BIOCELL

Tech Science Press

http://doi.org/10.32604/biocell.2026.079670

REVIEW

Costunolide as a Conceptual Framework for Host-Directed Antiviral Modulation: Mechanistic Insights and Future Perspectives

Leonardo Acuña[1], Mariam Ahumada Sabagh[2], Víctor David Osorio Castillo[1,2], Caverly Gooden[3], María Luisa Veisaga[4], Juan Liuzzi[5,6] and Manuel A. Barbieri[2,3,5,6,7,*]

[1]Ph.D. Biochemistry Program, Florida International University, Miami, FL, USA
[2]Department of Biological Sciences, Florida International University, Miami, FL, USA
[3]Graduate Certificate in Molecular and Biomedical Sciences, Herbert Wertheim College of Medicine, Florida International University, Miami, FL, USA
[4]Women's Studies Department, Florida International University, Miami, FL, USA
[5]Department of Dietetics and Nutrition, Robert Stempel College of Public Health and Social Work, Florida International University, Miami, FL, USA
[6]Biomedical Science Institute, Florida International University, Miami, FL, USA
[7]International Center of Tropical Botany, Florida International University, Miami, FL, USA
*Corresponding Author: Manuel A. Barbieri. Email: barbieri@fiu.edu

Received: 26 January 2026; Accepted: 07 May 2026; Published: 26 August 2026

ABSTRACT: Costunolide, a sesquiterpene lactone from *Saussurea lappa* Clarke, exhibits broad pharmacological properties, including anti-inflammatory and anticancer effects. This review examines its emerging potential as a host-directed antiviral compound. Costunolide modulates conserved host signaling pathways frequently exploited during viral infection, including nuclear factor-kappa B (NF-κB), the NOD-like receptor family pyrin domain-containing 3 (NLRP3) inflammasome, and mitogen-activated protein kinase (MAPK) cascades. Inhibition of NF-κB may suppress viral transcription in human immunodeficiency virus (HIV-1) infection, while NLRP3 blockade may limit inflammasome-driven viral reactivation in Epstein–Barr virus (EBV) infection and attenuate hyperinflammation in severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) disease. Additional modulation of phosphoinositide 3-kinase/AKT (PI3K/AKT), Janus kinase/signal transducer and activator of transcription (JAK/STAT), and Wnt/β-catenin pathways further expands its host-directed antiviral profile. These pleiotropic effects are mechanistically linked to the electrophilic α-methylene-γ-lactone moiety, which undergoes cysteine-directed Michael addition on target proteins. Collectively, these findings position costunolide as a promising host-directed antiviral scaffold, though pharmacokinetic optimization and virus-specific experimental validation remain necessary prerequisites for therapeutic translation.

KEYWORDS: Costunolide; antiviral activity; nuclear factor-kappa B pathway; NOD-like receptor family pyrin domain-containing 3 inflammasome; mitogen-activated protein kinase signaling; viral replication

1 Introduction

Plants have evolved a wide array of secondary metabolites as chemical defenses against microbial pathogens [1,2], many of which have since become valuable sources of therapeutic agents [3,4]. Among these compounds, sesquiterpene lactones, predominantly found in species of the Asteraceae family, have attracted sustained interest for their diverse biological activities, including anti-inflammatory, anticancer, and immunomodulatory effects [5–7]. Costunolide, a germacranolide-type sesquiterpene lactone traditionally

isolated from *Saussurea lappa* Clarke, is one of the most extensively studied members of this class and has been investigated in multiple disease contexts [5,8,9].

Although the pharmacological properties of costunolide have been extensively studied in inflammatory and oncological settings, its relevance in viral infections has only recently begun to emerge [10,11]. Many of the cellular pathways targeted by costunolide, including nuclear factor-kappa B (NF-κB), mitogen-activated protein kinase (MAPK), and phosphoinositide 3-kinase/AKT (PI3K/AKT) signaling, are conserved host dependency factors that viruses frequently co-opt to support replication, persistence, and immune evasion [12,13]. NF-κB is exploited by a broad range of DNA and RNA viruses to drive transcription of viral and pro-survival genes [14,15], while PI3K/AKT signaling is hijacked to sustain metabolic support and block apoptosis in infected cells [16]. Accumulating evidence indicates that costunolide modulates these same host pathways to disrupt viral replication, persistence, and immune evasion [5,8], positioning it as a candidate host-directed antiviral scaffold with an intrinsically higher barrier to resistance than direct-acting antivirals [17].

In this review, we integrate current evidence on the molecular pathways targeted by costunolide during viral infection, with particular emphasis on host signaling networks implicated in viral replication, inflammatory responses, and cell survival. We further discuss the translational challenges associated with its pleiotropic activity and highlight future directions for developing costunolide and related sesquiterpene lactones as antiviral therapeutic leads.

While prior reviews have documented the pharmacological properties of costunolide comprehensively, the present review addresses a distinct and previously uncharted dimension of its biological activity. Kim and Choi (2019) provided a foundational synthesis of costunolide's therapeutic potential across inflammatory, oncological, and metabolic disease contexts [5], and Matos et al. (2021) reviewed sesquiterpene lactones as anti-inflammatory natural compounds [8]. Neither work examined costunolide's mechanistic relevance to viral infection or its potential as a host-directed antiviral scaffold. More recently, Amen et al. (2025) reviewed antiviral activities across the sesquiterpene lactone class but did not provide a focused mechanistic analysis of costunolide's specific protein targets and their roles in virus-host biology [10]. The present review fills this gap by systematically reinterpreting costunolide's documented effects on NF-κB, NLRP3, MAPK, PI3K/AKT, JAK/STAT, and Wnt/β-catenin signaling within the framework of viral pathway dependencies, pathways that viruses exploit for transcriptional activation, immune evasion, and replication support. This recontextualization generates a mechanistically coherent rationale for evaluating costunolide as a host-directed antiviral candidate and identifies specific experimental priorities for the field, representing a contribution distinct from existing reviews of either costunolide pharmacology or sesquiterpene lactone biology.

2 Phytochemical Context and Pharmacology

Plants produce a vast repertoire of secondary metabolites to counter biotic and abiotic stresses, including microbial pathogens [1,2,18]. Many of these phytochemicals have been co-opted by humans for medicinal use and underpin several modern drugs, such as salicylates from willow (*Salix* spp.) and quinine from Cinchona bark [19,20]. Terpenes constitute the largest and most diverse class of plant secondary metabolites and are classified by the number of isoprene units into hemiterpenes, monoterpenes, sesquiterpenes, diterpenes, triterpenes, and higher oligomers [21–23]. Among these, sesquiterpenes (C15) display remarkable structural diversity, adopting acyclic, monocyclic, bicyclic, and tricyclic skeletons [24,25], and generating numerous oxygenated derivatives, including aldehydes, ketones, alcohols, acids, ethers, esters, lactones, and epoxides [26].

Sesquiterpene lactones (SLs) are a particularly bioactive subset of sesquiterpenes with well-documented antiparasitic, antimicrobial, anthelmintic, anti-inflammatory [8], and anticancer activities [3,27]. Structurally, these compounds are classified by their non-lactone ring skeletons into eudesmanolides (bicyclic systems composed of two fused six-membered rings), guaianolides and pseudoguaianolides (5–7 bicyclic systems), germacranolides (10-membered monocyclic systems), and xanthanolides (seven-membered monocyclic systems bearing a non-cyclic side chain) [25,28]. Germacranolides are widely regarded as biosynthetic precursors of other sesquiterpene lactone subclasses, with costunolide representing one of the simplest and most extensively studied germacranolide scaffolds (Fig. 1) [5,7,27].

