Antimicrobials – Complexities and effective clinical approaches

The human body is populated by trillions of microorganisms, requiring a complex interaction between host and microbe. Whilst net neutral or positive impacts on health are part of normal and in some circumstances essential symbiotic interactions, commensal relationships that create biological disadvantage to the host health may contribute to disease progression. Within this context, the microorganism is considered pathogenic.[1] At this stage, intervention may be required in the form of antimicrobial treatment, whilst we look for a 'cause' of imbalance and build immunity.

Antimicrobial resistance and natural antimicrobials

Antimicrobial resistance may develop through a variety of mechanisms, including direct inactivation of antibiotics, target site modification, bypass or alteration of metabolic pathways, prevention of drug uptake and export out of the cell by ATP-binding cassette (ABC) transporters, thereby reducing intracellular concentrations of antimicrobials.[2] As such, there is a growing need for prudent and calculated use of medical interventions, and the expansion of the use of alternative antimicrobials.[3]

Herbal antimicrobials represent an important area for both research and intervention. This resurgence of interest and the driving force of necessity have led to a growing body of data on the antimicrobial effects of natural herbal compounds.

Phellodendron amurense: Traditional uses and contemporary prescribing

Phellodendron is described in the oldest book in Chinese medicine, the Shennong Ben Cao Jing, which dates back to 2,800 BCE, as a member of the superior class of medicines, and recommends its use for diarrhoea, dysentery and vaginal discharge.[4]

Currently, it is regularly prescribed in the treatment of diarrhoea, gastroenteritis, genitourinary tract infections, abdominal pain, dysentery, and intestinal worms.[5-7]

In vitro trials have shown Phellodendron extract to significantly inhibit viral replication, and to induce innate immune responses including type 1 interferon and pro-inflammatory cytokines.[8]

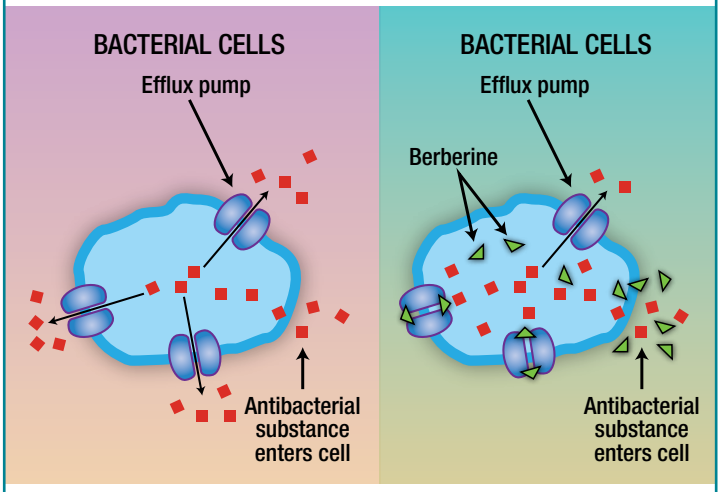
The main active constituent of Phellodendron is berberine, which has been shown to have extensive antimicrobial activity.

Berberine: Multiple mechanisms of action

Berberine is an isoquinoline quaternary alkaloid, and it exhibits a diverse number of pharmacological properties. Studies dating back to the 1960's have shown that berberine has the ability to inhibit certain bacteria and parasites directly.[9] Over the years, a number of mechanisms have been proposed.

Bacterial resistance to treatment may develop when bacteria increase the number or activity of active efflux pumps to remove drugs and other complexes that act as antibiotic compounds. Berberine has been shown to inhibit the action of the multidrug resistance pump (MDR) NorA of certain Gram-positive bacteria, including *Mycobacterium tuberculosis* and methicillin-resistant *Staphylococcus aureus* (MRSA).[10] Berberine may also accumulate within bacterial cells, particularly when the MDR Nor A is not upregulated.[10]

Figure 1: The role of berberine in reducing bacterial resistance to treatment by inhibiting the activity of efflux pumps.



Multidrug resistance: Bacterial cells increase efflux pumps to remove antibacterials from cell.

Berberine reduces efflux pump activity so the antibacterial substance such as berberine stays inside the cell.

