A quick method for testing antituberculosis drugs

Gaugas & Rees (1968) showed that progress of tuberculosis can be very rapid if the immunosuppressive antilymphocytic serum (ALS) is used in thymectomized mice. Here I propose a quick method for screening antituberculosis drugs using the above techniques.

Albino mice about 6 weeks old were thymectomized by sucking out the gland under anaesthesia and used after 14 days. Antilymphatic serum was raised against mouse thymocytes in rabbits (Greaves, Tursi & others, 1969) and assayed by using an agglutination test.

Mice, in groups of 10 were given ALS, 0·2 ml intramuscularly, every alternate day. The infective dose of 1 mg of H37Rv in 0·4 ml was given intravenously, and the drugs were administered on the 4th day. All the drugs were fed orally, except streptomycin which was given intramuscularly. The mortality of the animals was checked till 80% of the animals died.

From preliminary work it was found that more uniform and quicker results were achieved if the mortality of the animals was recorded at 80%, in one week than the customary method of recording 100% mortality. The drugs were administered only once on the 4th day, at which time the growth of the bacilli would be logarithmic. The results (Table 1) are taken to be significant if the survival period was increased from 7 in control to 9 or more in the drug-treated group. Conzelman & Jones (1956) found that the half life of cycloserine in mice was too short to have any effect. This was confirmed.

Table 1. *Time in days to kill* 8 *of ten infected mice.*

Drug	Dose/kg	Days required for 80% mortality
Saline	0·4 ml	7
Isoniazid	20 mg	11
PAS	400 mg	11
Streptomycin	25 mg	12
Cycloserine	100 mg	7
Ethionamide	40 mg	10
Pyrazinamide	20 mg	9
Thiacetazone	40 mg	11

According to Youmans & Youmans (1964) a screening test for *in vivo* bacteristasis should be conducted using animals in which only acute progressive tuberculous disease is manifest, thus eliminating the factor of acquired immunity. So the present method of using immunosuppressed animals would provide an ideal model. The only precaution required is that the animals do not die of other infections.

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Interactions in phenol–sodium dodecyl sulphate–water systems

Changes in environment resulting from micelle formation and solubilization can be followed by observing chemical shifts in the nuclear-magnetic-resonance spectra of the components. We have used nmr spectroscopy to study interactions between phenol, water and the surfactant sodium dodecyl sulphate.

Spectra were obtained in $D_2O:H_2O$ (60:40) at 35° using a Perkin-Elmer R-12 high resolution spectrometer. Chemical shifts were determined by locking to tetramethylsilane as an external standard and expanding the field to 50 or 100 Hz per chart width; the accuracy of the shifts is within about $\pm 0{\cdot}2$ Hz. Corrections for diamagnetic susceptibilities were made where necessary.

A change of medium from a polarizing to a more inert environment (such as the hydrocarbon environment of a micelle of the surfactant) may cause a considerable high-field shift (Eriksson & Gillberg, 1966). Fig. 1a shows the high-field shift for the phenol ring protons with increasing surfactant concentration. As this concentration increases, the ratio of micellar phenol to free phenol increases. Since the observed chemical shift is the weighted average of the free and solubilized peak positions this gives rise to a high-field shift.

Extrapolation of the phenolic proton shifts to zero chemical shift gives an intercept

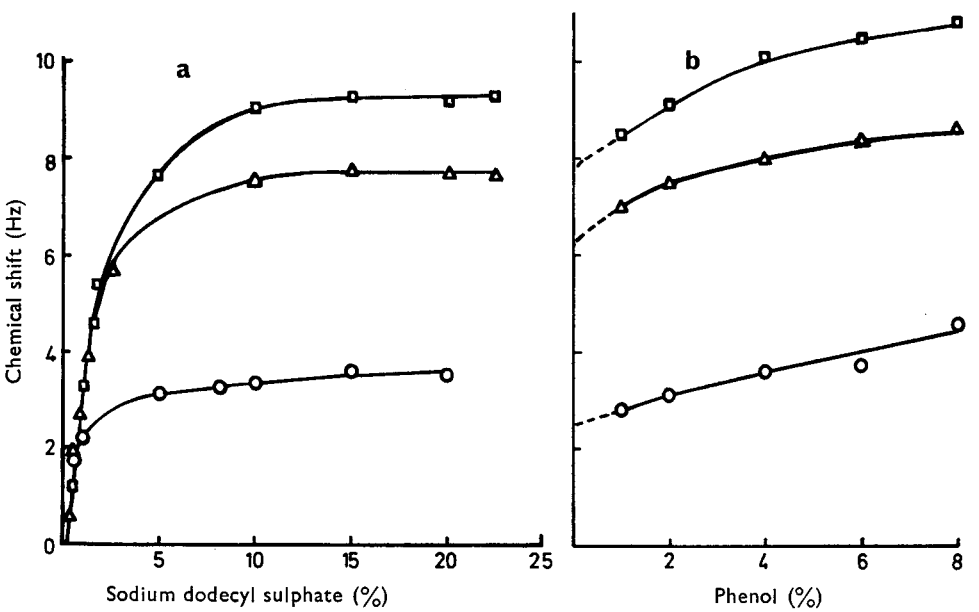

FIG. 1. Chemical shifts of ring protons of phenol: (a) 2% phenol in the presence of varying concentrations of sodium dodecyl sulphate (measured with respect to the peak positions of 2% phenol in water), (b) varying concentrations of phenol in the presence of 10% sodium dodecyl sulphate (measured with respect to the corresponding positions in water). □ *meta* protons; △ *para* protons; ○ *ortho* protons.