M, Pinilla M, Chua R, et al. Portimine A toxin causes skin inflammation through ZAK α -dependent NLRP1 inflammasome activation. *EMBO Mol Med*. 2025;17(3):535-562. <https://doi.org/10.1038/s44321-025-00197-4>
- [78] Szymanska I, Bauernfried S, Komar T, Hornung V. Vaccinia virus F1L blocks the ribotoxic stress response to subvert ZAK α -dependent NLRP1 inflammasome activation. *Eur J Immunol*. 2024;54(10):e2451135. <https://doi.org/10.1002/eji.202451135>
- [79] Chakrabarti A, Banerjee S, Franchi L, Loo YM, Gale M, Núñez G, et al. RNase L activates the NLRP3 inflammasome during viral infections. *Cell Host Microbe*. 2015;17(4):466-77. <https://doi.org/10.1016/j.chom.2015.02.010>
- [80] Misra J, Spandau DF, Vilseck JZ, Anthony TG, Staschke KA, Wek RC. An inhibitor of GCN2 and the integrated stress response directly targets ZAK protein kinase to limit cytotoxicity. *J Biol Chem*. 2026;302(6):111482. <https://doi.org/10.1016/j.jbc.2026.111482>
- [81] Vind AC, Wu Z, Firdaus MJ, Snieckute G, Toh GA, Jessen M, et al. The ribotoxic stress response drives acute inflammation, cell death, and epidermal thickening in UV-irradiated skin in vivo. *Mol Cell*. 2024;84(24):4774-4789.e9. <https://doi.org/10.1016/j.molcel.2024.10.044>
- [82] Snieckute G, Ryder L, Vind AC, Wu Z, Arendrup FS, Stoneley M, et al. ROS-induced ribosome impairment underlies ZAK α -mediated metabolic decline in obesity and aging. *Science*. 2023;382(6675):eadf3208. <https://doi.org/10.1126/science.adf3208>
- [83] Lee D, Malathi K, Okano T, Nakajima K, Cobat A, Morio T, et al. The OAS-RNase L pathway: Insights from experiments of nature. *Sci Immunol*. 2026;11(115):eads9407. <https://doi.org/10.1126/sciimmunol.ads9407>
- [84] Sidrauski C, McGeachy AM, Ingolia NT, Walter P. The small molecule ISRIB reverses the effects of eIF2 α phosphorylation on translation and stress granule assembly. *Elife*. 2015;4:e05033. <https://doi.org/10.7554/eLife.05033>
- [85] Requião RD, Lima-Silva LF, Estevão P, Costa WLG, Zani NC, Vasconcelos IC, et al. The integrated stress response kinase GCN2 prevents ZAK α -Dependent inflammatory hyperactivation in macrophages. *J Biol Chem*. 2026;302(10):113452. <https://doi.org/10.1016/j.jbc.2026.113452>
- [86] Liu J, Nagy N, Ayala-Torres C, Aguilar-Alonso F, Morais-Esteves F, Xu S, et al. Remodeling of the ribosomal quality control and integrated stress response by viral ubiquitin deconjugases. *Nat Commun*. 2023;14(1):8315. <https://doi.org/10.1038/s41467-023-43946-0>
- [87] Zhao T, Chen YM, Li Y, Wang J, Chen S, Gao N, et al. Disome-seq reveals widespread ribosome collisions that promote cotranslational protein folding. *Genome Biol*. 2021;22(1):16. <https://doi.org/10.1186/s13059-020-02256-0>