Within the cell, berberine may act as a DNA ligand and induce DNA mediated cell death by binding to DNA strands within the bacteria.[10] Berberine may also inhibit bacterial cell division via inhibition of FtsZ; a protein involved in the development of the Z-ring at the midcell site of the future septum during bacterial cell division. Cell elongation has been noted in dividing microbes exposed to berberine; this is a sign of reproductive disruption.[11]

Inhibition of fungal growth by berberine has also been noted. Species including *Aspergillus*, *Penicillium*, *Candida* and *Cryptococcus* have shown sensitivity to berberine extracts, including fluconazole-resistant strains.[10,12] One mechanism that has been proposed is the disruption of cellular redox homeostasis within the mitochondria, affecting manganese superoxide dismutase, thereby causing increases in oxidative stress and inhibition of cellular reproduction.[8] Another potential mechanism of action for berberine is the inhibition of alpha amylase production, an enzyme produced by fungal microorganisms in order to access nutrients from hosts.[13] Additionally, berberine has

This technical sheet (V.02/17) is intended for the further education of practitioners. Any statements made do not intend to claim a cure for any condition mentioned.

been shown to disrupt the biofilms of fluconazole-resistant strains of *Candida* and *Cryptococcus*.[12]

Berberine may also be useful in the prevention of microbial colonisation. Lipase production is a technique used by a diverse range of organisms, including those involved in acne, cholera and gastroenteritis, in order to better colonise skin and mucosal surfaces. Berberine has a mild anti-lipase action which may be an additional mechanism of Phellodendron's known antimicrobial effects.[14,15]

Berberine's Antimicrobial Mechanisms of Action:

- Reduces bacterial efflux pumps to counter antibacterial resistance
- Induces DNA mediated cell death
- Inhibits bacterial cell division
- Oxidises fungal cells so they apoptose
- Disrupts fungal biofilms
- Possible anti-lipase action

Garlic: Active constituents and antimicrobial actions

Allium sativum (Garlic) extracts have been shown to have antibacterial, antifungal, anti-protozoan and antiviral properties.[16] Even studies using simple Garlic powder extracts have been shown to produce an inhibitory effect on pathogenic gastrointestinal colonies.[17] In spite of this, the need for consistent, safe and efficacious Garlic extracts with enhanced stability of the primary active constituents, has led to advancements in production processes and the resultant therapeutic compounds.[18]

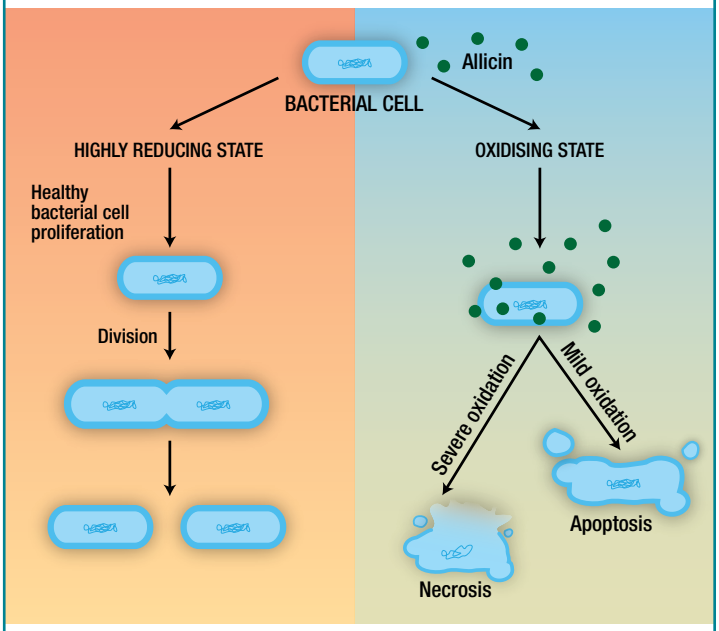
Allicin is the most widely studied component of Garlic, and is also known as diallyl thiosulphinate. Within the plant, alliin and alliinase are stored in separate sub-cellular compartments. When the cell wall is damaged, the enzymes, alliinase of the bundle sheath cells acts upon allin to form allicin.[17,19] Pure allicin is generally a highly volatile compound.[16] However, certain supplemental forms, such as those made with Allisure technology, may provide stabilised allicin that is bioavailable.[20]