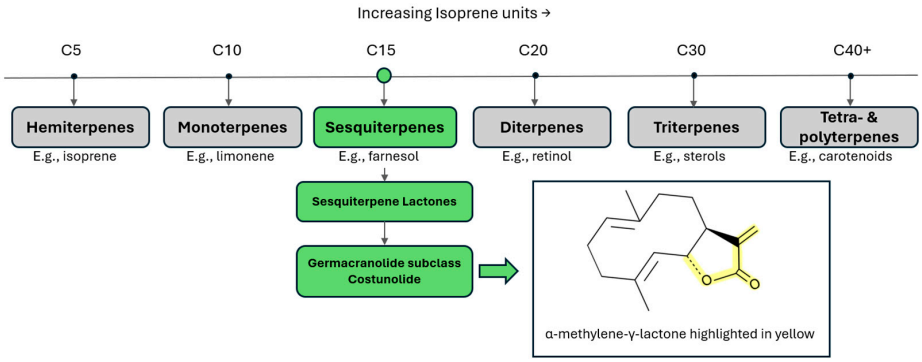

Figure 1: Classification of terpenes by isoprene unit number with representative examples. Sesquiterpenes (C15) are highlighted as the class containing sesquiterpene lactones, with costunolide shown as a representative germacranolide subclass member. The α-methylene-γ-lactone pharmacophore responsible for covalent cysteine alkylation is indicated in yellow. Figure created using Microsoft PowerPoint (Microsoft Corporation, Redmond, WA, USA).

3 Ethnobotanical Origins and Anti-Inflammatory Actions

Traditional herbal medicine is among the oldest forms of therapeutic practice, with ethnobotanical and archaeological evidence documenting the use of plants for medicinal purposes across diverse cultures for thousands of years [4,29]. Many of these traditional remedies rely on bioactive phytochemicals that exert a wide range of pharmacological effects, including anti-inflammatory activity [3,30,31]. In this context, sesquiterpene lactones derived from medicinal plants have attracted sustained interest for their contributions to the therapeutic efficacy of herbal medicines [8,27].

Costunolide derives its name from the plant Costus (*Saussurea lappa* Clarke) and the suffix "olide," which denotes a lactone group. For more than 2500 years, indigenous populations of the Indian Himalayan region have traditionally used costus to treat gastritis, ulcers, inflammatory conditions, liver diseases, diarrhea, joint pain, asthma, and other ailments [9]. Chemical analyses of costus extracts identified a monocarboxylic acid, later named costunolide, as one of the pharmacologically active constituents responsible for these traditional medicinal effects [5,9].

In vitro evaluation of costunolide supports many of these ethnobotanical claims, as a marked reduction in inflammatory markers is observed when cells are pretreated with costunolide [8]. Inflammation is an essential physiological process by which the body responds to noxious stimuli through immune cell activation and the release of inflammatory mediators; however, when sustained, this response becomes pathological and leads to tissue damage [8]. Extensive evidence indicates that persistent inflammation contributes to the development of numerous diseases, including cardiovascular disorders, neurodegenerative conditions, autoimmune diseases, and cancer [32,33].

Zhu et al. demonstrated *in vitro*, using murine peritoneal macrophages (MPMs) and RAW264.7 cells, that costunolide significantly decreases the expression of pro-inflammatory cytokines TNF-α, IL-6, IL-1β, IL-10, and MCP-1 in a dose-dependent manner [34]. The *in vivo* anti-inflammatory effects of costunolide were further evaluated in a murine model of lipopolysaccharide (LPS)-induced sepsis. Pretreatment with costunolide (10 mg/kg) for seven days before LPS challenge resulted in a 60% survival rate, whereas all mice receiving LPS alone succumbed to septic death [34].

In a related mechanistic study, Xu et al. demonstrated that costunolide effectively and selectively inhibits NLRP3 inflammasome assembly by alkylating Cys598 within the NACHT domain, providing a molecular basis for its potent anti-inflammatory activity [35].

4 Molecular Targets and Mechanisms of Action

4.1 Electrophilic α-Methylene-γ-Lactone and Michael Reactivity

The core pharmacophore of costunolide is the α-methylene-γ-lactone moiety, a conjugated α,β-unsaturated carbonyl system that can undergo Michael addition with nucleophilic residues, particularly cysteine thiols in proteins [5,28,36]. In this system, resonance stabilization of the conjugated π bonds, together with polarization by the lactone carbonyl oxygen, renders the β-carbon electrophilic and highly susceptible to nucleophilic attack. According to Pearson's hard–soft acid–base (HSAB) concept,α,β-unsaturated carbonyl compounds are classified as soft electrophiles because of their polarizable π-electron system and preferentially react with soft nucleophiles, such as thiolate groups on cysteine residues, rather than with hard nucleophiles such as hydroxyl groups [28,36]. This chemical reactivity underlies the ability of costunolide to covalently modify cysteine residues in target proteins (Fig. 2) [5,35].

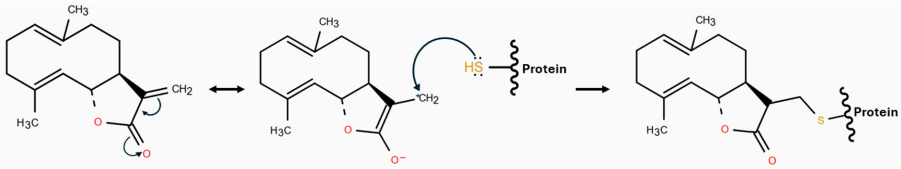

Figure 2: Mechanism of cysteine alkylation by the α-methylene-γ-lactone pharmacophore of costunolide via Michael addition. Resonance delocalization renders the exocyclic methylene electrophilic, enabling nucleophilic attack by a protein cysteine thiol and formation of an irreversible covalent adduct. Figure created using MolDraw (molDraw.com).

Structure-activity relationship (SAR) studies indicate that the α-methylene-γ-lactone moiety is critical for enhancing biological potency and covalent protein binding, and that the stereochemistry and three-dimensional conformation of the sesquiterpene lactone scaffold strongly influence target selectivity and overall biological behavior [37]. Covalent alkylation of cysteine residues has been implicated as a key mechanism underlying many of costunolide's effects on signaling proteins, including NLRP3 [35], IKKβ [38], TAK1 [39], and other kinases and transcription factors involved in inflammatory signaling pathways.

Consistent with this electrophilic reactivity, experimental studies have shown that costunolide covalently modifies specific cysteine residues in key signaling proteins. In particular, costunolide directly alkylates Cys598 in the NACHT domain of NLRP3, resulting in reduced ATPase activity, impaired inflammasome assembly, and suppression of downstream inflammatory signaling [35]. This covalent interaction is mediated by the electrophilic α-methylene-γ-lactone moiety via Michael addition to cysteine thiols, as demonstrated by mutational analysis, mass spectrometry, and irreversible inhibition assays [35]. Similar electrophile-driven mechanisms have been implicated in modulating additional signaling nodes,

including IKKβ and TAK1, providing a molecular framework for the broad anti-inflammatory effects of costunolide observed across diverse experimental models [38,39].

The most direct evidence for costunolide's antiviral activity at the molecular level comes from hepatitis B virus (HBV) studies. Chen et al. demonstrated that costunolide suppresses hepatitis B surface antigen (HBsAg) and hepatitis B e antigen (HBeAg) gene expression in human hepatoma cells (Hep3B and HepA2) in a dose-dependent manner, with an IC50 of 1.0 μM at concentrations that did not significantly affect cell viability. Northern blot analysis confirmed that suppression occurred primarily at the mRNA level, indicating interference with HBV transcriptional programs rather than direct cytotoxicity [40]. Although the precise molecular target mediating this effect was not characterized, the observed potency and selectivity are consistent with cysteine-directed disruption of host transcription factor complexes driving HBV gene expression, a mechanism that aligns with costunolide's documented inhibition of NF-κB and related transcriptional regulators discussed in subsequent sections.

