

Stabilised allicin (Allisure®) » a unique natural antimicrobial agent

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# Abstract

This paper describes the main biological properties of stabilised allicin (Diallyl thiosulfinate) and confirmed activity against multi-drug resistant bacterial infections such as methicillin resistant *Staphylococcus aureus* (MRSA). Several case studies are reported to show how stabilised allicin in capsule, cream, and liquid formulations can be used to resolve existing MRSA infections that were completely resistant to pharmaceutical antibiotic treatment. Patients generally found the treatments very acceptable and free from side effects. Clearly, stabilised allicin formulations offer a safe natural alternative or addition to drug treatment and are seen to be very effective in terms of both resolution of infection and wound healing capabilities.



# **INTRODUCTION**

# What is allicin?

The Allicin molecule is small and easily penetrates cell walls. This compound is highly active but generally unstable. Using a cold aqueous extraction method, we have obtained a novel extract of allicin that we can report is stable and highly active in vitro and in vivo against methicillin resistant *Staphylococcus aureus* (MRSA).

In 1944 an Italian chemist, C. J. Cavallito, with his colleague J H Bailey <sup>1</sup> first isolated an unstable, odourous sulphur containing compound from extracts of fresh garlic and demonstrated its antibacterial properties. The substance was named allicin, after the generic name for the plant Allium Sativum. Researchers Stoll and Seebeck, also working with garlic, discovered an odourless sulphur-containing compound called alliin which they fully characterised some years later.<sup>2</sup> This they found to be converted by a second garlic constituent, an enzyme called allinase, to form allicin. The researchers made an additional remarkable discovery: When they studied cloves in cross section they found that alliin and allinase are stored in different compartments. In an undamaged clove they remain completely separate, but once its structure is ruptured – typically by cutting – the two substances come into contact and form allicin.

This transformation is extremely rapid, taking mere seconds. And even more intriguing is the instability of the allicin. It remains active only for a short period before degrading. It is now generally accepted that garlic has evolved a defence mechanism against attack from the soil-borne organisms. It has been found that invasion of growing garlic cloves by fungi, virus and other soil pathogens causes the alliin and allinase to react, rapidly producing localised bursts of allicin which deactivates the invaders. This ability underlies the exceptional capacity of allicin to kill unwanted organisms. The reason why the highly reactive allicin molecules have such a short working life is that if they didn't they would continue to react with surrounding proteins, including the allinase enzyme itself, and this would use-up the garlic's protection, which it might need later. This extremely efficient binary chemical mechanism ensures that the clove's defence is highly localised and shortlived - just sufficient to repel an attack. The remaining alliin and allinase are held in reserve to fight off any subsequent attacks. While this is good for the well-being of a garlic crop, it poses distinct problems for anyone trying to extract and isolate the key active ingredient in a way that is beneficial. It was 3 decades after its initial discovery that allicin would be isolated in a stabilised form for the first time.<sup>3</sup> Chemists know that this configuration is highly reactive, giving allicin its remarkable antibiotic properties and in particular the potential to assist the immune system in a number of important ways, including stimulating immune cells, killing pathogens and detoxifying carcinogens.

Before the advent of pharmaceutical antibiotics, crushed garlic extracts were used to treat a wide range of infectious diseases including dysentery, typhoid, cholera, smallpox and tuberculosis. Then, in the 1920's, the first class of antibiotic drugs were invented, the sulphonamides.<sup>4</sup> The reason they were so successful was the presence of the reactive sulphur group – exactly the same group that allicin contains. Pharmacokinetic studies



indicate that allicin will reach a maximum level in the blood after 30-60 minutes and may still be present 72 hours later with more than 85% clearance through urinary and faecal pathways.<sup>5</sup>

