

WEILS DISEASE **(LEPTOSPIROSIS)**

A CONTAGIOUS DISEASE OF ANIMALS, OCCASIONALLY COMMUNICABLE TO MAN, CAUSED BY A PATHOGENIC SPIROCHETE OF THE GENUS LEPTOSPIRA.

THE RESERVOIR OF LEPTOSPIRES INCLUDES RODENTS AND CERTAIN DOMESTIC ANIMALS, THESE ANIMALS EXCRETE LIVE FULLY VIRULENT ORGANISMS IN THEIR URINE AND CONTAMINATE THE ENVIRONMENT OUTSIDE THE ANIMALS BODY.

LEPTOSPIRES CAN LIVE FOR SEVERAL WEEKS IN FRESH WATER. THUS INFECTION TAKES PLACE BY DIRECT CONTACT WITH URINE OF INFECTED ANIMALS OR BY INDIRECT CONTACT WITH CONTAMINATED FOOD OR WATER.

LEPTOSPIRES CAN READILY PENETRATE MUCOUS MEMBRANES BUT PROBABLY CANNOT GAIN ENTRANCE TO THE BODY THROUGH INTACT SKIN.

A SCRATCH OR ABRASION, AS WELL AS THE NASAL MUCOSA AND EYES, ARE EXCELLENT PORTALS OF ENTRY.

THE ORIGIN OF MANY INFECTIONS CAN BE TRACED TO WADING, SWIMMING OR OTHER CONTACT WITH WATER CONTAINING VIRULENT LEPTOSPIRES.

THE INCIDENT IN MAN DEPENDS UPON THE OPPORTUNITY FOR EXPOSURE I.E. PEOPLE WHO WORK IN SEWERS, OR UNDERGROUND CABLES AND MANHOLES, SWIMMING, HARVESTING AND CONTACT WITH ANIMALS

CLINICAL EVIDENCE OF THE DISEASE IN MAN VARIES DEPENDING UPON THE INFECTING TYPE OF LEPTOSPIRA. USUALLY AFTER AN INCUBATION PERIOD OF ABOUT A WEEK, FEVER, WEAKNESS AND PAINS IN THE LEGS, BACK AND ABDOMINAL MUSCLES ARE NOTED.

NAUSEA, VOMITING AND DIARRHOEA ARE NOT UNCOMMON.

ONE CHARACTERISTIC SYMPTOM IS CONGESTION OF THE CONJUNCTIVAL BLOOD VESSELS AROUND THE CORNEAS OF THE EYES. JAUNDICE MAY OCCUR AFTER THE FIRST WEEK OF ILLNESS.

THE DEATH RATE IS APPROXIMATELY 30% OF THE SEVERELY ILL AND JAUNDICED PATIENT.

PROTECT WORKING AREAS AGAINST WEILS DISEASE (LEPTOSPIRA ICTERHAEMORRHAGIAE) BY SPRAYING WITH VIRID-DC.

VIRID-DC IS EFFECTIVE IN DESTROYING AND CONTROLLING WEILS DISEASE.

VIRID-DC AT A CONCENTRATION OF 1:50 15 ANTI-LEPTOSPRIAL AFTER 5 MINUTES EXPOSURE EVEN WHEN LARGE NUMBERS OF LEPTOSPIRES ARE PRESENT.

Public Health Laboratory Service

Public Health Laboratory
County Hospital
Hereford HR1 2ER
Fax: 0432-351396
Telephone: 0432-277117

Leptospira Reference Laboratory
Telephone: 0432-277707

ASSESSMENT OF ANTI-LEPTOSPIRAL ACTIVITY OF VIRID-DC

Method

Preparation of inoculum

7 day culture of Leptospira interrogans serovar ICTEROHAEMORRHAGIAE icterohaemorrhagiae Ictero I. grown in Johnson and Harris' modification of Ellinghausen and McCullough medium at 30°C.

Four fold dilutions of leptospira culture, initial concentration 10^8 leptospores/ml. were made in PBS. Three master dilutions were used in this study. 10^8 , 10^4 and 10^2 leptospores/ml.