Perhaps the most discussed antimicrobial mechanism of allicin is its inhibition of the thiol-containing enzymes, contributing to the death of the microorganism. Thiol-containing enzymes are responsible for maintaining a cellular redox state within microbes.[16,19] In addition, interaction with thiol-containing compounds may reduce the virulence of pathogenic microorganisms even when death is not likely.[16] Allicin also inhibits other microbial enzyme systems necessary for the survival of microbes, such as the acetyl-CoA-forming system. Inhibition of RNA synthesis is considered part of the antimicrobial effect of allicin.[16,19]

Garlic extracts have shown antiviral activity against a number of viral strains, including human cytomegalovirus, influenza B and herpes simplex virus type 1 and 2. One study utilising a double-blind placebo controlled design, showed that the allicin-containing garlic supplement Allisure taken over a 12 week period, resulted in a statistically significant reduction in both the number of colds and the average duration of symptoms in the active group based upon self-report.[20] In addition to the antiviral effects of allicin, the allicin-derived compound ajoene has also demonstrated a potent antiviral propensity.[16]

Allicin has been shown to disrupt the electrochemical potential of yeast cells and cause apoptosis.[18,21] This alteration of the electrochemical potential of the cell is suggested to represent a shift in the cellular redox potential that induces cellular apoptosis through redox-regulated signal pathways.[21]

Figure 2: The action of allicin in bacterial cell apoptosis and necrosis.



The biological status of microbial cells is determined by their redox status. Proliferation can only occur when the cell is in a highly reducing state; apoptosis occurs when the cell shifts towards a more oxidising state. If cellular oxidation is too great for the microbial cells to apotose they necrose instead. Allicin inhibits the microbial enzyme systems which microbes rely on to maintain cellular redox state, leading to apoptosis, necrosis or reduced virulence.

Biofilm disruption and allicin

Allicin may reduce bacteria's tolerance to antimicrobial agents through its actions on biofilm. Allicin has been shown to reduce the surface adhesion potential of *Pseudomonas aeruginosa*, by inhibiting its production of exopolysaccharides. Allicin causes *P. aeruginosa* biofilms to lose complexity and density, thereby reducing the biofilms' ability to develop antimicrobial resistance.[22] Allicin has also been shown to reduce biofilm formation in uropathogenic *Escherichia coli*, even at subinhibitory concentrations.[23] Additionally, allicin has been shown to be superior to fluconazole in the inhibition of *Candida albicans* biofilm production.[24] It has also been shown that allicin inhibits biofilm formation by *Staphylococcus* epidermis by preventing the production of polysaccharide intercellular adhesin, a key adherence factor in the formation of biofilms.[25]

Thyme: Broad spectrum activity and biofilms

Thymus vulgaris (Thyme) has a long history of use in Western herbal medicine for respiratory, gastrointestinal and urinary tract infections.[26-28] In addition to its broad antibacterial, anthelmintic, antifungal and antiviral actions; it is carminative, antispasmodic, expectorant, diaphoretic and diuretic.[26-28] The combination of these qualities makes it well suited for a range of conditions, notably gastrointestinal and respiratory infections and their symptoms, such as gastritis, dyspepsia, diarrhoea, gas, nausea, bloating, coughs and upper respiratory catarrh.[26,27,29] It has also been used as a urinary antiseptic.[27,30]

Thyme's active constituents include essential oil (primarily the phenols thymol, eugenol and carvacrol), flavonoids, rosmarinic acid and

saponins.[31,32] The antibacterial and antifungal activity of thymol has been well documented.[32] The synergistic effects of combinations of Thyme's phenols has also been demonstrated.[32]

Thyme extract has been shown *in vitro* to disrupt the biofilms and inhibit the metabolic activity of *Staphylococcus aureus*, *Bacillus cereus*, *Streptococcus pneumoniae*, *Klebsiella pneumoniae*, *P. aeruginosa* and pathogenic *E. coli*.[33] *In vitro* studies have shown that Thyme extract is able to inhibit *Helicobacter pylori* growth and urease activity.[34]

Myrrh: Ancient use and modern research

Commiphora myrrha (Myrrh) has historically been used in European, Chinese, Tibetan, Middle Eastern and Ayurvedic medicine systems for the treatment of infections and respiratory conditions.[31] King's American Dispensatory recommends it as a mucous membrane stimulant and antiseptic for the treatment of chronic bronchitis, leucorrhoea, and chronic gastritis.[35]

