Duration of Cases up to December 31st, 1920, without any Symptoms or Microscopical Findings.

Case 2 ...	...	...	...	27 months
" 4 ...	...	...	...	Av. 24 "
" 7 ...	...	...	...	Av. 18 "
" 8 ...	...	...	...	Av. 15 "
" 9 ...	...	...	...	Av. 15 "
" 10 ...	...	...	...	14 "
Cases 11 to 15 ...	...	...	...	13 "
" 16 to 25 ...	...	...	...	8 "
" 26 to 29 ...	...	...	...	7 "
" 30 to 33 ...	...	...	...	6 "

Cases in which the Cerebro-spinal Fluid was Positive.

Case 7.	Case 39.
" 10 (since dead of sleeping sickness).	" 42 (since dead).
" 30.	" 44.
" 33 (since dead).	" 45.

Deaths and Causes.

Case 1 ...	Sleeping sickness.
" 3 ...	Disappeared; presumably dead.
" 5 ...	? Sleeping sickness (no records).
" 9 ...	Sleeping sickness.
" 10 ...	Sleeping sickness.
" 33 ...	? Delayed CHCl ₃ poisoning; marked mediastinal changes at necropsy.
" 42 ...	Pneumonia.
" 43 ...	Died on admission.

While it may be early to state positively that the present treatment promotes definite cure, we cannot help but feel optimistic in the face of the results we have obtained. Several of our cases have gone well beyond a year with one dose of treatment, and they have all invariably improved in health and no trypanosomes have been found, though they have been subjected to repeated and careful investigations.

Most of our cases have been treated with Burroughs Wellcome and Co.'s neo-kharsivan, a few cases were also treated by atoxyl, the object being to find out which is the more efficient drug. We also mean to use other drugs such as galy and also tartar emetic, and we hope to be able to publish during the next year (1921) comparative results of the various drugs used.

Experiments are also being made to see whether the results we have up to now obtained may not be due to the formation of trypanolysins or other antibodies. That the trypanosomes give rise to specific lysins was suggested by Louis Martin and Henri Darre, who attributed the fever to the massive destruction of the parasites by trypanolysis, and who showed that the blood of patients obtained at the onset of fever had a very marked trypanolytic power both *in vitro* and *in vivo*. We have therefore treated one or two well chosen cases with plain serum—that is, the serum from one of our treated cases was injected into the blood and cerebro-spinal fluid of another sleeping sickness patient; up to now it is much too early to form any opinion.

Conclusions.

1. Of 56 cases treated up to now, 8 are dead. Of 6 of these we have records, and of these 6 only 3 died of sleeping sickness, the others dying of an intercurrent disease, favoured, may be, by the trypanosome infection.

2. All the remaining cases are doing well, some (21) have only been treated during the last six months and so are excluded from this paper, though included in the index of cases.

3. The drugs tried so far have been neo-kharsivan most extensively, and to a less extent atoxyl, but all that has been said in this paper refers mainly to neo-kharsivan.

4. Plain serum treatment is also being tried in a few carefully selected cases. In advanced cases we are trying direct spinal medication; so far the results are uncertain—one case treated thus died; another, under observation, in whose spinal cord 0.003 gram neo-kharsivan was introduced, seems to present a certain amount of improvement.

Finally, we wish to express our thanks to Dr. C. A. Wiggins, Principal Medical Officer of this Protectorate, for his assistance and encouragement in carrying out these investigations and for his permission to publish this report.

REFERENCES.

- ¹ M. Nierenstein, BRITISH MEDICAL JOURNAL, June 5th, 1920. ² R. van Sackeghem, *Bull. Soc. Path. Exotique*, May, vol. 8, No. 5, pp. 339-348; and also with E. Nicolas in the same *Bulletin*, 1916, vol. 9, No. 10, pp. 815-823. ³ Hans Ritz, *Archiv f. Schiffs u. Trop. Hyg.*, 1916, September 20th, No. 17, pp. 397-420; also A. Stuehmer, *Zeit. f. Immunitätsforsch.*, I Teil, Orig., 1916, March 4th, vol. 24, No. 4, pp. 315-335.

THE VIRULENCE OF DIPHTHERIA-LIKE ORGANISMS.

A NOTE ON THE INTRACUTANEOUS TEST.*

BY

A. J. EAGLETON, M.D. LOND.,

AND

E. M. BAXTER, M.Sc. SHEFFIELD.

For testing the virulence of diphtheria-like organisms, Zingher and Soletsky¹ described a method of intracutaneous injection which was a modification of that originally described by Neisser. They advocated the method on the grounds of economy, and stated that in 60 cases it gave the same result as the subcutaneous method. These authors usually tested four strains on each pair of guinea-pigs.

We have been using a similar test for some months, and are submitting our experience of it in the hope that it may prove useful to the many public health pathologists in England who are working in the same field. The modifications we have introduced are as follows:

1. Standardization of inoculum by opacity.
2. Inclusion of a known virulent strain in each test.
3. Injection of control animal on the preceding day.
4. Administration to the unprotected animal of a "following dose" of antitoxin of such a size that the skin reactions are not obscured while the life of the guinea-pig is saved.

The technique of the test is as follows:

Two guinea-pigs are used for each test; both must be partially white.

Protecting Dose.—One animal is given 500 units of diphtheria antitoxin by intraperitoneal injection on the day preceding the test. This animal acts as a control.

Preparation of Inoculum.—The test strains and a known virulent diphtheria bacillus are grown eighteen hours on Loeffler slopes, emulsified in saline, diluted and standardized by opacity so that the suspension contains approximately fifty million organisms per cubic centimetre.

Injection of Suspensions.—White-haired portions of the guinea-pigs are depilated with calcium sulphide paste, and into corresponding skin areas of the two animals is injected 0.2 c.cm. of each suspension. The injections are strictly intradermal and at least half an inch apart. On each pair of animals we usually test eight or ten strains, including the control virulent.

Following Dose.—Four or five hours later the animal which was not given a protecting dose on the previous day receives 125 units of diphtheria antitoxin intraperitoneally.

Readings are taken for the next three days.

In the case of a virulent diphtheria bacillus the control animal shows nothing or a faint transient flush; the other animal shows a definite rose-red swelling which becomes more marked at each successive reading, and may terminate in slight necrosis. An avirulent diphtheria-like organism gives a negative result with both animals. If the test culture is contaminated with streptococci the result may be obscured by a reaction in both animals, but this does not always happen.

The advantages of the test are:

1. Economy of animals, as both survive.
2. Owing to the variability of growth shown by different strains, standardization by opacity is preferable to the use of a certain proportion of a slope culture (as in the American method).
3. Reliability. We have had the opportunity of comparing only a limited number of strains by the subcutaneous injection method; so far the agreement is satisfactory.

REFERENCE.

- ¹ Zingher and Soletsky: *Journal of Infectious Diseases*, vol. xvii, No. 3, 1915.

* From the Wellcome Physiological Research Laboratories.

MUCH is hoped in Germany from the regulation, made at the beginning of 1921, prohibiting the exploitation of Wassermann's test by persons unrecognized by the health authorities as competent to carry it out. A further precaution to secure a reliable Wassermann test is the decree that only those extracts must be used which have been tested at the Institute for Experimental Therapy, conducted by Professor Kolle at Frankfurt a. Maine.