Botanical Medicine and Medicinal Mushroom Therapies for Colds and Flu: Optimizing the Immune Response to Prevent and Treat Respiratory Infections

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The authors are solely responsible for the contents, which do not necessarily represent the official views of the NCCIH, or the NIH.
Elements required for an optimal anti-viral immune response against respiratory viruses

Adapted from Ruckwardt et al. Curr Opin Virol, 2016; 16:151–157

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<td>Follicular dendritic cell</td>
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<tr>
<td>DC</td>
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<td>CTL</td>
<td>Cytotoxic T cell</td>
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Elements involved in weak response to respiratory viruses

Adapted from Lambert et al. Front Imm, 2014; 5:1-14
## Botanical & mushroom extracts used in treating respiratory infections

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Echinacea spp.

Part used: Flowers and Roots

Types of extracts used:
- Ethanol-water extracts with alkylamides: anti-inflammatory
- Fresh pressed flower juice high in polysaccharides: pro-inflammatory

Indications: URIs including colds & viral influenza; tonsillitis; strep throat

- Distinct Echinacea extracts show varying results in URI trials:
  - *E. purpurea* given at first URI: no difference in URI severity and duration\(^{52}\) in children but URI recurrence significantly decreased vs. placebo\(^{59}\)
  - *E. pallida* extract reduced the length of URI infection from 13 to 9.8 days for bacterial infection and 13 to 9.1 days for viral infection\(^{10}\)
  - 60% ethanolic *E. angustifolia* extract given TID (1.5mL tincture with 300g equivalent of root) showed no effect vs. placebo in URI occurrence or severity after forced rhinoviral exposure.\(^{54}\)
Common Dosing Regimens for Echinacea

For treatment of common colds:

- **Fresh pressed juice:**
  - Children: used safely BID at 3.75 mL for 2-5 yr olds, 7.5 mL 6-11 yr olds
  - Adults: used safely BID at 5 mL per day
- **Alcoholic extract:** 20 drops in water every 2 hr on first day of symptoms, then TID up to 10 days
- **Whole plant extract:** 3-4 mL taken 8-10 times on first day, then 3-4 times daily for up to 6 days

For prevention of common colds:

- **Alcoholic extract:** 0.9 mL TID up to 4 months; increased to 0.9 ml 5 times daily at first sign of a cold.

For treatment of tonsillitis:

- **Throat spray of Echinacea whole plant extract with sage** every 2 hr up to 10 times daily for 5 days relieves sore throat due to tonsillitis or pharyngitis
- **Echinacea root extract with thuja and wild indigo** used TID up to 2wk in combination with antibiotic
Echinacea-derived constituents have distinct immune modulatory properties

- Immune-modulatory activities of Echinacea-derived constituents:
  - *E. purpurea* alkylamides have IL-2 suppressive effects
  - Alkylamide-induced IL-2 suppression decreased by CytP450 metabolism; may suppress alkylamide affinity for CB2 receptors on immune cells.\(^{46,5}\)
  - Echinacea-derived alkylamides act as agonists of CB2 receptors\(^{58}\) and PPARγ receptors\(^{47}\) to inhibit cytokine production by immune cells.
  - Endophytic bacterial compounds in Echinacea induce immune modulatory effects at lower concentrations than required for immune modulation by other constituents.\(^{44,53}\)

- Different Echinacea treatment conditions influence biological effects:
  - 75% ethanolic *E. purpurea* root extract alone stimulated production of TNF
  - Treatment with same *E. purpurea* extract during LPS stimulation suppressed TNF production.\(^{53}\)
*E. purpurea* alkylamides inhibit IL-2 secretion in activated human T cells

*E. Purpurea* ethanolic extract and alkylamide isolates dose-dependently inhibit IL-2 production in activated human Jurkat T cells\(^46\)