A number of clinical trials have been performed in which cure rates of over 90% were achieved through the use of 600mg of Myrrh extract per day for 6 days in the treatment of parasitic helminthic diseases including schistosomiasis and fascioliasis.[36]

In a 2001 study on 204 patients with Schistosomiasis, patients were given Myrrh at a dose of 10mg/kg/day for three days, resulting in a cure rate of 91.7%. Non-responsive patients were given the same dose again, this time for six days, resulting in a cure rate among this group of 76.5%, or an overall cure rate of 98.09%.[37]

In vitro trials have found Myrrh to be effective against a number of common pathogenic strains of fungal and bacterial species, including *C. albicans*, *E. coli*, *Staphylococcus aureus* and *P. aeruginosa*.[31,38]

Oregano oil: From *Blasto* to fungal and bacterial cell disruption

Oreganum vulgare (Oregano) essential oil is a well-known antimicrobial. It has traditionally been used as a respiratory antiseptic, for digestive disorders and for urinary tract infections.[28,39,40] It has a high content of carvacrol, which has been extensively studied for its antimicrobial activity.

Oregano oil has been found to be effective against a wide range of gram-positive and gram-negative organisms *in vitro*.[40]

Oregano oil and its isolated component carvacrol are understood to affect bacterial cell membranes possibly by affecting the fatty acid composition, causing them to become more permeable. Oregano's phenolic constituents may also enter bacterial cells and disrupt their metabolism.[40]

A clinical trial in 2000 showed that 6 weeks treatment with 600mg of Oregano oil per day was effective against *Blastocystis hominis*, *Entamoeba hartmanni* and *Endolimax nana*. 7 of 11 patients with *B. hominis* showed improved gastrointestinal symptoms after treatment.[41]

Increased survival rates were seen in *S. aureus*-infected mice which were treated with Oregano oil.[40]

Oregano oil has been found to have antifungal activity against a wide range of fungi, including fluconazole resistant *C. albicans*.[40,42] Both fluconazole resistant strains and regular strains of *Candida* are equally susceptible to Oregano oil, suggesting that Oregano oil's antifungal mode of action relates to a route completely separate from those affected by azole resistance.[42] Both carvacrol and eugenol appear to disrupt *Candida* cell surfaces.[40]

In a 2001 study, mice infected with systemic *C. albicans* were given 8.6mg/kg body weight of oregano oil per day for 30 days resulting in a survival rate of 80% compared to the control group mice who were given olive oil as a placebo, and all died within 10 days.[43]

Oregano oil has traditionally been used as a carminative, stomachic and antispasmodic, and so is an ideal component in the treatment of gastrointestinal disturbances. It is also expectorant, and has a long history of use for the treatment of bronchitis, tickling coughs and asthma.[28]

Oregano Oil's Antimicrobial Mechanisms of Action:

- Disrupts bacterial cellular metabolism
- Makes bacterial cell membranes more permeable
- Disrupts fungal cell surfaces

Natural Antimicrobials: A multifaceted approach

The growing body of research into natural antimicrobials is an important step towards the development of multifaceted and well-balanced antimicrobial strategies. While the current state of research is yet to provide comprehensive clinical trials, a growing body of literature is suggesting that natural medicine compounds warrant consideration as potent antimicrobial agents.

Enteric coating concerns and traditional herbal applications

Antimicrobial herbs have traditionally been administered orally for treatment of infections throughout the entire gastrointestinal tract, usually as a decoction or infusion and with no enteric coating, with well-recorded efficacy. It is therefore clear from traditional use, that it is unnecessary to use enteric coatings to specifically "target" antimicrobial herbal medicines at specific areas in the GIT, in order for the herbal supplement to be efficacious in the treatment of infections throughout the gastrointestinal tract.

Essential oils have traditionally been prepared in honey, syrup, alcohol, or a base oil, to improve compliance and reduce mucous membrane irritation of the oral and oesophageal mucosae. Enteric coating is often used to replicate these effects with the unfortunate side effect of bypassing the stomach altogether. This is counterproductive in the case of gastric infections, such as *H. pylori*. Additionally, enteric coatings typically consist of phthalates and other plasticisers, which many consumers prefer to avoid due to concerns regarding their safety.

