Acetic acid used for the elimination of Pseudomonas aeruginosa from burn and soft tissue wounds

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SUMMARY: Acetic acid was used topically at concentrations of between 0.5% and 5% to eliminate Pseudomonas aeruginosa from the burn wounds or soft tissue wounds of 16 patients. *In-vitro* studies indicated the susceptibility of *P. aeruginosa* to acetic acid; all strains exhibited a minimum inhibitory concentration of 2 per cent. *P. aeruginosa* was eliminated from the wounds of 14 of the 16 patients within two weeks of treatment. Acetic acid was shown to be an inexpensive and efficient agent for the elimination of *P. aeruginosa* from burn and soft tissue wounds.

Introduction

For many years different agents have been applied to burn wounds in an attempt to reduce the risk of septic complications developing in, and arising from, the wounds. Compounds such as polymyxin, gentamicin, sulphonamides, silver nitrate, mafenide acetate, povidone iodine and silver sulphadiazine have been used in an attempt to reduce bacterial colonisation. Many of these are toxic or are obsolete because of antibiotic resistance (1).

One third of burn wounds (59% of extensive burns) will yield *Pseudomonas aeruginosa* (2); this species causes the most common Gram-negative bacteraemia in patients with burns, carries a high mortality (3), and includes strains with reduced susceptibility to many antimicrobial agents (3). Furthermore, *P. aeruginosa* is the main cause of cross infection in burns units although other Gram-negative bacilli and *Staphylococcus aureus* are also implicated (4).

In 1916, Taylor (5) reported that application of a 1 per cent solution of acetic acid for a period of two weeks to purulent war wounds infected with "Bacillus pyocyaneus" led to the elimination of the organism. More recently, Phillips et al (6) demonstrated the efficacy of locally applied acetic acid, as compared with either chlorhexidine or hypochlorite, for the elimination of *P. aeruginosa* from superficial skin and burn wounds in a tropical climate. We report the results of a prospective clinical study undertaken in the burns unit of this hospital further to evaluate the ability of topical acetic acid to eliminate *P. aeruginosa* from colonised and infected burn and chronic superficial wounds.

Patients and Methods

Eight patients (5 males, 3 females, ages 23-75 years) had skin ulcers as previously described (7), and 8 patients (4 males, 4 females, aged 14-88 years) had burn wounds of between 2 and 30 per cent total body surface. All patients had purulent wounds or wound breakdown with delayed healing; *P. aeruginosa* was isolated from surface cultures of each wound on two occasions one week before treatment.

Glacial acetic acid (BDH Laboratory Supplies) was diluted in sterile water for Injection BP to 10 per cent. Sterile gauze swabs soaked in dilutions of between 1 to 5 per cent acetic acid were applied to the ulcers and to smaller burn wounds for 15 minutes twice daily for between one and two weeks. In addition, bath water containing approximately 0.5 per cent acetic acid was used to immerse each of 4 patients with purulent burn wounds for 20 to 45 minutes each day; during this time wounds were debrided and dressed.

The Minimum Inhibitory Concentration (MIC) *in-vitro* of acetic acid was determined for all strains of *P. aeruginosa* isolated from all wounds and for other bacterial species isolated from burn and soft tissue wounds.

Iso-sensitest agar plates (Oxoid Ltd, CM 471) were flooded with each organism to a density of 10^5 colony forming units (cfu); Iso-sensitest agar containing 5% v/v laked blood was used for haemolytic streptococci.

Glacial acetic acid was diluted to 10 per cent and 1 per cent in sterile water. Concentrations of acetic acid 0.1 per cent to 10 per cent were applied to either the surface of the agar plates or instilled in wells cut into the agar. Plates were incubated at 37°C for 18 hours and inhibition zones measured and recorded. Zones of inhibition obtained for clinical isolates were compared with those obtained for reference strains *Pseudomonas aeruginosa* NCTC 10662 and *Staphylococcus aureus* NCTC 6571.
Results

All patients tolerated acetic acid soaks or immersion, although 3 patients complained of stinging on application to wounds when used at 5 per cent, and most patients observed the odour of acetic acid; these factors did not necessitate interruption of treatment.

When used as soaks, a concentration of 5 per cent was the most effective for eliminating *P. aeruginosa* from wounds; the organism was eliminated from two wounds within 2 days, 6 wounds within 5 days and 2 wounds within 7 days. The longest period for elimination was 14 days. Despite treatment for 28 days, the burn wounds of one patient, an 88 year old female, remained colonised within 7 days. The longest period for elimination was 14 days. The organism was eliminated from two wounds within 2 days, 6 wounds within 5 days and 2 wounds within 7 days. The longest period for elimination was 14 days.

Follow-up surface cultures of all wounds, twice weekly, showed that once bacteria were eliminated, wounds remained free from *P. aeruginosa*, and healing occurred; no significant failures of skin grafts occurred in this group. Wounds consistently remained colonised with *Staphylococcus aureus* and occasionally *Proteus* species.

The MIC of acetic acid which produced an inhibition zone for clinical isolates from burn wounds is shown in Table 1. All organisms tested, with the exception of *Staphylococcus aureus* and micro cocci, were inhibited *in vitro* by acetic acid at a concentration of between 1 and 5 per cent. Inhibition zones were well defined with little variation in size despite increases in the concentration of acetic acid.