IL-2 inhibitory effects not due to ethanolic extract cytotoxicity
IL-2 suppression induced by Echinacea-derived alkylamide is reduced by PPAR-γ antagonist\textsuperscript{49}
Andrographis paniculata & Eleutherococcus senticosus

Part used: whole herb
Extracts & adult daily dose:
- 500-3,000mg TID
- 300mg tablet QID
4% andrographolides/tablet =48mg
Children’s dose: 1-2g TID or QID

Standardized extract equivalent to 4-6 mg andrographolides

Indications: URIs including colds & flu, bronchitis, tonsillitis, pharyngitis pneumonia, tuberculosis; also used for URI prevention

Common name: Siberian ginseng
Part used: root
Extracts & adult daily dose:
- 9-20g powder in tea
- 2-3g crude extract powder
- 300-400mg concentrate
- Tablet dose ~ 3g powder

4% andrographolides/tablet =48mg

Children’s dose: 1-2g TID or QID

Standardized extract equivalent to 4-6 mg andrographolides

Indications: influenza, swine flu, bronchitis, tuberculosis; also used for URI prevention

Standardized to Eleutherosides B and E
Concentrated extract: 10mg =120mg crude
**A. paniculata and E. senticosus for URIs**

**Evidence of efficacy:**
- *A. paniculata* alone or combined w/ *E. senticosus*: more effective than placebo for treatment of uncomplicated URI\(^7,16,41,43\)
- Combination significantly improved common cold symptoms compared to Echinacea or placebo

**Mechanisms of action:**
- *A. paniculata* alone is not antibacterial, but acts on immune cells
  - decreases neutrophil migration & inflammatory mediators (e.g., NO)
  - Inhibits NFkB binding to DNA promoters of inflammatory genes
- *E. Senticosus* alone may be antibacterial, and is antiviral (vs. RNA viruses)
  - Stimulates macrophages, Complement, Ab production, Tcell proliferation
- *A. paniculata* and *E. Senticosus* combination:
  - Induce peripheral blood lymphocyte & IFN-\(\gamma\) and TNF production
  - Increase activation markers: neopterin, \(\beta\)-2-microglobulin and IL-2R\(^{41}\)
- These actions indicate ↑ in TH1 and ↓ in TH17 responses induced by combination extract
Sambucus nigra

- Part used: Berries
- Extract types: Syrup of elderberry juice, lozenge
- Dosing: within 24-48 hr of symptom onset:
  - Adults: 15 mL QID 3-5 days
  - Children: 15 mL BID for 3 days
- Indications: influenza A & B; H1N1 swine flu; Streptococcus pyogenes (S. pyogenes) infection

- Sambucus extracts and constituents inhibit influenza virus and S. pyogenes
  - Elderberry extract inhibits several strains of influenza virus \textit{in vitro}^{33}
    - Flavonoids from elderberry extract bind to H1N1 virion and block ability of virus to infect host cells \textit{in vitro}^{45}
  - Elderberry extract dose-dependently inhibits H1N1 virus infection
  - Elderberry extract reduces S. pyogenes proliferation upon contact\textsuperscript{33}

- Increases inflammatory cytokines (IL-1β, TNF, IL-6, IL-8) compared to LPS\textsuperscript{1}
- Suppresses virus replication & induces neutralizing Ab In influenza A infected mice\textsuperscript{31}
- 15 mL QID syrup within 48 hr of onset reduced symptoms & duration of influenza A and B infections in double blind, placebo-controlled RCT\textsuperscript{62}
Glycyrrhiza glabra / uralensis

- Part used: Root
- Extract type: hot water extract
- Dosing: typically used in combination formulas, optimally standardized to 4% glycyrrhizin per European Pharmacopoeia
- Indications: bacterial and viral URIs, sore throat, bronchitis, tuberculosis

Has both direct antimicrobial actions and TH1-inducing immunological actions:

- Constituents show antibacterial activity against respiratory bacteria\(^{51}\)
  - licorcidin and glycocoumarin inhibited *S. pyogenes* and *H. influenzae*
- Active against human RSV in human respiratory tract cell lines\(^{14}\)
  - prevented viral attachment & internalization, and induced IFN-β secretion
- Glycyrrhizin induced T cell differentiation toward Thelper 1 (TH1) response
  - Glycyrrhizin-treated DCs increased proliferation of allogenic T cells
  - T cells showed increased IFN-\(\gamma\) and decreased IL-4 production\(^{3}\)
- 18-\(\beta\)-glycyrrhretinic acid induced TH1 response and IFN-\(\gamma\) production *in vivo*\(^{29}\)
Culinary Herbs: *Allium sativa* and *Thymus vulgaris*

- **Common name:** garlic
- **Part used:** bulb
- **Powder or extract standardized to allicin (1-2.5 mg) in 200-400 mg**
- **Indications:** colds and flu, whooping cough, tuberculosis, bronchitis
  - Bactericidal: *H. influenza, S. pyogenes*
  - Allicin (9 mg/kg) immune enhancing:
    - increases IFN-\(\gamma\) and TNF
    - promotes expansion of mature DCs after oral treatment in mice

- **Common name:** thyme
- **Part used:** Aerial parts and volatile oils
- **Ethanolic extract; steam inhalant**
- **Dosing:** insufficient data available
- **Indications:** Bronchitis, cough
  - Thymol bactericidal: *S. pyogenes*, *H.influenza*, *Klebsiella pneumonia*
  - Thymol, carvacrol reduce IL-2, IFN\(\gamma\) secretion in stimulated Jurkat T cells
Propolis

- Part used: Conifer & *Populus spp.* bud resin made by bees
- Extract type: powder, ethanolic extract
- Typical dose: 2 x250mg capsules TID 3 days
- Indications: Common cold, H1N1 influenza, bacterial URIs, tuberculosis

- Hydroethanolic extract bactericidal and antiviral\textsuperscript{4,9}
  - Inhibits *S. pyogenes, H. influenzae*, adenovirus, influenza virus
- Immune-stimulatory effects
  - Caffeic acid ophenethyl ester, cinnamic acids and artepilin-C activate macrophages *in vitro* and *in vivo*\textsuperscript{40,6,9,30}
- Clinical evidence supporting use in treating URIs:
  - Propolis treatment decreases duration of rhinovirus infection (common cold) by 2.5 times vs. placebo\textsuperscript{68}
  - Propolis combined with Echinacea and vitamin C for 12 wk treatment decreased URI incidence, number and duration of infection in children
Local botanical used in respiratory infections: *Lomatium dissectum*

- Part used: Root
- Aqueous extract, added to steam bath
- Dosing: insufficient data available
- Indications: colds, cough, influenza, pneumonia, tuberculosis,

- Okanagan-Colville Native Americans of British Columbia traditionally use Lomatium root in treatment of respiratory infection$^{38}$
- May resolve lower respiratory symptoms in influenza virus infection
  - Lomatium extract treatment decreased CXCL10 secretion by BEAS-2B human bronchial epithelial cells$^{63}$
Lomatium inhibits chemokine secretion

Astragalus membranaceus

- Part used: Root
- Aqueous extract, powder, decoction, tincture
- Typical dosing: 20-500mg extract TID or QID; 1-30g of dried powder daily; 500-1,000mg capsules TID; 3-5mL of a tincture (1:5) in 30% EtOH TID
- Indications: common cold, upper respiratory infections, H1N1 swine flu

- Prevents acute URIs
- Induces T cell-dependent immune response
  - Promotes proliferation of human peripheral blood immune cells
  - Elevates CTL activity
  - Enhances phagocytosis and increases TNF and IL-6 production in vitro
- Astragalus with G. glabra and E. purpurea enhances T cell response
  - Induces CD8 and CD4 T cell activation within 24 hr of ingestion
  - Effect continued for at least 7 days with twice daily dose of tincture
  - T cell-enhancing effects could improve immune response to URI
**Lentinus edodes**: Shiitake