Beyond this experimentally demonstrated activity, the electrophilic α-methylene-γ-lactone moiety raises the mechanistic possibility of direct interactions with viral proteins containing solvent-exposed cysteine residues in their catalytic or regulatory domains. Several clinically validated antiviral targets harbor reactive cysteines amenable to covalent modification, including the catalytic Cys145 of SARS-CoV-2 main protease (Mpro), which is the target of the approved covalent inhibitor nirmatrelvir [41], and cysteine residues present in HIV reverse transcriptase and integrase [42]. The structural basis for covalent inhibition of viral cysteine proteases by natural electrophiles is well established, and the Michael addition chemistry of sesquiterpene lactones is chemically compatible with these targets [41,43]. No experimental studies have directly evaluated costunolide against viral proteases, polymerases, or structural proteins, and this represents a tractable and scientifically well-motivated priority for future investigation. Systematic *in silico* docking of costunolide against the cysteine-containing active sites of HBV polymerase, SARS-CoV-2 Mpro, and HIV reverse transcriptase, followed by biochemical validation, would directly address whether the compound's electrophilic pharmacophore engages viral protein targets in addition to its established host-directed activity.

4.2 Targeting the NLRP3 Inflammasome

The NLRP3 inflammasome is a pattern-recognition receptor that senses a wide range of pathogenic and danger-associated signals and functions as a multiprotein cytosolic complex composed of NOD-like receptor family, pyrin domain-containing 3 (NLRP3) [44,45]. This sensor assembles with caspase-1, an effector protease responsible for the proteolytic maturation of pro-inflammatory cytokines such as IL-1β and IL-18 [45,46]. The apoptosis-associated speck-like protein containing a CARD (ASC) adaptor protein links NLRP3 and caspase-1 [45]. Although inflammasome activation is required for anti-pathogen responses, aberrant activation of the NLRP3 inflammasome has been implicated in the initiation and progression of numerous inflammatory and metabolic disorders [44,45], including type 2 diabetes, atherosclerosis, ischemic heart disease, liver disease, inflammatory bowel disease [46], and neurodegenerative conditions such as Parkinson's and Alzheimer's disease [47].

In EBV-infected cells, viral exploitation of host metabolic stress activates the NLRP3 inflammasome, driving lytic reactivation and viral production [48]. Burton et al. showed that in EBV-positive endemic Burkitt lymphoma cells, oxidative stress or increased glucose availability elevates thioredoxin-interacting protein (TXNIP), promoting NLRP3 inflammasome assembly and resulting in caspase-1–mediated depletion of the heterochromatin-promoting factor KAP1, thereby inducing EBV lytic gene expression and a nearly 4- to 100-fold increase in released viral genomes [48].

In other viral infections, such as SARS-CoV-2 and influenza A virus (IAV), excessive NLRP3 activation is linked to hyperinflammation and cytokine storm, contributing to respiratory failure and multiorgan dysfunction, including central nervous system effects [49,50]. Costunolide directly inhibits NLRP3 activation by covalently modifying the NACHT domain, thereby blocking the ATPase activity required for inflammasome oligomerization [35]. This inhibition suppresses caspase-1 activation and downstream IL-1β maturation in cellular and animal models of NLRP3-driven inflammation [35], consistent with the established role of aberrant NLRP3 signaling in inflammatory disease [47]. In viral contexts, costunolide-mediated NLRP3 inhibition limits inflammasome-dependent viral reactivation, as directly demonstrated in herpesviruses [48,51], and attenuates the excessive inflammatory responses that exacerbate disease pathology in coronavirus infections characterized by hyperinflammation [49,50]. These findings identify NLRP3 as a relevant host target of costunolide and support its role as a modulator of inflammasome-driven inflammatory pathways engaged during viral infection.

4.3 Inhibition of NF-κB Signaling

NF-κB is a central transcription factor complex that regulates inflammatory gene expression [14,15]. Dysregulated NF-κB signaling contributes to chronic inflammation and inflammation-associated tissue damage [38] and plays a central role in the pathogenesis of viral infections [14]. Furthermore, NF-κB is frequently exploited by viruses, including HIV-1, to promote viral transcription, persistence, and immune evasion. In particular, NF-κB activity is essential for efficient transcription from the long terminal repeat (LTR) of HIV-1 [14], where two conserved NF-κB binding sites upstream of the transcription start site drive viral gene expression [15].

Costunolide has been shown to inhibit NF-κB activation across multiple experimental models by targeting key components of the canonical NF-κB pathway. Gene expression analysis and immunochemical studies implicate IKKβ as a critical regulatory node modulated by costunolide in murine models of unilateral ureteral obstruction and inflammatory lung injury [34,38]. In experimental models of pulmonary fibrosis, costunolide likewise suppresses NF-κB–dependent inflammatory responses. This effect is evidenced by reduced phosphorylation of p65, IκBα, and decreased expression of inflammatory mediators, alongside modulation of TGF-β1/Smad2 and Nrf2/NOX4 signaling pathways [52], indicating attenuation of NF-κB signaling rather than direct targeting of upstream IKKβ in this context.

Upstream of IKKβ, transforming growth factor-β–activated kinase 1 (TAK1) has been identified as a molecular target of costunolide. Binding studies, including pull-down assays and biolayer interferometry, demonstrate a direct interaction between costunolide and TAK1, leading to disruption of TAK1/TAB2 complex formation and subsequent inhibition of canonical NF-κB signaling [39]. Given that TAK1 activation is required for NF-κB signaling downstream of multiple receptors, its inhibition by costunolide suggests a unifying mechanism for broad suppression of inflammatory and potentially virus-supportive transcriptional programs [14,39]. In the context of HIV-1 infection, NF-κB binding sites within the 5′ LTR play a central role in HIV-1 transcriptional activation; costunolide-mediated inhibition of this pathway therefore targets a critical node of viral gene expression and replication [14,15].

4.4 Modulation of MAPK Signaling

Mitogen-activated protein kinase (MAPK) pathways regulate fundamental cellular processes, including proliferation, differentiation, stress responses, and apoptosis, and their dysregulation underlies oncogenic transformation as well as a broad spectrum of developmental disorders [53]. Beyond these established roles, MAPK signaling is a central host signaling axis frequently activated during viral infection. A diverse range of

DNA [12] and RNA viruses [13] engage MAPK pathways to support viral gene expression, genome replication, and the modulation of host inflammatory and stress responses that favor productive infection. In particular, the ERK, JNK, and p38 MAPK cascades have been implicated at multiple stages of viral life cycles [12,13], highlighting MAPK signaling as a shared host dependency factor conserved across viral families.

Costunolide has been reported to modulate MAPK signaling primarily by inhibiting stress-activated kinases. Experimental studies in neuronal cell models show that costunolide suppresses JNK and p38 MAPK phosphorylation, whereas its effects on ERK signaling are more variable and depend on cellular context, stimulus, and dose [54,55]. In models of oxidative stress–induced neuronal injury, costunolide-mediated attenuation of MAPK activation correlates with reduced apoptosis and improved cell viability, supporting a role for MAPK modulation in its cytoprotective and anti-inflammatory effects [54,55].