Preparation of detergent

Dilutions of detergent were prepared in distilled water at the following concentrations:

Neat, 1:50, 1:100 and 1:200

Assessment of anti-leptospiral activity

Culture

100ul volumes of detergent and inoculum were mixed in a sterile tube and 10 ul volumes of the mixture removed at the following time intervals. 0, 5, 10 and 20 minutes. The reactant mixtures were then inoculated into 4 ml volumes of EMJH containing 2% rabbit serum and 0.1% agar and incubated at 30°C for 6 weeks. The cultures were examined at weekly intervals for evidence of leptospiral growth. A control for each concentration of inoculum was included comprising of leptospiral culture without disinfectant.

Mortality

An initial assessment of antileptospiral activity was also undertaken by placing 5 ul volumes of the reactant mixture into a Thomas counting chamber at the same time intervals as above and observing for loss of motility. The percentage loss of motility was determined by counting 100 leptospores and estimating the number of non-motile organisms under dark ground illumination at a final magnification of x 125.

These investigations were only carried out on reactant mixtures containing an initial leptospiral concentration of 10^3 organisms/ml because at leptospiral concentrations of 10^4 and 10^2 organisms/ml, there were too few leptospire visible to give an accurate estimate

RESULTS

Time (mins)	<u>Percentage motility</u>			
	<u>Initial concentration of disinfectant</u>			
	Neat	1:50	1:100	1:200
0	5	80	100	100
5	5	10	10	8
10	1	5	7	7
20	1	1	1	1

Concentration of leptospire = 10^8 organisms/ml. Above figures represent percentage of organisms still motile.

Culture results for initial inoculum of 10^8 leptospire/ml

Initial concentration of disinfectant

Time	Neat	1:50	1:100	1:200
0	NG	G	G	G
5	NG	NG	NG	G
10	NG	NG	NG	NG
20	NG	NG	NG	NG

G = growth of leptospire
 NG = No growth

Culture results for initial inoculum of 10^4 leptospire/ml

Time	Neat	<u>Initial concentration of disinfectant</u>		
		1:50	1:100	1:200
0	NG	NG	NG	NG
5	NG	NG	NG	NG
10	NG	NG	NG	NG
20	NG	NG	NG	NG

Culture results for initial inoculum of 10^2 leptospire/ml
Initial concentration of disinfectant

Time	Neat	1:50	1:100	1:200
0	NG	NG	NG	NG
5	NG	NG	NG	NG
10	NG	NG	NG	NG
20	NG	NG	NG	NG

The three control cultures of 10^8 , 10^4 and 10^2 leptospire/ml without disinfectant all grew.

Conclusions

Virid-DC was totally anti-leptospiral when used neat against all dilutions of L.interrogans serovar Icterohaemorrhagiae. At concentration of 1:50 and 1:100 it was anti leptospiral after 5 minutes exposure even when large numbers of leptospire (10^8) were present.

Dr IR Ferguson
Director



Laboratories at Bristol, Exeter, Gloucester,
Hereford, Plymouth, Taunton, Truro

PUBLIC HEALTH LABORATORY SERVICE

Public Health Laboratory
County Hospital, Hereford HR1 2ER
Tel 01432-277117 Fax 01432-351396
Leptospira Reference Unit
Tel 01432-277707

CPA Accredited
a UKAS TESTING Laboratory No 1613

7.4.99

Ms Lynne Ratcliffe,
Technical Support Manager,
NCH (UK) Ltd,
Lanchard House,
Victoria Street,
West Bromwich, B70 8ER

Dear Ms Ratcliffe,

Laboratory number: L98/2840

The Healthy Hands hand cleanser was tested for leptospiral activity by the method described in the protocol of 31.01.98.

The product was used as supplied.

Results for the product after six weeks incubation:

Tested against *Leptospira interrogans* serovar *Icterohaemorrhagiae* RGA.

Minutes	Inoculum		
	10^8	10^4	10^2 orgs/ml
0	NLG	NLG	NLG
5	NLG	NLG	NLG
10	NLG	NLG	NLG
20	NLG	NLG	NLG
Control : RGA	LG	LG	LG

NLG : No leptospiral growth

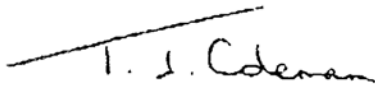
LG: Leptospiral growth

Comment

Results suggest that Healthy Hands hand cleanser, will kill leptospire on exposure.

Should you wish to discuss these results further, please contact me.

Yours sincerely,

A handwritten signature in black ink that reads "T. J. Coleman". The signature is written in a cursive style and is positioned above a horizontal line that extends to the left.

Dr T J Coleman
Head of Unit