Biological activity is the key to killing drug resistant infections and in particular methicillin resistant *Staphylococcus aureus* (MRSA) is commonly related to delayed closure for many chronic and acute wounds. This is associated with high levels of bacteria in tissues but they can also close through toxin secretion. These toxins can cause local necrosis and disrupt the delicate balance of critical mediators such as cytokines and proteases necessary for healing progression. Allicin has been reported to have a strong SH-modifying and antioxidant properties.<sup>6</sup> Allicin reacts very rapidly with free thiol groups, via a thiol-disulphide exchange reaction. The main antimicrobial effect of allicin may be due to its chemical reaction with thiol groups of various enzymes, e.g. alcohol dehydrogenase, thioredoxin reductase, and RNA polymerase.<sup>6</sup> In previous work we have demonstrated that allicin is bactericidal against MRSA at concentrations of 128 to 256 ug/ml.<sup>7,8,9</sup>

# WHY IS MRSA SUCH A GLOBAL PROBLEM?

# Introduction

In recent months reports from California, The Netherlands, Great Britain and several other European Countries suggest that MRSA (methicillin resistant Staphylococcus aureus) is now beginning to infect thousands of people across America and Europe. It is now a fact that a new strain is emerging that spreads through skin contact and can even infect healthy people – Community Acquired MRSA or cMRSA.<sup>10</sup> The United States Centre for Disease Control in Atlanta stated "We are greatly concerned that MRSA has emerged into the healthy population". The strain has been spreading guickly in crowded jails and across cities and towns all over the United States. Athletes, schoolchildren, homosexuals and newborns have all fallen victim to this superbug. The infection usually appears as sores that resemble insect bites and nasty boils and abcesses can develop usually requiring repeated courses of antibiotics and sometimes surgery. The worry is that it could reach the lungs or bloodstream where it could cause pneumonia or septicaemia. Furthermore in San Francisco the emergence of a strain identified as USA300 has caused concern as it is infecting suburban mothers, executives, doctors, athletes and children. It has turned up in tattoo parlours and newborn nurseries. People with HIV infection seem especially prone to it,<sup>11</sup> but it also strikes patients who have no previous health problems.<sup>10</sup> Antibiotic use continues unabated and we have also seen cases resistant even to vancomycin.<sup>12</sup> All manner of infections with Staphylococcus aureus can be dangerous especially in weakened patients, particularly if they can't be cleared up quickly with antibiotic treatments. MRSA infections can prove tough to treat because they are resistant to treatment, making them more dangerous than a simple infection.

Doctors are very worried about what the future holds for MRSA. The number of reports of MRSA infections rises year by year (over 90,000 last year in the USA) – and the latest evidence suggests that deaths due to MRSA are increasing at a similar rate. Already, the spectre of a bug resistant to all antibiotics is approaching. VRSA, or vancomycin resistant *Staphylococcus aureus*, has acquired resistance to a drug considered the "last line of

defence" when all other antibiotics have failed. The UK has already seen several cases of GISA, or glycopeptide intermediate resistant *Staphylococcus aureus*, a kind of "halfway house" between MRSA and VRSA, which has developed a resistance to antibiotics of the vancomycin family.<sup>13</sup> This is already a common infection in Japan and the USA.<sup>12</sup>

Although new antibiotics are being developed all the time, pessimistic experts believe it is only a matter of time at current rates until virtually every weapon in the pharmaceutical arsenal is nullified.

The government is already trying to at least slow down the apparently relentless march of the bacterial Super-germs; virulent, multiple antibiotic resistant bacteria such as MRSA, VRSA, GISA, PRSP, MDRTB and Acinteobacter baumanii are a major problem throughout the world.<sup>13</sup> Given the failure of conventional antibiotics to control and eradicate these micro-organisms, the search for a more natural solution has provided a safe and effective answer. Fortunately the first commercial production of stabilised allicin in the United Kingdom has now been demonstrated to be capable of killing even multi-drug resistant strains of *Staphylococcus aureus* as well as VRSA, GISA, PRSP and MDRTB.<sup>14-19</sup> Recently, work completed in our Department of Life Sciences at The University of East London has shown that all the formulations can not only kill a wide range of different strains of *Staphylococcus aureus* but can also kill the drug resistant strains.<sup>15</sup> Confirmation of this ground breaking work has recently been reported in several medical journals including presentations at the American Society of Microbiology (Atlanta 2005)<sup>17</sup> and the European Congress of Chemotherapy and Infectious Diseases (ECCMID Copenhagen 2005)<sup>17</sup>. All forms of stabilised allicin that carry the branded name Allisure® may have a key role to play in the fight against the so called MRSA "superbug" and the way it infects patients whilst in hospital, care home, nursing home or community setting. Many healthcare workers may also find that a natural allicin based cream/gel and capsule intake may reduce their own risk of developing an infection. With very few staff screening programs in place anywhere in the westernised world it is estimated that 1 in 2 health care workers actually carry Staphylococcus aureus in their nasal cavities<sup>12</sup> and this could represent a simple route to re-infection and potential resistance.