MAPK signaling does not operate in isolation but is tightly interconnected with other inflammatory pathways. Transforming growth factor β–activated kinase 1 (TAK1) is a critical upstream regulator of both MAPK and NF-κB signaling, integrating cellular stress and inflammatory cues. Costunolide has been shown to disrupt the formation of TAK1/TAB2 complexes, thereby inhibiting downstream activation of MAPK and NF-κB signaling cascades [39]. Costunolide-mediated modulation of MAPK activity occurs within a broader network of interconnected host signaling pathways rather than through isolated kinase inhibition.

In viral infection, suppression of MAPK signaling disrupts virus-induced activation of host stress and inflammatory pathways that facilitate viral replication and contribute to tissue damage [12,13]. Costunolide's documented suppression of JNK- and p38-dependent signaling, together with upstream disruption of TAK1-mediated pathway activation, positions it as a host-directed modulatory of signaling networks critically engaged during viral infection [39,54,55].

4.5 PI3K/AKT, JAK/STAT, and Wnt/β-Catenin Pathways

The PI3K/AKT pathway regulates cell growth, survival, metabolic activity, and resistance to apoptosis, and is frequently hijacked by viruses to promote replication and persistence across hepatitis, herpesvirus, and respiratory virus families [16]. Costunolide suppresses AKT phosphorylation and downstream signaling across multiple cellular contexts [56,57], directly impairing the pro-survival and metabolic support that infected cells provide to the replicating virus. These findings identify AKT as a conserved molecular target of costunolide and establish PI3K/AKT interference as a mechanistically relevant component of its host-directed activity during viral infection.

The JAK/STAT pathway presents a critical pharmacological tension that must be explicitly addressed. As a core mediator of interferon-induced transcriptional programs, JAK/STAT signaling is essential for restricting viral replication during early infection [58]; inhibition of this axis by costunolide therefore carries the theoretical risk of blunting antiviral innate immune defenses and exacerbating infection. This concern is not hypothetical, broad JAK inhibition during the early, viremic phase of infection could impair interferon-stimulated gene expression and compromise viral clearance [58]. However, the therapeutic calculus shifts substantially depending on the disease context. Clinical experience with baricitinib, a JAK1/2 inhibitor approved for the treatment of severe COVID-19, demonstrates that during the hyperinflammatory phase of disease, dysregulated JAK/STAT-driven cytokine amplification causes greater harm than the loss of interferon-mediated antiviral activity [59]; JAK/STAT inhibition in this setting reduces immunopathology rather than promoting viral persistence. A distinct but equally instructive context is provided by HIV latency reversal strategies. In shock and kill approaches, reactivation of latently infected $CD4^+$ T cells requires robust LTR-driven viral gene expression; however, JAK/STAT pathway activity in latently infected primary $CD4^+$ T cells contributes to the maintenance of viral latency, such that JAK/STAT inhibition may

facilitate rather than impair latency reversal in this context [60]. The available evidence for costunolide reflects this same disease-stage and context dependence: its documented inhibition of STAT3 and STAT5 in oncogenic and inflammatory models targets constitutively activated, pathological JAK/STAT signaling rather than the transient interferon-driven activation that characterizes early antiviral responses [61–63]. Whether costunolide differentially affects these two modes of JAK/STAT activation remains unresolved and represents a critical question for future experimental evaluation. Until this selectivity is characterized, the antiviral application of costunolide is most relevant to hyperinflammatory viral disease and chronic infection contexts such as HIV latency reversal, where JAK/STAT inhibition is mechanistically supportive rather than detrimental.

Wnt/β-catenin signaling intersects with innate immunity and inflammatory regulation through suppression of virus-induced innate immune responses downstream of the canonical WNT/CTNNB1 axis [64]. Aberrant β-catenin-dependent transcription supports immune evasion and enhanced replication across influenza virus [64] and herpesvirus families [65,66], which actively exploit this pathway to promote viral propagation and evade host antiviral defenses. Costunolide inhibits Wnt/β-catenin signaling by suppressing β-catenin nuclear accumulation and transcriptional activity [67], and given this pathway's established role in viral immune evasion and replication, its modulation extends costunolide's mechanistic reach into host-directed antiviral territory [64,66].

The convergent modulation of PI3K/AKT, JAK/STAT, and Wnt/β-catenin signaling by costunolide disrupts interconnected nodes of viral persistence, immune evasion, and host cell survival across hepatitis, herpesvirus, and respiratory virus infections, positioning this compound at multiple critical intersections of virus-host biology independent of direct virucidal activity.

4.6 Oxidative Stress, ROS, and Autophagy

Oxidative stress and redox signaling are central determinants of cellular fate during both pathological and infectious conditions [68,69]. Viruses actively manipulate host redox homeostasis to create intracellular environments permissive for replication, and ROS accumulation, mitochondrial dysfunction, and ER stress are established mediators of viral pathogenesis [69].

Costunolide exhibits context-dependent redox-modulating activity with direct relevance to these processes. In neuronal models of oxidative injury, costunolide reduces intracellular ROS levels, stabilizes mitochondrial membrane potential, and suppresses stress-activated MAPK signaling through inhibition of p38 and ERK phosphorylation [55]. In cancer cell systems, costunolide acts as a pro-oxidant, inducing ROS accumulation that drives mitochondrial dysfunction, caspase-dependent apoptosis, and ER stress responses with downstream activation of JNK and p38 MAPK pathways [70,71]. Costunolide-induced redox imbalance further drives autophagic flux through AKT/GSK3β suppression [56], engaging autophagy as a cooperative pro-death mechanism. This dual redox capacity is mechanistically relevant to viral infection because viruses similarly depend on mitochondrial integrity, ER homeostasis, and autophagic flux to support replication and evade host cell death programs [68,69,72], cellular dependencies that overlap extensively with those exploited in oncogenesis.

The tissue-dependent and concentration-dependent nature of costunolide's redox activity has direct implications for its antiviral applicability that merit explicit consideration. Different viruses establish infection in cell types with distinct redox environments and transformation states, and the available evidence suggests that costunolide's redox behavior, cytoprotective in non-transformed neuronal cells and pro-oxidant in transformed cancer cells, tracks primarily with cellular transformation status and baseline oxidative tone rather than tissue of origin per se. In the context of HIV-1 infection, where the primary cellular reservoirs

are non-transformed $CD4^+$ T cells and macrophages, the cytoprotective antioxidant profile observed in non-malignant cell models is the more likely predominant effect, suggesting that costunolide would preserve host cell viability in infected reservoirs while modulating the inflammatory and survival signaling that supports viral persistence. In influenza and SARS-CoV-2 infections of respiratory epithelial cells, the relevant question shifts to concentration dependence: at sub-cytotoxic concentrations, costunolide's NLRP3 and MAPK inhibitory effects are predicted to dominate in non-transformed epithelium, while the pro-oxidant profile is expected to emerge only at higher concentrations that would compromise host cell integrity. In EBV-associated Burkitt lymphoma cells, which are transformed, the pro-oxidant profile could constitute an additional therapeutic mechanism operating in parallel with NLRP3-dependent lytic reactivation suppression, cooperating to reduce both viral reservoir size and transformed cell viability. Critically, none of these cell-type-specific predictions have been experimentally validated in the context of active viral infection, and costunolide's redox effects in primary immune cells, including $CD4^+$ T lymphocytes, macrophages, and B cells, remain entirely uncharacterized. Establishing concentration-response relationships in infection-relevant primary cell types represents a prerequisite for a realistic assessment of costunolide's therapeutic window across these distinct viral contexts.

