are the most fequently dispensed liquid pharmaceutical preparations in hospitals around Ile-Ife, Nigeria.

The Mixtures were prepared as directed by relevant British Phrmaceutical Codex (BPC, 1975) monographs. Prior to preparation, the various liquid ingreidients were sterilised by autoclaving at 115°C for 30 minutes. In the case of volatile substances these were autoclaved in sealed ampoules to prevent evaporation. Powders were sterilised by heating in a hot air over at 115°C for 1 hour. Following this, the ingredients were aseptically compounded under a laminar flow cabinet (John Bass Limited, Sussex, England). A 100 ml portion of each preparation was dispensed into a glass stoppered bottle and challenged with 1 ml of thebacterial suspension prepared as earlier described. Each preparation therefore contained approximately 1.0×10^3 micro-organisms/ml. The inoculated preparations were then incubated at room temperature (abut 27oC) over a period of 28 days.

SAMPLING

From the incubated preparation, duplicate 1 ml portions were removed at 4-day intervals, serially diluted in Ringers solution and 1 ml of the final dilution was transferred into each of four sterile petridishes to which 20 ml of molten nutrient agar was then added. The plates were throughly mixed, allowed to set and then incubated at 37°C for 48 hours after which the number of colonies in each plate was counted and the viable count calculated from figures obtained. Samples of preparations to which 1 ml of sterile water was added were treated in an identical manner and used as controls. No organisms were detected in any of the control samples throughout the 28-day period of the experiment.

RESULTS AND DISCUSSION

The results are shown in Figures 1-6 from which it can be seen that most of the organisms employed proliferated very rapidly in each of the four preparations. The organisms employed in this study included four Gram-positive cocci. The two Gram-positive rods not only survived but proliferated within each of the four preparations used. A substantial number of these organisms were isolated from each preparation throughout the period of study (Figures 1 and 2). Of the four Gram-negative rods from in the challenge, two died off in all the preparations within 8 days (Figure 4), but the other two survived and proliferated within the preparations (Figure 3 and 5). The two Gram-positive cocci employed were not detectable in the preparations shortly after they were incubated except in the case of the organisms which seemed to have become adapted to conditions in the ipecacuanha and ammonia mixture paediatric BPC (Figure 6). The situation was such that it proliferated within the preparation towards the end of the 28-day study period.

On the whole, the observations are in broad agreement with earlier work in this field. Westwood and Pin-lin (1972) have, for example, reported the survival of various organisms including E coli, Ps, aeruginosa and S. aureus in both Magnesium trisilicate and Potassium citrate mixture oyer period exceeding 7 days. Furthermore, Sykes (1969); as well as Akinmoji and Ogunlana (1972) were able to isolate large numbers of organisms from many different preparations presumably becasue of the ability of these microorganisms to survive and grow within the preparations considered.

It is interesting to note that organisms were able to survive and even multiply in the various preparations even though each of these preparations contained chloroform water which is supposed to act as a preservative. The inability of chloroform water to prevent bacterial growth in these preparation might have been caused by the ability of the various microorganisms to utilise chloroform water as a source of carbon and energy. Examples of this phenomenon have been provided by Bock (1967) who reported the ability of Aspergilus niger to degrade benzoic acid, and also by Burdon and Whitly (1967) who isolated Ps. cepacia from various antibacterials solutions including chlorhexidine and certrimide. Brown (1971) has also demonstrated that peppermint water is virtually a selective medium for Ps. pyocyanea. It is also worth remembering that free living organisms are generally resistant to the activity of anti-bacterial agents becasue of the characteristic structure of their cell envelope (Brown, 1975).

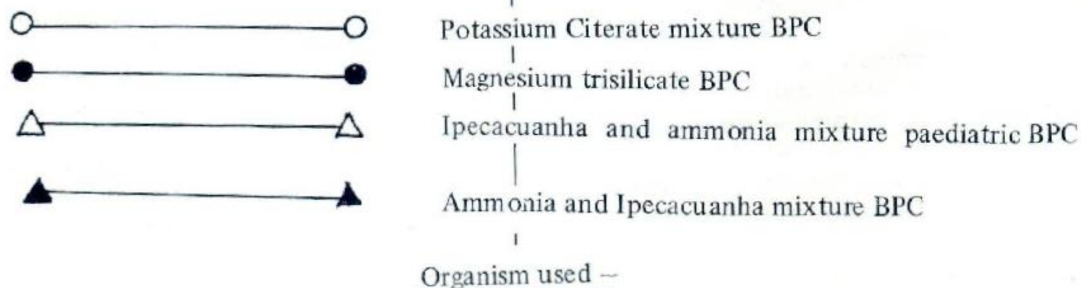
From the results obtained, it can be seen that some of the organisms employed in the test failed to survive when inoculated into the different preparations used. The inability of these organisms to survive may be attributed to the antibacterial activity of the chloroform water which was incorporated into the preparation. On the other hand, it is possible that these organisms failed to survive in the preparations because they were unable to utilise any of the ingredients of these preparations as a source of carbon and energy For such organisms, the inclusion of preservatives in pharmaceutical preparations is unnecessary, the internal environment of pharmaceuitcals being sufficiently hostile to prevent their survival (Westwood and Pancholi, 1974).