It may be preferable to deliver essential oils in a hard capsule without enteric coating. This may be achievable through the development of new methods of microencapsulating the essential oils within the structure of specially chosen dry ingredients. The sequestered essential oil may then gradually disassociate as it traverses the upper GIT, partially releasing in the stomach before the remainder is released in the small intestine.

Table 1. Antimicrobials and commonly presenting clinical conditions	
SIBO	Small Intestinal Bacterial Overgrowth (SIBO), is a condition defined by increased bacterial load in the small intestine, causing abdominal pain, bloating, flatulence, malabsorption and weight loss. This condition is increasingly well-recognised as a factor in various conditions, including Irritable Bowel Syndrome (IBS), non-alcoholic fatty liver disease (NAFLD) and deep vein thrombosis (DVT).[44-47] A broad-spectrum antimicrobial is indicated in SIBO due to the variety of organisms implicated.
Dysbiosis	Dysbiosis, or the disruption of the intestinal microbiome, has been linked with an ever-increasing range of effects on health, including neurological diseases, depression, autism, metabolic disorders, cardiovascular disease, liver diseases including NAFLD and cirrhosis, immunological diseases, inflammatory bowel disease and IBS, and multiple sclerosis. Herbal antimicrobials, probiotics and prebiotics can be used to modulate the intestinal microbiome.[48]
URTI	Upper respiratory tract infections (URTIs) are almost universally caused by viruses; only 2.4% are bacterial in origin. However, URTIs are the most common cause of antibiotic use, a significant factor in the rise of antibiotic resistance.[49] A broad spectrum herbal antimicrobial which can reduce viral load at the same time as preventing secondary bacterial and fungal infections, is an ideal adjunct to immunomodulation therapies and mucous membrane support for the treatment of URTIs.
Bacterial Vaginosis	Bacterial Vaginosis (BV) is the most common form of infectious vaginitis. It is often described as a deficiency of lactobacilli, leading to an overgrowth of a wide range of bacteria.[50] It is therefore important to repopulate the lactobacilli with a targeted probiotic supplement, in addition to treatment with a broad spectrum antimicrobial.
UTI	Urinary tract infections (UTIs) are most commonly bacterial in origin, but may also be fungal (usually candidal), viral, mycobacterial or parasitic in origin.[51] Recurrent UTIs are common, and prophylactic antibiotics are often prescribed, leading to the growth of antimicrobial resistant bacteria. The chemical complexity of herbal antimicrobials limits the ability of microbes to develop resistance, making antimicrobial herbal formulae the preferable choice for the treatment of UTIs in combination with specifically targeted probiotics.
Candida	*Candida sp* infections are usually caused by *C. albicans*, though an increasing number are caused by *C. glabrata* or *C. krusei*, which display varying degrees of resistance to standard antifungal treatments such as fluconazole.[52] Antifungal herbal medicines can be used in conjunction with probiotics to both treat the initial infection, and to help recolonise the site of infection with beneficial bacteria in order to prevent reinfection.
Chronic sinus	Chronic sinus infections are usually caused by bacteria, often with biofilm involvement, but may be viral or fungal in origin, and are often exacerbated by chronic inflammation caused by such factors as airborne pollution, structural abnormalities or allergies. Infection by multiple pathogens simultaneously is common.[53] A broad-spectrum antimicrobial with the capacity to disrupt biofilms is therefore indicated to ensure that all pathogens are treated simultaneously. Anti-inflammatories, immune support and mucous membrane support are also indicated.
Total pathogen burden	Total pathogen burden is increasingly recognised as a driver for chronic low-grade inflammation. Epidemiological studies have shown a link between total pathogen burden and the progression of coronary artery disease. Chronic infection increases the expression of inflammatory cytokines, chemokines and cellular adhesion molecules, thereby having a proatherosclerotic effect.[54] Insulin resistance has also been shown to be positively associated with a higher total pathogen burden; the mechanism for this is understood to be low-grade inflammation.[55]

Full reference list available at biomedica.com.au

©BioMedica Nutraceuticals. BioMedica is dedicated to providing high-quality and unbiased technical and education materials for healthcare practitioners. The content of these materials remains the Intellectual Property of BioMedica Nutraceuticals.
*This brochure is intended for the further education of healthcare practitioners only.