- Part used: Fruiting body and mycelium
- Extract used: Hot water extract; lentinan
- Typical dosing: 6-16g whole, dried mushroom, 4g powder or 1-3g mycelium BID or TID
- Indications: influenza and other viral infections, including common cold, strep throat

- *L edodes* extracts and lentinan have direct antimicrobial actions:
  - Culture fluid of mycelium was active against *S. pyogenes*\(^2\)\(^3\)
  - Lentinan active against adenovirus
- Lentinan induces strong antiviral immune response\(^6\)\(^4\),\(^6\)\(^6\),\(^3\)\(^9\)
  - Enhances IL-12, IFN\(\gamma\) and NO production
  - Increases TH1 response
  - Stimulates maturation of dendritic cells
  - Increases activity of neutrophils and NK cells
Proposed antiviral actions for lentinan

Fig. 7. Mechanisms of antitumor activity of lentinan as a β-1,3-glucan. Taken from Moradali et al., 2007.
Other edible gilled mushrooms: *Agaricus blazeii* & *Pleurotus ostreatus*

- **Common names:** almond mushroom; himematsutaki
- **Part used:** Fruiting body extract
- **Typical dosing:** 500 mg TID
- **Indications:** immune stimulant and antioxidant
  - Extract protective against lethal *S. pneumonia* infection in mice
  - Stimulates TNF and chemokine CXCL8 (IL-8) production

- **Common name:** oyster mushroom
- **Part used:** Fruiting body
- **Dosing:** insufficient data available
- **Indications:** immune stimulant; directly bactericidal
  - Extract inhibits *K. pneumonia* and *S. pyogenes in vitro* \(^{61}\)
  - 8 week hot water extract increased IFN\(\gamma\), IL-12, and NK cell activity \(^{50}\)
Polypore mushrooms: *Ganoderma lucidum* (Reishi)

- Part used: Carpophores
- Raw powder, decoction, encapsulated powder, ethanol and aqueous extracts
- Dosing: 2-6g or equivalent dosage of concentrated extract
- Indications: influenza, bronchial diseases

Ganoderma constituents have antimicrobial and immune-stimulatory effects

- Triterpenoids ganoderic acid TQ and TR inhibited activity of different influenza neuraminidase subtypes
- Effects ranged from 55.4% to 96.5% inhibition for different NA subtypes
- *G. lucidum* isolates showed inhibitory effects against Influenza A
- Treatment of dendritic cells with *G. lucidum*-derived polysaccharide:
  - Enhanced cell-surface expression of CD80, CD86, CD40, CD54
  - Increased T cell stimulatory capacity and secretion of IFN\(\gamma\) and IL-10
- Ganoderic acid enhances NK and IL-2 activity in vivo
Cordyceps sinensis and other Cordyceps spp.

- Entomopathogenic fungi
- Part Used: Mycelium
- Dried aqueous extract of mycelium
- Traditional dosing: 3-9g daily in tea or meal; 1g TID of CS-4 strain
- Indications: coughs, chronic bronchitis, respiratory disorders

- Cordyceps extracts induce strong antimicrobial immune responses
  - Aqueous extract of mycelium increases phagocytic activity of human monocytic U937 cells
  - Extract abrogates inhibitory effect of Group A Streptococcal (GAS) virulence factor SPE B on phagocytosis
  - Extract also Increases expression of cytokines IFN-γ, IL-12 and TNF, involved in augmenting phagocytosis
  - *C. militaris* extract enhances NK cell activity, lymphocyte proliferation and partially increases TH1 cytokine secretion *in vivo*.28
**Crytoporus volvatus & Phellinus igniarius**

- Common names: Pouch Fungus, Cryptic Globe, Veiled Polypore
- Part Used: Fruiting body
- Aqueous extract
- Dosing: insufficient data available
- Indications: influenza and other URIs; immune-stimulating