Control of the spread of antibiotic-resistant bacteria and the treatment of infections caused by them is a major problem worldwide. In particular, methicillin-resistant *Staphylococcus aureus* (MRSA) presents major infection control problems for patients and hospital staff, as its incidence in Europe has risen from 3% in 1992 to 37% in 1999. Topical agents are important in controlling the carriage and spread of MRSA. Mupirocin (pseudomonic acid), a fermentation product produced by *Pseudomonas fluorescens* (NCIB 10586), is a standard product used to deal with MRSA carriage and to prevent its spread. It has also proved to be an effective treatment for skin infections and plays a crucial role in the control of MRSA outbreaks.

However, resistant strains were described soon after its introduction. Moreover, the increased use of pseudomonic acid, especially for chronic infections, has led to an increased incidence of resistance. In a recent survey from Spain, levels of resistance in clinical isolates was reported to have increased from 7.7% in 1998 to 19% in 2000, and some hospitals have reported incidences as high as 63%.<sup>19</sup> The continuing spread of



MRSA and the increase in pseudomonic acid -resistant strains highlight the need for alternative topical agents such as a stabilised allicin cream or gel formulation.

# Volunteer patient recruitment

In excess of 50% of the volunteers/patients on this study program, numbering 52 in total, suffered wounds or lesions as a result of an operation on their leg, hip, back or knee. Some wounds varied in size from 20mm to 100mm diameter. Other wounds covered the shin area, the area around and above the ankle, the back of the leg, the hip joint, the stump end of an amputated leg and pressure sores on the buttocks. All wounds were first checked for MRSA using swabs. These swabs were analysed at the University of East London, Department of Life Sciences, and all were confirmed and graded as MRSA positive.

For such wounds, it was recommended that the patient take 1350mg of stabilised allicin powder capsules per day, apply the stabilised allicin liquid in spray form and where appropriate apply stabilised allicin cream to the infected area twice daily and then apply their dressing in the normal fashion. The concern was that this procedure would keep the wound wet and therefore delay or prevent healing. Within a few weeks, it became apparent that the wounds were not healing as well as expected. So, it was decided to change tactics and recommend to the patient what could be described as "Dry wound" healing. This change of routine involved the use of the stabilised allicin spray only. Patients were asked to spray three times daily and then leave the wound open to the air as long as possible to accelerate the healing process. All patients were very keen to try this and, without exception, it had the desired result. Wounds healed faster and any pain associated with the wound was reduced. MRSA levels in the wounds were checked by swabs taken on a six weekly basis. The dosing of 1350mg of stabilised allicin powder capsules per day remained unchanged.

There was some concern that leaving wounds open could be embarrassing or distressing for the patient and, if this was the case, it was recommended that the patient should cover the wound with a dry dressing. Fortunately, most of the patients were homebound and they were all able to carry out the "dry wound" healing procedure. However, upon retiring to bed, it was recommended that all patients cover their wounds with a dry dressing for protection.

One of the noteworthy features of applying the stabilised allicin spray was that it took about 2 to 3 minutes for the liquid to penetrate and dry on the infected area. It was believed that applied three times daily enabled the allicin in the spray to continually act on the MRSA bacteria. The benefits of this on the wound were evident within a few weeks. Most wounds healed between 8 and 12 weeks of starting treatment. Other, larger wounds took up to 18 weeks.

We present 3 case reports from the initial program concerning individuals with confirmed MRSA infections at multiple sites. All had been treated with a series of pharmaceutical antibiotics (oral, topical and intravenous) without effect and had therefore been discharged



home to deal with their condition in a familiar environment. All had a carer and various medical staff involved in either daily or weekly visits to re-dress open wounds. The effect on their lifestyle was considerable and all reported feeling very frustrated at being told "nothing could be done" to improve their potential to heal the wound sites and return to a normal lifestyle.