This mechanistic convergence between viral and oncogenic programs extends to telomerase biology. Costunolide suppresses hTERT activity in breast cancer cells through inhibition of c-Myc and Sp1 transcription factors [73], and the structurally related sesquiterpene lactone helenalin directly inhibits telomerase through cysteine-directed alkylation of the reverse transcriptase domain [74]. This is mechanistically relevant because HIV-1 reverse transcriptase, like telomerase, is a template-dependent polymerase that harbors cysteine residues susceptible to electrophilic modification [42], raising the possibility that the α-methylene-γ-lactone pharmacophore may engage reverse transcriptase activity through the same Michael addition chemistry that underlies costunolide's broader target profile.

Given that ROS accumulation, ER stress, and autophagy are established determinants of cellular fate during viral infection, costunolide's documented modulation of these processes places it at a mechanistically relevant intersection of host stress biology and antiviral defense. Virus-specific experimental models are now needed to define the antiviral consequences of these effects.

5 Pharmacokinetics, Metabolism, and BBB Permeability

Pharmacokinetic properties critically shape the translational potential of phytochemicals as therapeutic agents. Natural sesquiterpene lactones, including costunolide, commonly exhibit limited oral bioavailability and extensive metabolic clearance, reflecting their lipophilicity, electrophilic reactivity, and susceptibility to hepatic biotransformation [75]. Experimental pharmacokinetic studies in rodents indicate that costunolide is rapidly absorbed and extensively metabolized, resulting in a relatively short systemic half-life [76]. Metabolic profiling further reveals biotransformation through oxidative and conjugative pathways, yielding multiple metabolites with reduced electrophilic reactivity compared to the parent compound [75,76]. Notably, costunolide and structurally related sesquiterpene lactones cross the blood-brain barrier and modulate barrier integrity by regulating tight and adherens junction proteins and efflux transporter expression, supporting potential relevance for viral infections involving central nervous system pathology [77].

The broad cysteine reactivity conferred by the α-methylene-γ-lactone moiety represents the principal barrier to clinical translation of costunolide. Simultaneous covalent modification of multiple signaling proteins, including NLRP3, IKKβ, TAK1, and AKT, raises significant concerns regarding therapeutic selectivity, as indiscriminate pathway suppression *in vivo* could produce immunosuppression, impaired tissue repair, and off-target cytotoxicity [78]. Although costunolide demonstrates cytotoxic selectivity

toward transformed cells *in vitro*, with relatively lower toxicity in non-malignant cell lines, the therapeutic window in the context of infectious disease remains undefined [5,55]. Preclinical toxicology data are sparse; the rapid metabolic clearance documented in rodent models may inherently limit systemic exposure to reactive species [75,76], but this has not been systematically evaluated against efficacy thresholds in any viral infection model.

Structure-activity relationship studies indicate that modification of the α-methylene-γ-lactone group can attenuate electrophilic reactivity while partially preserving biological activity [37,79], and strategies including prodrug design and advanced formulation approaches offer additional routes to improving bioavailability and tissue targeting. Until selectivity profiling, dose-response toxicology, and antiviral efficacy data are generated in parallel in relevant infection models, the therapeutic window of costunolide remains uncharacterized and its clinical development premature.

6 Limitations of Current Evidence

The mechanistic framework developed in this review reflects the current state of a nascent field, and several evidence gaps warrant acknowledgment as the foundation for future research priorities. The majority of costunolide studies employ *in vitro* models using transformed cell lines, and direct evaluation in primary infection-relevant cell types, including $CD4^+$ T lymphocytes, macrophages, respiratory epithelial cells, and B cells, represents a logical next step for the field [5,8]. Similarly, most *in vivo* evidence derives from inflammatory disease models, and virus-specific infection studies are needed to confirm that the pathway-level effects documented in these contexts translate to infectious disease settings [5,8].

Concentration-response relationships in primary immune cells remain to be established, and pharmacokinetic optimization, as discussed in Section 5, will be central to ensuring that effective concentrations are achievable at sites of infection *in vivo*. The pretreatment paradigm employed in key *in vivo* studies, while valuable for mechanistic proof-of-concept, does not capture therapeutic dosing scenarios [34] and points toward the need for studies evaluating costunolide after viral challenge.

These gaps are not unique to costunolide but reflect the broader stage of development for sesquiterpene lactones as antiviral candidates [3,10]. The mechanistic coherence of the evidence synthesized here, grounded in well-characterized host pathway dependencies conserved across viral families, provides the rationale for the targeted experimental investment needed to advance this compound class toward direct antiviral evaluation.

7 Antiviral Implications and Future Directions

Costunolide exerts pleiotropic biological effects by modulating host inflammatory [14], stress-responsive [12], and survival signaling pathways [16] that viruses depend on for replication, persistence, and immune evasion. Rather than functioning as a direct-acting antiviral, costunolide operates as a host-directed modulator targeting conserved cellular processes that sustain viral infections across multiple virus families. As illustrated in Fig. 3, its mechanistic reach spans NLRP3 inflammasome activation, NF-κB transcriptional programs, MAPK stress cascades, and PI3K/AKT survival signaling, establishing a multi-node host-directed antiviral framework. Validating these mechanistic implications in virus-specific experimental models remains the critical next step toward therapeutic development.

Figure 3: Proposed mechanistic framework for costunolide as a host-directed antiviral scaffold. Covalent alkylation of NLRP3 (Cys598), TAK1, and IKKβ drives downstream suppression of inflammasome activation, NF-κB transcription, MAPK, PI3K/AKT, and Wnt/β-catenin signaling across EBV, SARS-CoV-2, influenza A, and HIV-1 infection contexts. Translational advancement requires pharmacokinetic optimization and virus-specific experimental validation. ↓ inhibition/decrease; ↑ induction/increase; ↑↓ context-dependent. Figure created using Microsoft PowerPoint (Microsoft Corporation, Redmond, WA, USA).

The convergence of costunolide's molecular actions on NF-κB signaling, NLRP3 inflammasome activation, MAPK cascades, PI3K/AKT signaling, redox regulation, and autophagy establishes a multi-node capacity to disrupt viral life cycle progression while simultaneously attenuating excessive inflammatory responses. This pleiotropic mode of action confers a particular advantage in emerging and re-emerging viral infections, where host-directed targeting reduces susceptibility to the rapid resistance mechanisms that undermine direct-acting antivirals [17].

Meaningful comparison of costunolide's antiviral potential is most productively conducted within the sesquiterpene lactone class, where shared pharmacophore chemistry enables direct mechanistic inference and clarifies what makes costunolide the appropriate focal point for this analysis. As the simplest and most extensively mechanistically characterized germacranolide scaffold, costunolide occupies a foundational position within its subclass, it is the biosynthetic precursor from which structural diversity across germacranolides derives [6,7], and its relative simplicity has made it uniquely tractable for the kind of target-level biochemical characterization, including direct identification of alkylated cysteine residues in NLRP3 [35], IKKβ [38], and TAK1 [39], that underpins the mechanistic arguments developed in this review. This depth of mechanistic characterization distinguishes costunolide from structurally related compounds, including parthenolide, which shares the α-methylene-γ-lactone moiety, comparable NF-κB and NLRP3 inhibitory activity, and similar blood-brain barrier permeability [77,80], and has accumulated

more direct antiviral evidence in experimental models, but lacks equivalent target-level mechanistic resolution. Rather than undermining the rationale for focusing on costunolide, this comparison highlights the complementary contributions of the two compounds: parthenolide provides proof-of-concept antiviral activity for the germacranolide scaffold, while costunolide provides the mechanistic framework that explains how that activity is generated and how it might be optimized. Helenalin, a pseudoguaianolide SL cited in this review for cysteine-directed telomerase inhibition [74], further illustrates that subclass-level differences in ring geometry determine which cysteine residues are alkylated and therefore which signaling proteins are targeted, a principle that both validates the cysteine-directed mechanistic framework established here for costunolide and underscores the importance of scaffold-specific structure-activity relationship studies for improving target selectivity. Thapsigargin, a guaianolide SL currently in clinical development as a broad-spectrum antiviral, demonstrates that the sesquiterpene lactone class is capable of producing clinically viable antiviral leads [81,82], establishing a precedent that the mechanistic framework developed here for costunolide is worth pursuing toward experimental validation. Collectively, these comparisons position this review not as an argument for costunolide in isolation but as an exercise in using the best-mechanistically-characterized member of a pharmacologically coherent compound class to establish the conceptual and experimental foundations for broader sesquiterpene lactone antiviral development.