- Inhibited Influenza A *in vivo & in vitro*\(^17\)
- Immune-modulatory polysaccharides:
  - Reduce LPS-induced expression of TLR2 mRNA\(^60\)
  - May help prevent LPS-induced lung injury in respiratory infections

- Common name: Willow Bracket
- Part Used: Fruiting Body
- Aqueous extract
- Dosing: insufficient data available
- Indications: influenza and other URIs; immune-stimulating

- Interferes with influenza virus replication cycle:
  - Inhibits viral attachment to cells\(^35\)
- Enhances antiviral responses
  - Increases CD8 T cells and NK cell activity *in vivo*\(^65\)
**Trametes versicolor** (Turkey Tail)

- **Fungi Class:** Basidiomycetes
- **Common Name:** Turkey Tail
- **Fruiting body and mycelium extracts**
  - **PSK:** pharmaceutical grade hot water mycelium extract
- **Typical dosing:** 1.5g BID
- **Indications:** URIs, pulmonary disorders, cancer treatment adjuvant

**Adjunctive treatment for several cancer types:**
- Stomach cancer: 16 RCTs in 6462 patients
- Colorectal cancer: 8 RCTs in 1374 patients
- Esophageal cancer: 4 RCTs in 279 patients
- Breast cancer: 3 RCTs in 1517 patients

- **PSK induces TH1-dependent antitumor and antiviral immune responses**
  - TLR2 agonist actions prime strong dendritic cell activity
  - Induces TH1 cytokines, CTL and NK cell responses
- **Active constituents proposed to be beta-1,3-D-glucans,** shown to be bioavailable after oral ingestion

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57, 69
PSK induces TH1 response needed for effective antitumor and antiviral immune responses

PSK dose-dependently increases Th1 cytokines after oral gavage in Her2/neu tumor bearing mice

PSK increases DCs in tumor draining lymph nodes (TDLN)

PSK increases cytotoxic effector molecules in tumor target cells
PSK acts as a TLR2 agonist to activate DCs

A

TLR2

Concentration of TLR Agonists (AU)

SEAP Activity (OD560)

HKLM
PSK

B

TLR4

Concentration of TLR Agonists (AU)

SEAP Activity (OD560)

LPS
PSK

C

T cells

B cells

NK cells

DC

D

E

WT
TLR2−/−
TLR4−/−

F

% of IFN-γ+ NK cells

Treatment Groups

PBS
PSK

IL-12p40 (pg/ml)

IL-12p40 (pg/ml)

PBS
LPS
PSK

PBS
LPS
PSK

PBS
LPS
PSK

PBS
LPS
PSK

PBS
LPS
PSK
PSK dose-dependently enhances NK cell activation

PSK dose-dependently induces CD69 activation marker in human NK cells
Proposed mechanism for mushroom-derived beta-glucans enhancing antiviral immune responses

Beta-glucans

Activates accessory cells

BG Receptors

TLR2
CR3
Dectin-1

Activates NK cells

NK cells

Virally infected cell

Dead target cell

CD4+ Th1

IL-2

CD8+ CTL

Augments antiviral CMI
Limitations & future research

Most studies conducted in human cell lines *in vitro*

- More clinical trials needed to determine optimal parameters (e.g., dosing regimen) for treating and preventing respiratory infections

Need to ensure modulatory effects observed *in vitro* are not due to bacterial endotoxin contamination

- Several studies report cytokine-suppressive actions, not caused by bacterial endotoxins which induce inflammatory cytokines
- Bacterial endotoxin testing is requirement for *in vitro* assay

Quality control of botanical and mushroom extracts needed

- Ensuring product quality and stability are key to accurately assessing botanical and medicinal mushroom extracts for safety and efficacy

Some immune-enhancing effects may be due to endophytic bacterial compounds in botanical and mushroom extracts

- Further research needed to identify actions of constituents in bioactive extracts and correlate levels with growth and extraction conditions.
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