Chapter 9

COMPARATIVE SUSCEPTIBILITY OF STRAINS OF TREPONEMES TO PENICILLIN

One of the important practical questions in the field of treponematosis control is whether all species and strains of pathogenic treponemes are equally susceptible to antibiotics, particularly penicillin; whether one scheme of treatment developed for, and laboriously tested in, one of the clinical syndromes of the treponematosis group of diseases, as it occurs in one locality, may reasonably be expected to be equally effective in other localities and against other clinical syndromes.

In addition to this very practical question, there is a more theoretical problem concerned with the relation of one strain of treponeme to another. In other words: Are there significant similarities or differences in the susceptibility of various strains of treponemes to penicillin that may serve as an index to the biological relationships existing within the family of treponemal organisms?

With the foregoing considerations in mind, studies have been made of the comparative susceptibility of most of our newly isolated strains to penicillin G.

Test Procedures

Tests for penicillin sensitivity have been made by both the *in vivo* and the *in vitro* methods described in Chapter 6. The *in vivo* method was developed in this laboratory by Turner, Cumberland & Li.[2] The *in vitro* method follows, in principle, procedures used for other organisms, but was adapted as a practical procedure for treponemes only after painstaking experimentation by Nell,[1] working in this laboratory.

Results of " In Vivo " Tests

A rather wide experience with the Nichols strain has established that a dosage level of 0.25 mg of crystalline penicillin G per kg of body-weight will, in a high proportion of rabbits, reduce the treponeme count from 200 or

more organisms in 200 fields to less than 10 treponemes in 200 fields, within 24 hours after the initiation of treatment. The total dose was given in 3 equal amounts 2 hours apart. Turner, Cumberland & Li [2] also reported that half this dose (0.125 mg) will reduce the count to 10 treponemes or less in approximately one half of the animals. It was further found that a total dose of 0.025 mg/kg body-weight rarely reduced the count to this level when the Nichols strain was the test organism.

In the comparative tests with various strains, carried out in collaboration with our associate, Dr Katherine Schaeffer, dosages of 0.25 mg and 0.05 mg of penicillin G were given. Difficulty was experienced with some strains in obtaining lesions with sufficient treponemes for the *in vivo* tests, so that many animals which were inoculated for this purpose were not used. The results shown in Table LXIII represent only completed tests.

TABLE LXIII. COMPARATIVE SENSITIVITY OF NEWLY ISOLATED STRAINS OF TREPONEMES TO PENICILLIN G AS DETERMINED BY A SHORT "IN VIVO" METHOD

Group	Strain	Results of indicated dose [a]	
		0.25 mg/kg body-weight	0.05 mg/kg body-weight
Syphilis	Nichols	1/12	10/11
,,	Chicago	1/3	2/3
,,	Baghdad A	0/3	2/4
,,	Baghdad B	0/3	3/3
,,	Mexico A	0/3	1/2
Yaws	Haiti A	0/2	
,,	Haiti B	1/4	2/3
,,	Indonesia B	0/2	0/2
,,	Samoa D	0/3	1/3
,,	Samoa F	1/1	
Bejel	Syria A	1/3	2/3
,,	Syria B	0/2	0/1
,,	Iraq B	0/1	1/1
Endemic syphilis	Bosnia A	0/3	2/3
,,	Bosnia B	0/2	0/1

[a] Numerator = number of rabbits in which treponeme count remained higher than 10 in 200 fields; denominator = number of rabbits in which tests were made

It is evident that these data are far from definitive. Taken as a whole, however, they suggest that most of the newly isolated strains have the same degree of penicillin sensitivity as the Nichols strain.

Results of " In Vitro " Tests

The results of *in vitro* tests of penicillin sensitivity on newly isolated strains, recorded in Table LXIV, are those reported by Nell [1] from this laboratory. The sensitivity is expressed in terms of the concentration of penicillin G that will immobilize 50% of the treponemes in the suspension (IC_{50}) during 18 hours' incubation at 35°C. The concentrations of penicillin are expressed in μg/ml.

TABLE LXIV. COMPARATIVE "IN VITRO" SENSITIVITY OF NEWLY ISOLATED STRAINS OF TREPONEMES TO PENICILLIN *

Group	Strain	IC_{50} [a] after 18 hours' incubation at 35°C — Individual assay—μg/ml	Mean
Syphilis	Nichols	0.002, 0.0011, 0.0018, 0.002, 0.002 0.0015, 0.0021, 0.0021, 0.0031, 0.0022, 0.0025	0.002
,,	Chicago	0.0011, 0.0017	0.0014
,,	Baghdad A	0.0012, 0.0083, 0.0021, 0.0049	0.0041
,,	Baghdad B	0.001, 0.0037, 0.0049	0.0032
Yaws	Haiti A	0.0011, 0.0021	0.0016
,,	Haiti B	0.0008, 0.0035, 0.0039	0.0027
Bejel	Syria A	0.002, 0.0016, 0.001, 0.0043	0.0022
,,	Syria B	0.0019, 0.0023	0.0021
,,	Iraq B	0.0032, 0.001, 0.002, 0.0024	0.0021
Endemic syphilis	Bosnia A	0.0010, 0.0022	0.0016
,,	Bosnia B	0.0017, 0.0007	0.0012

* Adapted from Nell [1]

[a] IC_{50} = penicillin concentration at which 50% of treponemes were immobilized

It will be noted from Table LXIV that the effective concentration for all the strains tested is about the same, to judge from the mean figures for several assays. It is doubtful if any of the differences can be regarded as significant and certainly there is no evidence suggesting that any of these strains have an abnormally high resistance to penicillin.

REFERENCES

1. Nell, E. E. (1954) Comparative sensitivity of treponemes of syphilis, yaws and bejel to penicillin in vitro, with observations on factors affecting its treponemicidal action, *Amer. J. Syph.*, **38**, 92
2. Turner, T. B., Cumberland, M. C. & Li, H.-Y. (1947) Comparative effectiveness of penicillins G, F, K, and X in experimental syphilis as determined by a short in vivo method, *Amer. J. Syph.*, **31**, 476