EVALUATION OF LACTIC ACID AS AN ANTIBACTERIAL AGENT

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Summary

Fifty strains each of *Staphylococcus aureus*, beta haemolytic streptococci, *Proteus* species, *Esch.* coli and *Pseudomonas aeruginosa* were subjected to 2%, 1% and 0.1% lactic acid in peptone water. Minimum inhibitory concentration of lactic acid for all the strains of each of these organisms was 0.1% or 1%. Depending upon its concentration, lactic acid added to peptone water brings down the pH to 2.5 - 4 which by itself has some inhibitory effect on the micro-organisms. Lactic acid however, retains its inhibitory effect even if the pH of the peptone water is brought back to 7.3. Lactic acid is a non-toxic and non-sensitizing agent because it is a normal metabolite of the body. Thus, it can be used as a safe and effective antibacterial agent for local application.

Lactic acid is an intermediate product of glucose metabolism and is present in almost all tissues. When incorporated in a balanced ionic solution containing serum albumin, it inhibited the growth of **Staphylococcus aureus** and **Mycobacterium tuberculosis** in vitro[1]. In diabetics, the levels of lactic acid in skin were found to be lower in patients having skin infection[2] which suggests that lactic acid may be having a significant role in protecting the skin against micro-organisms, more so because lactic acid is an important constituent of sweat and accounts for the acidity of the skin surface. The present study was undertaken to establish the antibacterial spectrum of lactic acid with a view to use it as an antibacterial agent.

Material and Methods

In a pilot study, 10 strains each of Staphylococcus aureus and Pseudomonas aeruginosa were inoculated in a peptone water medium containing lactic acid in concentrations of 10, 5, 2, 1, 0.1 and 0.01 percent. All the strains were found to be inhibited by concentrations of lactic acid as low as 2% while 0.01% lactic acid was unable to inhibit any of the strains.

Subsequently, 50 strains each of Staphylococcus aureus, Pseudomonas aeruginosa, Proteus species and Esch. coli were cultured in peptone water at 37°C for 4 hours, while beta haemolytic streptococci were cultured in heart infusion broth (Difco) at 37°C for 16 to 18 hours. The growths of beta haemolytic streptococci thus obtained were diluted 1000 times with heart infusion broth, while those of other organisms were used as such. One drop of these growths was added to each of three tubes containing 0.1, 1.0 and 2.0 percent of lactic acid in 3 ml peptone water. A 4th tube containing 3 ml of only peptone water was also inoculated to serve as a control. All these tubes were incubated at 37°C for 16 to 18

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hours and then subcultured on nutrient agar to check for the presence of live organisms. Beta haemolytic streptococci were subcultured on sheep's blood agar. The minimum concentration of lactic acid which completely inhibited the growth of a particular strain was considered to be the minimum inhibitory concentration (MIC) of lactic acid for that organism.

Results

None of the organisms was found to be resistant to lactic acid. Lactic acid in 0.1% concentration was sufficient to inhibit all the strains of Proteus species and Esch. coli, 23 strains of Staphylococcus aureus, 32 strains of beta haemolitic streptococci and 31 strains of Pseudomonas aeruginosa. The remaining strains were susceptible to 1% of lactic acid except one strain of Staphylococcus aureus which required 2% lactic acid for complete inhibition (Table 1).

TABLE 1

Susceptibility of different organisms to various concentrations of lactic acid in the culture medium.

Organism	Number of strains having minimum inhibitory concentration of lactic acid 2%	1%	0.1%	Total
Pseudomonas aeruginosa	0	19	31	50
Staphylococcus aureus	1	26	23	50
Beta haemolytic streptococci	0	18	32	50
Proteus species	0	0	50	50
Escherichia coli	0	0	50	50

Discussion

The chief drawbacks of the antibacterial agents currently available for local application include, (1) contact hypersensitivity, and (2) bacterial resistance. Lactic acid is a natural metabolite of the body and is freely available in all tissues. There are therefore, practically no chances of an individual becoming allergic to lactic acid or developing a toxicity reaction following percutaneous absorption. Moreover, lactic acid seems to have a wide range of antibacterial activity, because it was able to inhibit all the strains of even Pseudomonas aeruginosa and Staphylococcus aureus which are well known to show resistance to other commonly used antibiotics.

The mode of action of lactic acid has not yet been worked out but addition of lactic acid in concentrations of 0.1% to 2% results in lowering of the pH of peptone water to between 4 and 2.5. To assess if the antibacterial activity of lactic acid depends upon only lowering of the pH, culture tubes were prepared with peptone water and brought to pH 2.5., 3.0 and 4.0 by the addition of HCl. 10 strains each of Staphylococcus aureus, Pseudomonas aeruginosa, beta haemolytic streptococci, Proteus species and Esch. coli were inoculated into each of these tubes. All the strains of each of these organisms were completely inhibited at pH 2.5 and 3.0 while 3 strains each of Pseudomonas aeruginosa and Staphylococcus aureus and 7 strains of Esch. coli were able to grow at pH 4.0 (Table 2). In a second experiment,

TABLE 2

Effect of lowering the pH of the culture medium with HCl.

Organism	Number of strains tested	Number of strains showing growth in the culture medium containing HCl to make the pH 2.5	3	4
Pseudomonas aeruginosa	10	0	0	3
Staphylococcus aureus	10	0	0	3
Beta haemolytic streptococci	10	0	0	0
Proteus species	10	0	0	0
Escherichia coli	10	0	0	7

the same 10 strains of each of these organisms were inoculated in peptone

water containing 0.1., 1.0 and 2.0% lactic acid in which the pH of the medium had been brought back to 7.3 with sodium hydroxide. All the strains of these organisms were completely inhibited by 2.0 and 1.0% concentrations of lactic acid while 0.1% lactic acid was not able to inhibit any of the organisms (Table 3) indicating that the

TABLE 3

Minimum inhibitory concentration of lactic acid for various organisms after the pH of the culture medium had been brought back to 7.3 with NaOH

Organism	Number of strains tested	Number of strains showing growth in the culture medium containing various concentrations of lactic acid neutralised with NaOH to pH 7.3		
		2%	1%	0.1%
Pseudomonas aeruginosa	10	0	0	10
Staphylococcus aureus	10	0	0	10
Beta haemolytic streptococci	10	0	0	10
Proteus species	10	0	0	10
Escherichia coli	10	0	0	10

inhibitory effect of lactic acid does not depend only upon lowering of the pH of the culture medium. Local applications of 5% lactic acid to the skin of human volunteers have been found to be non-irritant and non-sensitizing[3] and thus there is a significant margin between the therapeutically effective concentration and the clinically safe concentration. Although further studies are being continued, it seems that lactic acid is a promising agent which can be employed as a safe and effective antibacterial agent.

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References

1. Dubos RJ : Effect of ketone bodies and other metabolites on the survival and multiplication of staphylococci and tubercle bacilli. J Exp Med 98 : 145, 1953.
2. Kandhari KC, Pasricha JS, Duggal SK et al : Investigations into the causation of increased susceptibility of diabetics to cutaneous infections. Indian J Med Res 57 : 1295, 1969.
3. Pasricha JS : Personal communication, 1976.