# Case 1 Female, 26 years old.

DB's wounds were on her spine. One close to the top, which was approximately 20mm by 15mm this was over-granulated and weeping. The other is approximately 7.5mm by 5mm and near her waistline this was also over-granulated and weeping a little. She had had a major spinal operation two years previously and although she has had many antibiotic treatments nothing has cleared her MRSA infection. Many courses of both oral antibiotics and creams over several months were prescribed and nothing has been able to shift the distressing MRSA infection. The patient had received continuous antibiotic treatment for approximately 2 years both in hospital and in her home environment without success. The only option available to her via the hospital was to have all the metalwork removed. This was not the patients preferred option as she did not want to go back into hospital nor did she want the metalwork removed.

Following confirmation from swab samples that both the wound and nasal cavities were infected with MRSA, sensitivity tests toward stabilised allicin were performed and showed very good activity against this particular strain of MRSA.

The patient was given a daily dose of 1080mg of stabilised allicin in capsule form and asked to apply allicin cream at least twice daily. She was also asked to insert a "pea sized" sample of cream into each nostril 2/3 times each week. This treatment regimen continued for 4 weeks, at which time both wounds had closed. Swab samples confirmed that MRSA was absent from the wound site and nasal cavity.

The patient commented that "Having had these two wounds on my back weeping for 2 years I don't know quite how to thank you and hope that I get the opportunity to thank you in person at some point. I will also be telling anyone who may benefit from Allicin how miraculous it is. I am going to the hospital on Thursday. I am not sure if my consultant can quite believe what has happened, as he, along with some of my district nurses, are not too happy about the thought of using alternative remedies. When I think how many courses of antibiotics I have been instructed to take in the last 2 years and how many biopsies came back positive for MRSA, I am not surprised that the medical staff cannot believe it! Thank you once again for all you have done. You saved me from another horrendous operation."





Figure 1 » Chronic wound infection-releasing pus before treatment with stabilised allicin formulations.



Figure 2 » Both wound sites have healed following 4 weeks treatment with stabilised allicin capsules and cream.

#### Case 2 Female 14 years old

Miss SC is a Female, 14 yrs old with 2 pins inserted into her spine. Both wound sites failed to heal for 18 months. Patient had several courses of antibiotics with no effect. Surgeons were keen to re-admit her and replace the pins and deal with the infection. Her medical staff had confirmed that she was MRSA positive at the wound site.

We tested her as ++MRSA sensitive to stabilised allicin





Figure 3 » Miss SC showing 2 open wounds on her spine before treatment with stabilised allicin powder capsules only

Treatment was 1800 mg of allicin powder capsules daily in divided doses (Allisure® capsules only) for a period of 4 weeks. No creams or spray were needed in this case.

At 4 weeks, both wounds had healed completely and swabs post treatment showed no MRSA infection present in the healed area, groin or nasal cavity.



Figure 4 » Miss SC's spine following 4 weeks treatment. Both sites are completely healed. The patient remains healed and reports that she has been able to start wall climbing again after a 2 year break and her parents are both delighted that she has got her young life back again.

Case 3 Details of the patient DB. A 67 year old woman who had a leg amputated due to poor circulation in January 2004.

Her history reveals that in January 2004, following her amputation, she was due to leave hospital in February and the leg stump wound became infected. A swab was taken in



March 2004 and MRSA of an unknown strain was confirmed. It is believed that she had had MRSA for up to 2 months. The patient also suffers from arthritis/rheumatism. The patient also suffers sometimes from an acid stomach. The patient also takes medication for her heart. Following a course of stabilised allicin capsules cream and spray treatment the patient's wound closed and no trace of infection could be found at the site or in her nasal cavities.

Later that month another operation was carried out to remove some bone so that an artificial limb can be fitted. Unfortunately the patient contracted MRSA, whilst in hospital for the second time. However following a short course with stabilised allicin formulations once again her swabs were negative for MRSA infection.