The electrophilic reactivity of the α-methylene-γ-lactone moiety, while mechanistically central to costunolide's pleiotropic activity, simultaneously defines its primary translational liability, as discussed in Section 5. Realizing the therapeutic potential of this multi-node activity profile, therefore, requires addressing several key challenges. Limited pharmacokinetic stability [75,76], context-dependent biological effects, and off-target cysteine reactivity require systematic optimization and rigorous validation [77]. Future studies should prioritize evaluation of costunolide and its derivatives in virus-specific infection models, with emphasis on defining antiviral efficacy, selectivity, and immune modulation within a host-directed therapeutic framework.

In parallel, medicinal chemistry efforts to refine electrophilic reactivity, improve bioavailability, and enhance tissue targeting will be essential to developing costunolide-derived scaffolds with improved therapeutic indices [37,79]. Combining chemical biology, virology, and pharmacology within an integrated experimental framework represents the most direct path toward establishing costunolide as a viable host-directed antiviral lead.

Acknowledgement: We also thank the members of our laboratory for their generous assistance.

Funding Statement: This research was funded by the National Institute of Health, grant number UG HG013615.

Author Contributions: Leonardo Acuña conceptualized the review framework, wrote the original draft, and contributed to review and editing. Mariam Ahumada Sabagh contributed to writing sections of the original draft and participated in review and editing. Víctor David Osorio Castillo contributed to writing sections of the original draft and participated in review and editing. Caverly Gooden contributed to the investigation, review, and editing. María Luisa Veisaga contributed to formal analysis, review, and editing. Juan Liuzzi contributed to methodology, review, and editing. Manuel A. Barbieri supervised the project and contributed to writing, review, and editing. All authors reviewed and approved the final version of the manuscript.

Availability of Data and Materials: Not applicable.

Ethics Approval: Not applicable.

Conflicts of Interest: The authors declare no conflicts of interest.

References

1. Shahrajabian MH, Sun W, Cheng Q. The importance of flavonoids and phytochemicals of medicinal plants with antiviral activities. Mini Rev Org Chem. 2022;19(3):293–318. [CrossRef].
2. Ghildiyal R, Prakash V, Chaudhary VK, Gupta V, Gabrani R. Phytochemicals as antiviral agents: Recent updates. In: Plant-derived Bioactives. Singapore: Springer; 2020. p. 279–95. [CrossRef].
3. Sülsen VP. Sesquiterpene lactones and diterpenes: Promising therapeutic candidates for infectious diseases, neoplasms and other chronic disorders. Molecules. 2021;26(5):1251. [CrossRef].
4. Petrovska B. Historical review of medicinal plants' usage. Phcog Rev. 2012;6(11):1. [CrossRef].
5. Kim DY, Choi BY. Costunolide—A bioactive sesquiterpene lactone with diverse therapeutic potential. Int J Mol Sci. 2019;20(12):2926. [CrossRef].
6. Nguyen TD. Biochemical and evolutionary studies of sesquiterpene lactone metabolism in the sunflower (Asteraceae) family [dissertation]. Calgary, AB, Canada: University of Calgary; 2013.
7. de Kraker JW, Franssen MCR, Joerink M, de Groot A, Bouwmeester HJ. Biosynthesis of costunolide, dihydrocostunolide, and leucodin. Demonstration of cytochrome P450-catalyzed formation of the lactone ring present in sesquiterpene lactones of chicory. Plant Physiol. 2002;129(1):257–68. [CrossRef].
8. Matos MS, Anastácio JD, Nunes dos Santos C. Sesquiterpene lactones: Promising natural compounds to fight inflammation. Pharmaceutics. 2021;13(7):991. [CrossRef].
9. Nadda RK, Ali A, Goyal RC, Khosla PK, Goyal R. *Aucklandia Costus* (Syn. *Saussurea costus*): Ethnopharmacology of an endangered medicinal plant of the Himalayan region. J Ethnopharmacol. 2020;263:113199. [CrossRef].
10. Amen Y, Abdelwahab G, Heraiz AA, Sallam M, Othman A. Exploring sesquiterpene lactones: Structural diversity and antiviral therapeutic insights. RSC Adv. 2025;15(3):1970–88. [CrossRef].
11. Atampugbire G, Adomako EEA, Quaye O. Medicinal plants as effective antiviral agents and their potential benefits. Nat Prod Commun. 2024;19(9):1934578X241282923. [CrossRef].
12. DuShane JK, Maginnis MS. Human DNA virus exploitation of the MAPK-ERK cascade. Int J Mol Sci. 2019;20(14):3427. [CrossRef].
13. Kumar R, Khandelwal N, Thachamvally R, Tripathi BN, Barua S, Kashyap SK, et al. Role of MAPK/MNK1 signaling in virus replication. Virus Res. 2018;253:48–61. [CrossRef].
14. Heusinger E, Kirchhoff F. Primate lentiviruses modulate NF-κB activity by multiple mechanisms to fine-tune viral and cellular gene expression. Front Microbiol. 2017;8:198. [CrossRef].
15. Wang M, Yang W, Chen Y, Wang J, Tan J, Qiao W. Cellular RelB interacts with the transactivator Tat and enhance HIV-1 expression. Retrovirology. 2018;15(1):65. [CrossRef].
16. Diehl N, Schaal H. Make yourself at home: Viral hijacking of the PI3K/Akt signaling pathway. Viruses. 2013;5(12):3192–212. [CrossRef].
17. Aw DZH, Zhang DX, Vignuzzi M. Strategies and efforts in circumventing the emergence of antiviral resistance against conventional antivirals. npj Antimicrob Resist. 2025;3:54. [CrossRef].
18. Sulsen V, Elso OG, Borgo J, Laurella L, Catalan C. Recent patents on sesquiterpene lactones with therapeutic application. Stud Nat Prod Chem. 2021;69:129–94. [CrossRef].
19. Mahdi JG. Medicinal potential of willow: A chemical perspective of aspirin discovery. J Saudi Chem Soc. 2010;14(3):317–22. [CrossRef].
20. Bruce-Chwatt LJ. *Cinchona* and quinine: A remarkable anniversary. Interdiscip Sci Rev. 1990;15(1):87–93. [CrossRef].
21. Cox-Georgian D, Ramadoss N, Dona C, Basu C. Therapeutic and medicinal uses of terpenes. In: Medicinal plants. Cham, Switzerland: Springer International Publishing; 2019. p. 333–59. [CrossRef].
22. Mosquera MEG, Jiménez G, Tabernero V, Vinueza-Vaca J, García-Estrada C, Kosalková K, et al. Terpenes and terpenoids: Building blocks to produce biopolymers. Sustain Chem. 2021;2(3):467–92. [CrossRef].
23. Shu HZ, Peng C, Bu L, Guo L, Liu F, Xiong L. Bisabolane-type sesquiterpenoids: Structural diversity and biological activity. Phytochemistry. 2021;192:112927. [CrossRef].
24. Beer MF, Bivona AE, Sánchez Alberti A, Cerny N, Reta GF, Martín VS, et al. Preparation of sesquiterpene lactone derivatives: Cytotoxic activity and selectivity of action. Molecules. 2019;24(6):1113. [CrossRef].
25. Herz W. Biogenetic aspects of sesquiterpene lactone chemistry. Isr J Chem. 1977;16(1):32–44. [CrossRef].