The undesirability of having grossly contaminated liquid pharmaceuticals has earlier been discussed (Lamikanra and Onwudike, 1979). The results of this investigation show that free-living bacteria can remain viable in pharmaceutical preparations over long periods. It is therefore important to ensure that all the raw materials used in the preparation of pharmaceuticals are free large bacterial population. Special care is required in hospital pharmacies which is why Baird, Parks and Awad (1977) have suggeted the introduction of an environmental control programme to minimise the possibility of contamination of hospital preparations. In places where such program-

mes cannot be introduced it may be desirable to routinely incorporate adequate concentrations of preservatives into all hospital preparations. Such preservatives must however be rigorously tested so as to ensure that they are effective under the conditions of use.

Figures 1 – 6

The survival patterns of some free living microorganisms in



Organism used –

Fig. 1. Gram positive rod
Fig. 2. Gram positive rod
Fig. 3. Gram negative rod
Fig. 4. Gram negative rod
Fig. 5. Gram negative rod
Fig. 6. Gram positve coccus

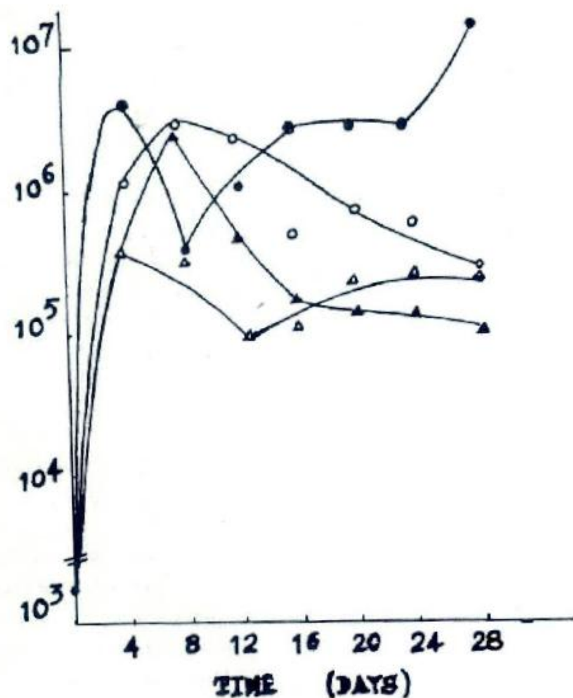


Fig 1

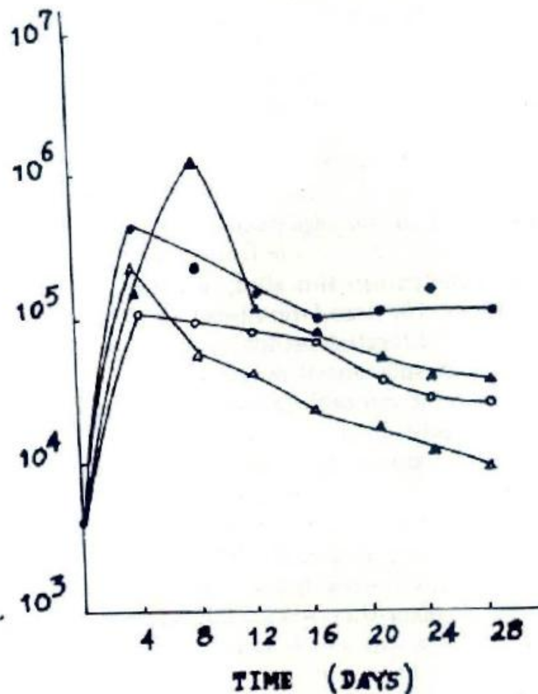


Fig 2

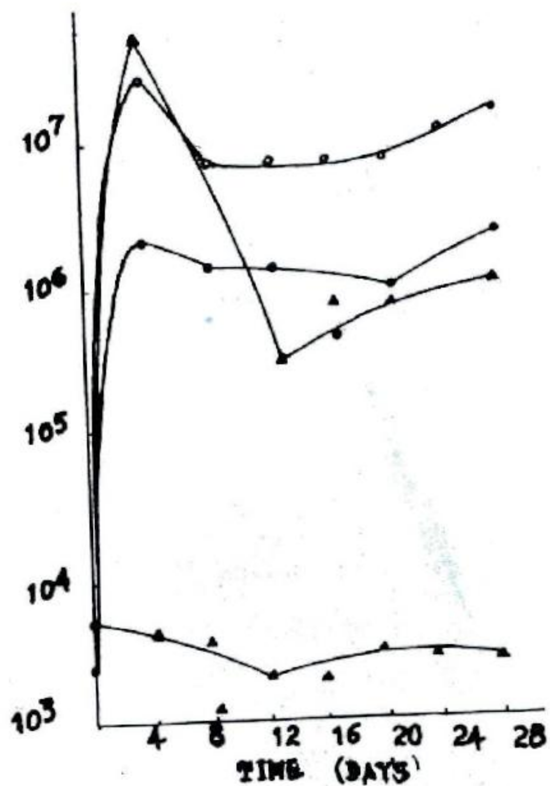


Fig. 3

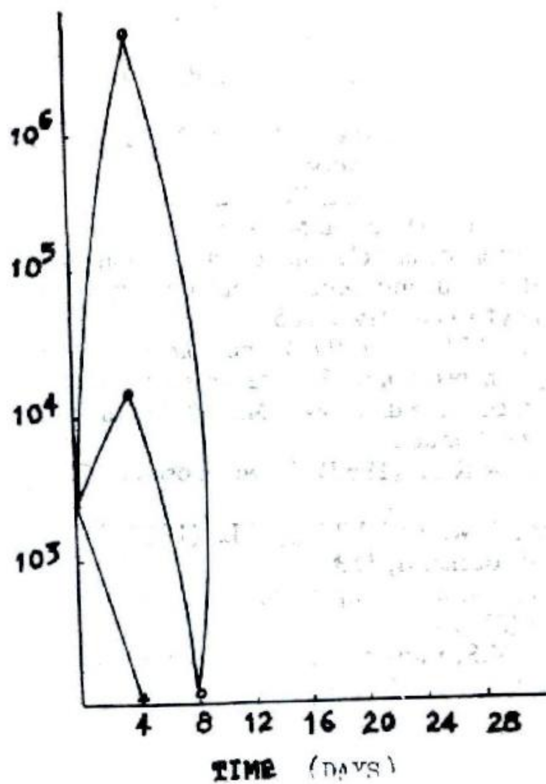


Fig. 4

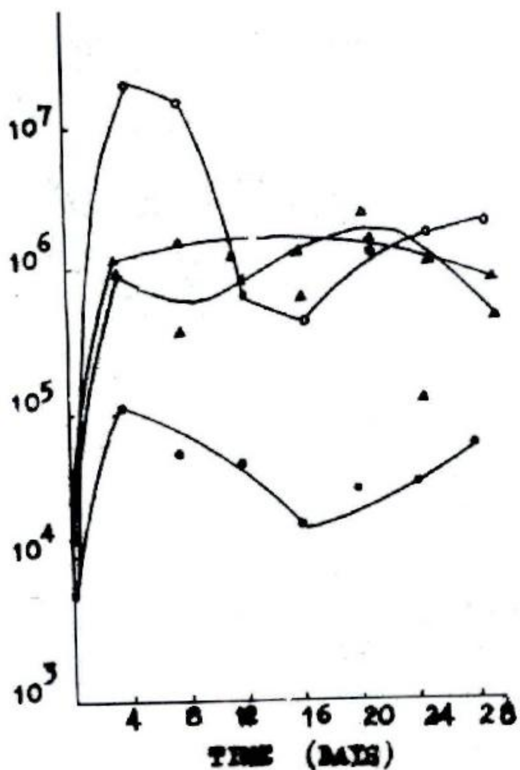


Fig. 5

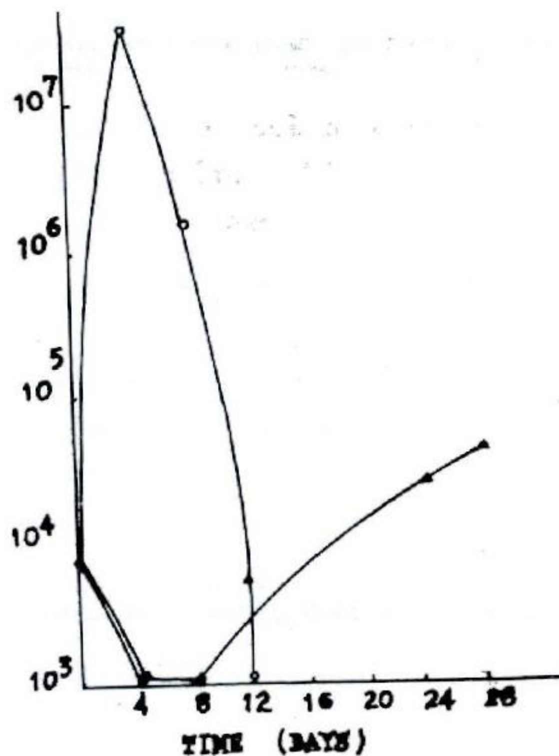


Fig. 6

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