Clinical Symptoms	Infection on leg stump 10cm x 2cm	
Medical Treatment	Glyceryl Tinitrate	2.6 1x2 a day
	Ranitidine	150mg 1x2 a day
History	Asprin	75mg 2x1 a day
	Celliprolol Hydrochloride	200mg 1x1 day
	Zomorph Sulphate	10mg 1x1 day
	Liquid Paraffin & Magnesium Hydroxide	10ml-20ml 2x day
	Diclofenac Diethylammonium	1.16 apply 3x a day
	Peptac Liquid	20ml 4x a day
	Frusemide	20mg 1x a day
	Clopidogrel Hydrogen Sulphate	75mg 1x a day
	Amitriptyline Hydrochloride	25mg 1 daily
	Atorvastatin	20mg 1 daily
	Gabapentin	300mg 1x3 daily
	Co-Codamol	8/500 2x4 times a day
Devices Units	Oinne flaure eine	
Previous History		
	Doxycycline	
Allergic to	Amovil	
Allergie to	Distrodos	
	Septrium	
	Ceprim	
	Penicillin	
	NSAID's	
		1

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Allicin treatment	1350mg allicin powder caps.	Daily
	1-2 sprays of allicin liquid	Daily
	1-2 applications of allicin	Daily
	cream	

#### **Patient response**

Patient reported higher energy levels after approximately 1 weeks treatment. No side effects were reported and concomitant use with aspirin and clopidogrel did not pose any problems at all.

Patient delighted to achieve a wound closure and following the second removal of MRSA infection now reports being able to wear a calliper and go dancing! Patient continues to take 450mg of stabilised allicin powder capsules daily.

# **Further Information**

Although a few relapses have happened over the months the wound is now completely clear of MRSA. The wound site started to breakdown during the summer months due to the hot weather and rubber sleeve worn over leg stump, but the MRSA did not return. January 2006 - Patient now has prosthesis and is bowling and dancing again after many years.

As of April 2008, patient is still on maintenance dose of 450mg of stabilised allicin powder daily and still clear of MRSA.

Picture below shows weeping inflamed lesion (before allicin treatment). The other pictures show the gradual progression of healing through "dry wound" treatment. No side effects were reported by the patient.









Pictures of leg wounds before and after 2 months treatment using only stabilised allicin powder capsules, liquid and cream formulations







# Discussion

The Allicin molecule is small with a molecular mass of 162 and easily penetrates cell walls. This compound is highly active but generally unstable. Using a cold aqueous extraction method, followed by proprietary freeze drying, we have obtained a stabilised novel extract of allicin (Allisure®) that is highly active *in vitro* and *in vivo* against methicillin resistant *Staphylococcus aureus* (MRSA) in powder, liquid, cream and gel formulations. MRSA is commonly related to delayed closure for many chronic and acute wounds. This is associated with high levels of bacteria in tissues but they can also close through toxin secretion. These toxins can cause local necrosis and disrupt the delicate balance of critical mediators such as cytokines and proteases necessary for healing progression. Allicin has been reported to have a strong SH-modifying and antioxidant properties. Allicin reacts very rapidly with free thiol groups, via a thiol-disulphide exchange reaction. The main antimicrobial effect of allicin may be due to its chemical reaction with thiol groups of various enzymes, e.g. alcohol dehydrogenase, thioredoxin reductase, and RNA polymerase.<sup>5</sup>



We presented initial findings from a number of patients who have completed a course of treatment. These courses consisted of stabilised allicin capsules (1350 mg allicin powder per day); spraying liquid stabilised allicin (1000 ug ml-1) onto the affected areas once per day and applying stabilised allicin cream (500 ug ml-1) to the infected area once daily. Patients reported an improvement in their condition after 2 to 6 weeks treatment and the infections resolved in 3 to 4 months. Although the timescales required for treatment may be longer than those normally required using antibiotics, the initial relief from weeping ulcers and pain was much quicker. It should be noted that these patients had been receiving unsuccessful treatment with antibiotics for months or years prior to treatment with allicin.