26. Mabou FD, Yossa IBN. TERPENES: Structural classification and biological activities. IOSR J Pharm Biol Sci. 2021;16(3):25–40.
27. Moujir L, Callies O, Sousa PMC, Sharopov F, Seca AML. Applications of sesquiterpene lactones: A review of some potential success cases. Appl Sci. 2020;10(9):3001. [CrossRef].
28. LoPachin RM, Geohagen BC, Nordstroem LU. Mechanisms of soft and hard electrophile toxicities. Toxicology. 2019;418:62–9. [CrossRef].
29. Pan SY, Litscher G, Gao SH, Zhou SF, Yu ZL, Chen HQ, et al. Historical perspective of traditional indigenous medical practices: The current renaissance and conservation of herbal resources. Evid Based Complementary Altern Med. 2014;2014:525340. [CrossRef].
30. Jalmakhanbetova RI, Suleimen YM, Oyama M, Elkaeed EB, Eissa IH, Suleimen RN, et al. Isolation and *in silico* anti-COVID-19 main protease (mpro) activities of flavonoids and a sesquiterpene lactone from *Artemisia sublessingiana*. J Chem. 2021;2021:5547013. [CrossRef].
31. Twaij BM, Hasan MN. Bioactive secondary metabolites from plant sources: Types, synthesis, and their therapeutic uses. Int J Plant Biol. 2022;13(1):4–14. [CrossRef].
32. Kujawska M, Domanskyi A, Kreiner G. Editorial: Common pathways linking neurodegenerative diseases—The role of inflammation. Front Cell Neurosci. 2021;15:754051. [CrossRef].
33. Zhao H, Wu L, Yan G, Chen Y, Zhou M, Wu Y, et al. Inflammation and tumor progression: Signaling pathways and targeted intervention. Sig Transduct Target Ther. 2021;6:263. [CrossRef].
34. Zhu X, Bai B, Ge X, Zheng B, Xiao Z, Tang Y, et al. Costunolide attenuates LPS-induced inflammation and lung injury through inhibiting IKK/NF-κB signaling. Naunyn Schmiedeberg's Arch Pharmacol. 2024;397(3):1601–10. [CrossRef].
35. Xu H, Chen J, Chen P, Li W, Shao J, Hong S, et al. Costunolide covalently targets NACHT domain of NLRP3 to inhibit inflammasome activation and alleviate NLRP3-driven inflammatory diseases. Acta Pharm Sin B. 2023;13(2):678–93. [CrossRef].
36. Schultz TW, Yarbrough JW. Trends in structure–toxicity relationships for carbonyl-containing α, β-unsaturated compounds. SAR QSAR Environ Res. 2004;15(2):139–46. [CrossRef].
37. Schmidt TJ. Structure-activity relationships of sesquiterpene lactones. Stud Nat Prod Chem. 2006;33:309–92. [CrossRef].
38. Zhao Y, Wang YH, Tu WC, Wang DW, Lu MJ, Shao Y. Costunolide inhibits chronic kidney disease development by attenuating IKKβ/NF-κB pathway. Drug Des Dev Ther. 2024;18:2693–712. [CrossRef].
39. Ye B, Chen X, Chen Y, Lin W, Xu D, Fang Z, et al. Inhibition of TAK1/TAB2 complex formation by costunolide attenuates obesity cardiomyopathy via the NF-κB signaling pathway. Phytomedicine. 2023;108:154523. [CrossRef].
40. Chen HC, Chou CK, Lee SD, Wang JC, Yeh SF. Active compounds from *Saussurea* lappa Clarks that suppress hepatitis B virus surface antigen gene expression in human hepatoma cells. Antivir Res. 1995;27(1–2):99–109. [CrossRef].
41. Owen DR, Allerton CMN, Anderson AS, Aschenbrenner L, Avery M, Berritt S, et al. An oral SARS-CoV-2 M^{pro} inhibitor clinical candidate for the treatment of COVID-19. Science. 2021;374(6575):1586–93. [CrossRef].
42. Hizi A, Shaharabany M, Tal R, Hughes SH. The effects of cysteine mutations on the reverse transcriptases of human immunodeficiency virus types 1 and 2. J Biol Chem. 1992;267(2):1293–7. [CrossRef].
43. Varela K, Arman HD, Berger MS, Sponsel VM, Lin CA, Yoshimoto FK. Inhibition of cysteine proteases via thiol-Michael addition explains the anti-SARS-CoV-2 and bioactive properties of arteannuin B. J Nat Prod. 2023;86(7):1654–66. [CrossRef].
44. Anand PK, Subbarao Malireddi RK, Kanneganti TD. Role of the Nlrp3 inflammasome in microbial infection. Front Microbio. 2011;2:12. [CrossRef].
45. Wang Z, Zhang S, Xiao Y, Zhang W, Wu S, Qin T, et al. NLRP3 inflammasome and inflammatory diseases. Oxid Med Cell Longev. 2020;2020:4063562. [CrossRef].
46. Leemans JC, Cassel SL, Sutterwala FS. Sensing damage by the NLRP3 inflammasome. Immunol Rev. 2011;243(1):152–62. [CrossRef].
47. Fusco R, Siracusa R, Genovese T, Cuzzocrea S, Di Paola R. Focus on the role of NLRP3 inflammasome in diseases. Int J Mol Sci. 2020;21(12):4223. [CrossRef].

