Phylogeny  
Orthologous RNASEL genes are present in human, mouse, rat, bovine, pig, elephant, guinea-pig and chicken, indicating deep conservation across vertebrates (Chakrabarti et al., 2011). The protein’s KEN module (pseudo-kinase plus RNase lobe) is structurally related to Ire1 RNases but lacks catalytic kinase motifs, placing RNase L in a distinct pseudokinase-containing nuclease lineage outside the canonical protein-kinase superfamily. Accordingly, RNase L is not classified within any group of the human kinome (Silverman, 2003).

Reaction Catalyzed  
2′-5′-oligoadenylate-activated endoribonucleolytic cleavage of single-stranded RNA at UpN sites:  
ssRNA-UpN + H₂O → 5′-OH RNA fragment + RNA fragment bearing a 2′,3′-cyclic phosphate (Cooper et al., 2014; Silverman, 2003).

Cofactor Requirements  
• Activation strictly requires 5′-triphosphorylated 2′,5′-linked oligoadenylates (2-5A) of ≥3 AMP units (Bisbal & Silverman, 2007).  
• Catalysis is metal-ion independent (Cooper et al., 2014).  
• Two Walker-A P-loops can bind ATP/GTP, but no intrinsic kinase activity has been detected (Madsen et al., 2008).

Substrate Specificity  
Cleavage occurs preferentially at UU > UA ≫ UG/UC dinucleotides within single-stranded, AU-rich regions; transcriptome-wide mapping confirms a dominant UA/UU consensus independent of extensive secondary structure (Cooper et al., 2014; Silverman, 2003).

Structure  
RNase L comprises an N-terminal ankyrin repeat domain (aa 1–330), a pseudo-kinase lobe (aa 331–535) and a C-terminal RNase domain (aa 536–741) (Madsen et al., 2008). Nine ankyrin repeats form the high-affinity 2-5A-binding pocket, with repeats 2–4 contributing most contacts (Ezelle et al., 2016). The catalytic triad His672–Tyr727–Lys789 constitutes a metal-independent active centre (Cooper et al., 2014). Binding of 2-5A bridges the ankyrin surfaces of two monomers, driving antiparallel dimerisation that unlocks the RNase domains; the ankyrin–pseudo-kinase scaffold functions solely as an allosteric switch (Silverman, 2003; Silverman, 2007).

Regulation  
Femto- to picomolar 2-5A enhances dimer affinity by 10⁵–10⁶-fold, converting latent monomers into active nucleases (Silverman, 2003). Endogenous ABCE1/RLI binds the ankyrin domain and blocks 2-5A-induced dimerisation (Ezelle & Hassel, 2012). Cellular 2-5A is rapidly degraded by 2′-phosphodiesterases and phosphatases, terminating signalling (Ezelle et al., 2016). Proteasome-dependent degradation and stress-induced proteolysis generate 83 kDa and 37 kDa fragments that modulate activity and localisation (Bisbal & Silverman, 2007; Liang et al., 2006). Viral antagonists (vaccinia E3L, influenza A NS1, Theiler’s virus L\*, murine coronavirus ns2) either prevent activation or destroy 2-5A (Liang et al., 2006).

Function  
Basal expression is low but is strongly up-regulated by type I and III interferons in many cell types (Ezelle et al., 2016). Upstream, double-stranded RNA activates OAS enzymes to synthesise 2-5A, which binds and activates RNase L (Silverman, 2003). Downstream, cleavage products stimulate RIG-I/MDA5-MAVS and the NLRP3 inflammasome, boosting IFN-β and IL-1β production (Ezelle et al., 2016). Direct effector functions include degradation of viral ssRNA, rRNA-mediated translation arrest and JNK-dependent mitochondrial apoptosis (Silverman, 2007). Additional roles encompass antibacterial cytokine responses (Ezelle & Hassel, 2012), maintenance of epithelial barrier integrity via interactions with Filamin A and E3-ligase LNX (Ezelle et al., 2016), regulation of mitochondrial mRNA turnover through IF2mt binding (Ezelle & Hassel, 2012) and suppression of androgen-receptor transcriptional programmes in prostate cells (Ezelle & Hassel, 2012).

Inhibitors  
Endogenous ABCE1/RLI competitively blocks 2-5A binding (Ezelle & Hassel, 2012). Viral proteins vaccinia E3L, influenza A NS1, Theiler’s virus L\* and coronavirus ns2 antagonise RNase L activation or degrade 2-5A, while host 2′-phosphodiesterases limit pathway output by degrading 2-5A (Ezelle et al., 2016; Liang et al., 2006).

Other Comments  
RNASEL (chromosome 1q25.3, HPC1) acts as a tumour suppressor; germline variants (e.g., M1I, G59S, I97L, E265X, K392R, R462Q, D541E, 471delAAAG) are linked to hereditary prostate cancer (Carpten et al., 2002; Silverman, 2003). The common R462Q allele lowers catalytic activity ~3-fold and may contribute to up to 13 % of prostate-cancer cases (Casey et al., 2002). Associations have also been reported with pancreatic, head-and-neck, breast and cervical cancers (Gusho et al., 2020; Madsen et al., 2008). Chronic fatigue syndrome features elevated RNase L proteolysis and disease-specific 37 kDa fragments (Bisbal & Silverman, 2007).

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