A possible reason for the initial relief from symptoms could relate to the reported activity of allicin extracts to neutralise bacterial exo-enzymes *in vitro*. In initial studies we have demonstrated that even after brief exposure, allicin can reduce the activity of microbial enzymes. The activity of alcohol dehydrogenase in producing NADH from NAD+ reduced as allicin in the solution increased. Patients were all screened and both nasal and wound swabs were tested for MRSA prior, during and after treatment. All patients were nose and wound swab MRSA positive prior to treatment.

We have successfully treated both community acquired and hospital acquired infections. The strains isolated from each patient were tested *in vitro* against stabilised allicin powder and liquid.

It appears that Allicin has the potential to reduce the activity of extracellular virulence factors since many patients got relief from their symptoms before the MRSA were fully removed from the lesion site. Generally the product formulations were very well tolerated and did not appear to cause any side effects. Many patients have continued on long term treatment with capsules to try and prevent symptoms from recurring. It seems that long term treatment can improve the immune system significantly. Stabilised allicin formulations can be safely added to almost any other pharmaceutical agent including Warfarin and Coumadin. However, friendly bacteria such as Lactobacillus spp, Enterococcus spp. and Pediococcus spp. are concentration dependent to allicin and currently not affected by the treatment protocols we have adopted. It has also been noted that allicin is synergistic with antibiotics.<sup>19</sup> Further research also indicates that various multi-drug resistant bacteria are also effectively killed by allicin. This includes a broad range of bacteria, including E.coli, Staphylococcus Aureus, Streptococcus pyogenes, Proteus mirabilis, Pseudomonas aeruginosa, Penicillin Resistant Streptococcus Pneumoniae, Acetobacter baumanii, Klebsiella pneumoniae. Enterococcus faecium. Mycobacterium tuberculosis. Helicobacter pylori, Salmonella, Clostridium spp and Shigella are all allicin-sensitive. Some of the bacteria listed are killed by allicin concentrations as low as 3-15 ppm (3-15ppm (15-30 mcg/ml).<sup>19</sup> Allicin also has a powerful antifungal effect, with a minimum inhibitory concentration (MIC) against various Candida species of only 0.15 to 0.8 mcg/ml. Allicin is effective against other fungal species including Cryptococcus, Trichophyton, *Epidermophyton*, and *Microsporum* at MIC's of 1.57-6.25 mcg/ml.<sup>19</sup>

As we enter a new frightening age of multi-drug resistant organisms it is nice to know that with a little new technology and some help from mother nature we still have a better than even chance of defeating the so called "superbugs"



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# Table 1

Bacterial infections against which Allisure® allicin powder, cream, gel or liquid may be effective (all with very low concentrations required to prevent infection).

Acinetobacter baumanii, Acinetobacter calcoaceticus, Escherichia coli, Bacillus cereus, Bacillus subtilis, Campylobacter jejuni, Camplylobacter fetus, Campylobacter coli, Campylobacter doyley, Campylobacter hyointestinalis, Campylobacter ureolyticus, Campylobacter mucosalis, Campylobacter helveticus, Clostridium difficile, Citrobacter spp., Hafnia spp., Provindencia spp., Micrococcus, Mycobacterium tuberculosis, Mycobacterium leprae, Mycobacterium bovis, Mycobacterium kansasii, Helicobacter pylori, Cornyebacterium spp., Pasteurella spp., Cryptococcus spp., Salmonella typhimurium, Salmonella dublinii, Salmonella enteriditis, Shigella, Bacillus anthracis, MRSA (Methicillin resistant staphylococcus aureus), GISA (Glycopeptide intermediate resistant staphylococcus aureus), VISA (Vancomycin intermediate resistant staphylococcus aureus), Staphylococcus aureus in skin disease, Streptococcus pyogenes (flesh eating bacteria), Klebsiella aerogenes, Saccharomyces cerevisiae, Meningitis, Cholera, Listeria monocytogenes, Enterococcus faecium, Pseudomonas aeroginosa, Proteus mirabilis, Nesseria Gonorrhea, Clostridium perfringens, Chlamydia, Haemophilus Influenza, Enterococcus histolytica.

Direct observation reported in Scientific literature, recorded at The University of East London or personal communication to the authors