48. Burton EM, Goldbach-Mansky R, Bhaduri-McIntosh S. A promiscuous inflammasome Sparks replication of a common tumor virus. Proc Natl Acad Sci U S A. 2020;117(3):1722–30. [CrossRef].
49. Dutta D, Liu J, Xiong H. NLRP3 inflammasome activation and SARS-CoV-2-mediated hyperinflammation, cytokine storm and neurological syndromes. Int J Physiol Pathophysiol Pharmacol. 2022;14(3):138–60.
50. Yalcinkaya M, Liu W, Islam MN, Kotini AG, Gusarova GA, Fidler TP, et al. Modulation of the NLRP3 inflammasome by sars-CoV-2 envelope protein. Sci Rep. 2021;11:24432. [CrossRef].
51. Murata T, Sugimoto A, Inagaki T, Yanagi Y, Watanabe T, Sato Y, et al. Molecular basis of Epstein–Barr virus latency establishment and lytic reactivation. Viruses. 2021;13(12):2344. [CrossRef].
52. Liu B, Rong Y, Sun D, Li W, Chen H, Cao B, et al. Costunolide inhibits pulmonary fibrosis via regulating NF-kB and TGF-β1/Smad2/Nrf2-NOX4 signaling pathways. Biochem Biophys Res Commun. 2019;510(2):329–33. [CrossRef].
53. Hebron KE, Hernandez ER, Yohe ME. The RASopathies: From pathogenetics to therapeutics. Dis Models Mech. 2022;15(2):dmm049107. [CrossRef].
54. Ham A, Lee SJ, Shin J, Kim KH, Mar W. Regulatory effects of costunolide on dopamine metabolism-associated genes inhibit dopamine-induced apoptosis in human dopaminergic SH-SY5Y cells. Neurosci Lett. 2012;507(2):101–5. [CrossRef].
55. Cheong CU, Yeh CS, Hsieh YW, Lee YR, Lin MY, Chen CY, et al. Protective effects of costunolide against hydrogen peroxide-induced injury in PC12 cells. Molecules. 2016;21(7):898. [CrossRef].
56. Xu C, Huang X, Lei X, Jin Z, Wu M, Liu X, et al. Costunolide-induced apoptosis via promoting the reactive oxygen species and inhibiting AKT/GSK3β pathway and activating autophagy in gastric cancer. Front Cell Dev Biol. 2021;9:722734. [CrossRef].
57. Huang H, Park S, Zhang H, Park S, Kwon W, Kim E, et al. Targeting AKT with costunolide suppresses the growth of colorectal cancer cells and induces apoptosis *in vitro* and *in vivo*. J Exp Clin Cancer Res. 2021;40(1):114. [CrossRef].
58. Platanias LC. Mechanisms of type-I- and type-II-interferon-mediated signalling. Nat Rev Immunol. 2005;5(5):375–86. [CrossRef].
59. Marconi VC, Ramanan AV, de Bono S, Kartman CE, Krishnan V, Liao R, et al. Efficacy and safety of baricitinib for the treatment of hospitalised adults with COVID-19 (COV-BARRIER): A randomised, double-blind, parallel-group, placebo-controlled phase 3 trial. Lancet Respir Med. 2021;9(12):1407–18. [CrossRef].
60. de Armas LR, Gavegnano C, Pallikkuth S, Rinaldi S, Pan L, Battivelli E, et al. The effect of JAK1/2 inhibitors on HIV reservoir using primary lymphoid cell model of HIV latency. Front Immunol. 2021;12:720697. [CrossRef].
61. Cai H, He X, Yang C. Costunolide promotes imatinib-induced apoptosis in chronic myeloid leukemia cells via the Bcr/Abl–Stat5 pathway. Phytother Res. 2018;32(9):1764–9. [CrossRef].
62. Jin X, Wang C, Wang L. Costunolide inhibits osteosarcoma growth and metastasis via suppressing STAT3 signal pathway. Biomed Pharmacother. 2020;121:109659. [CrossRef].
63. Lee SH, Cho YC, Lim JS. Costunolide, a sesquiterpene lactone, suppresses skin cancer via induction of apoptosis and blockage of cell proliferation. Int J Mol Sci. 2021;22(4):2075. [CrossRef].
64. Xu Z, Elaish M, Wong CP, Hassan BB, Lopez-Orozco J, Felix-Lopez A, et al. The Wnt/β-catenin pathway is important for replication of SARS-CoV-2 and other pathogenic RNA viruses. npj Viruses. 2024;2:6. [CrossRef].
65. SoRelle ED, Haynes LE, Willard KA, Chang B, Ch'ng J, Christofk H, et al. Epstein-Barr virus reactivation induces divergent abortive, reprogrammed, and host shutoff states by lytic progression. PLoS Pathog. 2024;20(10):e1012341. [CrossRef].
66. Wang C, Hu R, Duan L, Hou Q, Yang M, Wang T, et al. The canonical Wnt/β-catenin signaling pathway facilitates pseudorabies virus proliferation and enhances virus-induced autophagy. Vet Microbiol. 2022;272:109502. [CrossRef].
67. He Y, Moqbel S, Xu L, Ran J, Ma C, Xu K, et al. Costunolide inhibits matrix metalloproteinases expression and osteoarthritis via the NF-κB and Wnt/β-catenin signaling pathways. Mol Med Report. 2019;20(1):312–22. [CrossRef].
68. Kayesh MEH, Kohara M, Tsukiyama-Kohara K. Effects of oxidative stress on viral infections: An overview. npj Viruses. 2025;3:27. [CrossRef].
69. Foo J, Bellot G, Pervaiz S, Alonso S. Mitochondria-mediated oxidative stress during viral infection. Trends Microbiol. 2022;30(7):679–92. [CrossRef].

70. Zhang C, Lu T, Wang GD, Ma C, Zhou YF. Costunolide, an active sesquiterpene lactone, induced apoptosis via ROS-mediated ER stress and JNK pathway in human U2OS cells. Biomed Pharmacother. 2016;80:253–9. [CrossRef].
71. Choi YJ, Choi YK, Ko SG, Cheon C, Kim TY. Investigation of molecular mechanisms involved in sensitivity to the anti-cancer activity of costunolide in breast cancer cells. Int J Mol Sci. 2023;24(4):4009. [CrossRef].
72. Jassey A, Jackson WT. Viruses and autophagy: Bend, but don't break. Nat Rev Microbiol. 2024;22(5):309–21. [CrossRef].
73. Choi SH, Im E, Kang HK, Lee JH, Kwak HS, Bae YT, et al. Inhibitory effects of costunolide on the telomerase activity in human breast carcinoma cells. Cancer Lett. 2005;227(2):153–62. [CrossRef].
74. Zhang Z, Xu L, Cheung HY. The inhibitory effect of helenalin on telomerase activity is attributed to the alkylation of the CYS445 residue: Evidence from QM/MM simulations. J Mol Graph Model. 2014;51:97–103. [CrossRef].
75. Yu Z, Chen Z, Li Q, Yang K, Huang Z, Wang W, et al. What dominates the changeable pharmacokinetics of natural sesquiterpene lactones and diterpene lactones: A review focusing on absorption and metabolism. Drug Metab Rev. 2021;53(1):122–40. [CrossRef].
76. Peng Z, Wang Y, Gu X, Guo X, Yan C. Study on the pharmacokinetics and metabolism of costunolide and dehydrocostus lactone in rats by HPLC-UV and UPLC-Q-TOF/MS. Biomed Chromatogr. 2014;28(10):1325–34. [CrossRef].
77. Ávila-Gálvez MÁ, Marques D, Figueira I, Cankar K, Bosch D, Brito MA, et al. Costunolide and parthenolide: Novel blood-brain barrier permeable sesquiterpene lactones to improve barrier tightness. Biomed Pharmacother. 2023;167:115413. [CrossRef].
78. Paço A, Brás T, Santos JO, Sampaio P, Gomes AC, Duarte MF. Anti-inflammatory and immunoregulatory action of sesquiterpene lactones. Molecules. 2022;27(3):1142. [CrossRef].
79. Amer MFA, Hattab D, Bakhtiar A. Anticancer potential of synthetic costunolide and dehydrocostus lactone derivatives: A systematic review. Eur J Med Chem. 2025;291:117648. [CrossRef].
80. Whipple RA, Vitolo MI, Boggs AE, Charpentier MS, Thompson K, Martin SS. Parthenolide and costunolide reduce microtentacles and tumor cell attachment by selectively targeting detyrosinated tubulin independent from NF-κB inhibition. Breast Cancer Res. 2013;15(5):R83. [CrossRef].
81. Al-Beltagi S, Preda CA, Goulding LV, James J, Pu J, Skinner P, et al. Thapsigargin is a broad-spectrum inhibitor of major human respiratory viruses: Coronavirus, respiratory syncytial virus and influenza a virus. Viruses. 2021;13(2):234. [CrossRef].
82. Shaban MS, Mayr-Buro C, Meier-Soelch J, Albert BV, Schmitz ML, Ziebuhr J, et al. Thapsigargin: Key to new host-directed coronavirus antivirals? Trends Pharmacol Sci. 2022;43(7):557–68. [CrossRef].