### Table 1

<table>
<thead>
<tr>
<th>Organisms</th>
<th>No. of isolates</th>
<th>Concentration of acetic acid</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aeromonas hydrophila</td>
<td>1</td>
<td>1%</td>
</tr>
<tr>
<td><em>Pseudomonas aeruginosa</em></td>
<td>18</td>
<td>2%</td>
</tr>
<tr>
<td><em>Pseudomonas stutzeri</em></td>
<td>1</td>
<td>2%</td>
</tr>
<tr>
<td><em>Pseudomonas maltophilia</em></td>
<td>1</td>
<td>2%</td>
</tr>
<tr>
<td>Streptococcus pyogenes Gp A</td>
<td>2</td>
<td>2%</td>
</tr>
<tr>
<td>Streptococcus Gp G</td>
<td>1</td>
<td>2%</td>
</tr>
<tr>
<td>Corynebacterium sp</td>
<td>1</td>
<td>2%</td>
</tr>
<tr>
<td>Bacillus cereus</td>
<td>2</td>
<td>2%</td>
</tr>
<tr>
<td>Escherichia coli</td>
<td>4</td>
<td>4%</td>
</tr>
<tr>
<td>Klebsiella pneumoniae</td>
<td>2</td>
<td>4%</td>
</tr>
<tr>
<td>Salmonella typhimurium</td>
<td>1</td>
<td>4%</td>
</tr>
<tr>
<td>Flavo. meningosepticum</td>
<td>1</td>
<td>4%</td>
</tr>
<tr>
<td>CDC VE Type I</td>
<td>1</td>
<td>4%</td>
</tr>
<tr>
<td>Proteus mirabilis</td>
<td>3</td>
<td>4%</td>
</tr>
<tr>
<td>Serratia marcescens</td>
<td>3</td>
<td>4%</td>
</tr>
<tr>
<td>Serratia liquifaciens</td>
<td>1</td>
<td>4%</td>
</tr>
<tr>
<td>Enterobacter cloacae</td>
<td>3</td>
<td>4%</td>
</tr>
<tr>
<td>Acinetobacter calcoaceticus</td>
<td>3</td>
<td>4%</td>
</tr>
<tr>
<td>Micrococcus sp</td>
<td>3</td>
<td>8%</td>
</tr>
<tr>
<td>Staph aureus</td>
<td>4</td>
<td>8-10%</td>
</tr>
<tr>
<td>Methicillin-resistant Staph aureus</td>
<td>4</td>
<td>8-10%</td>
</tr>
</tbody>
</table>

*This includes three strains of *Pseudomonas aeruginosa* isolated from the wounds of casualties returning from the Gulf Conflict 1991.*

Discussion

Infection continues to be the predominant cause of morbidity and mortality in patients with burn wounds (8). Burn wounds readily become colonised with many species of potentially pathogenic bacteria including *P. aeruginosa*. Nevertheless, burn and other skin wounds will heal despite heavy bacterial colonisation (9), though numbers greater than 10^5 cfu g^-1 have been associated with a higher risk of infection and failure of skin grafts (10, 11).

In the absence of infection, wound healing has been shown to be adversely affected by toxic metabolites from *P. aeruginosa*, Streptococci group A and *Staphylococcus aureus* (12). The pigment pyocyanin produced by *P. aeruginosa* is one of the many extracellular substances and proteases that are toxic to skin epithelium, cause tissue destruction, impairment of local and microbial defence mechanisms and cleavage of immunoglobulins (13, 14, 15). These pigments also have an inhibitory effect on other micro-organisms suggesting an advantage for this species over others in a bacterial population (16).

The results obtained in this study were similar to and further confirm, the results of Phillips et al (6) and indicate that acetic acid remains effective for the elimination of *P. aeruginosa* from burn and other skin wounds and leads to clinical improvement and wound healing.

Acetic acid (5 per cent ) used as topical soaks twice daily was the most effective method, the organism being eliminated within 7 days and wounds remaining free from *Pseudomonas*.

Acetic acid has a narrow anti-bacterial spectrum as previously described (6). Although *P. aeruginosa* was consistently eliminated from wounds together with certain Gram-negative bacilli, Gram-positive organisms and *Proteus* species continued to colonise wounds. These clinical observations were verified by the *in-vitro* inhibition tests for clinical isolates, which showed that all strains of *P. aeruginosa* were inhibited by acetic acid at 2 per cent. Studies have shown that the routine use of antibiotics does not prevent the bacterial colonisation of patients with burn wounds, and may result in colonisation with antibiotic resistant strains (17, 18).

The choice of antibacterial agents for topical use on burn wounds is limited. Although silver sulphadiazine is widely used in burn units in the UK, problems of bacterial resistance have occurred during prolonged use (19). In view of the results in this and previous studies, we conclude that the use of acetic acid (5 per cent) topically to burn and soft tissue wounds is an effective treatment for reducing the incidence of *P. aeruginosa*, avoiding the expense and adverse effects of other topical agents and antibiotics.
Acknowledgements

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REFERENCES