

A new serovar *mogdeni* of serogroup Tarassovi of *Leptospira interrogans* isolated from a sewage plant in England

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SUMMARY

Among 30 strains of leptospires isolated from samples of sewage taken before and during treatment at two sewage plants in England, only one appeared to belong to *Leptospira interrogans*, the species that comprises the leptospires that are pathogenic to man and animals. That strain, Compton 746, was isolated from settled sewage, before treatment at a treatment plant that deals mainly with human sewage. It was shown serologically to belong to serogroup Tarassovi and appears to represent a new serovar that has been named *mogdeni* after the name of the sewage plant, Mogden, from which it was isolated.

INTRODUCTION

In 1977 an investigation into the possibility that leptospires may be harboured in sewage was carried out jointly by the Institute for Research on Animal Diseases of the Agricultural Research Council at Compton, Berkshire, England and the *Leptospira* Reference Laboratory of the Public Health Laboratory Service, Colindale, London. The project was financed by the Thames Water Authority. Results dealing mainly with the saprophytic leptospires isolated during the investigation were reported by Cinco, Coghlan & Matthews (1980), while in a second report on the occurrence and significance to animal health of various pathogenic bacterial species including *Leptospira interrogans* in sewage and sewage sludges, mention is made of the single strain of pathogenic leptospire isolated from those sources (Jones *et al.* 1981). This paper deals with the identification of that single pathogenic strain referred to as Compton 746 that proved to be a new serovar within the serogroup Tarassovi.

MATERIALS AND METHODS

Sources investigated

Two different sewage plants were chosen, one of which, Plant D receives waste from a cattle market and local farms while the other, Plant F, a large urban treatment plant, processes material that is mainly of human origin, with only a

Table 1. *Agglutination of Compton 746 by antisera to serovars within the Tarassovi and Bataviae serogroups**

Antiserum to serovar	Strain	Titre to Compton 746
Tarassovi serogroup		
<i>tarassovi</i>	Mitis Johnson	400
<i>kisuba</i>	Kisuba	<100
<i>bravo</i>	Bravo	<100
<i>chagres</i>	1913 K	<100
<i>gatuni</i>	1473 K	<100
<i>tunis</i>	P2/05	<100
<i>vughia</i>	LT 80-68	<100
<i>bakeri</i>	LT 79	200
<i>guidae</i>	RP 29	200
<i>atchafalaya</i>	LSU 1013	<100
<i>kaup</i>	LT 64-68	<100
<i>kanana</i>	Kwale	<100
<i>darien</i>	637 K	<100
<i>langati</i>	M 39090	<100
<i>atlantae</i>	LT 81	<100
<i>sulzeræ†</i>	LT 82	<100
<i>rama</i>	316	<100
<i>navet</i>	TVRL 1098 37	1600
Bataviae serogroup		
<i>bataviae</i>	van Tienen	<100
<i>paidjan</i>	Paidjan	<100
<i>djatzi</i>	HS-26	<100
<i>claytoni</i>	LT 818	100
<i>losbanos†</i>	LT 101-69	3200
<i>kobbe</i>	CZ-320	<100
<i>argentiniensis</i>	Peludo	<100
<i>balbao</i>	LT 761	200
<i>brasiliensis</i>	AN 776	100
<i>santarosa†</i>	LT 21-74	100

* Homologous titre, 12800. † Not yet officially recognized.

small amount of animal waste material from industries processing animal products. Samples of 20 ml volumes were taken at different stages of the treatment. Details of the method of isolation, culture, purification and preliminary identification of leptospiral isolates have already been given in two previous reports (Cinco, Coghlan & Matthews, 1980; Jones *et al.* 1981). The medium used for culture was Ellinghausen & McCullough's semisolid medium (1965) with the addition of 1% rabbit serum, 100 µg per ml 5-fluorouracil (Johnson & Rogers, 1964) and 50 µg per ml of amphotericin B (Jones *et al.* 1981).

Classification

The serological properties of strain Compton 746 were studied by the microscopic agglutination test (MAT) and by agglutinin-absorption tests (Dikken & Kmety, 1978) in three *Leptospira* Reference Laboratories in London, Bratislava

Table 2. Agglutination titres of antiserum to strain Compton 746 against serovars within the Tarassovi and Bataviae serogroups

Serovar	Strain	Titre
Tarassovi serogroup		
<i>tarassovi</i>	Mitis Johnson	100
<i>kisuba</i>	Kisuba	< 100
<i>bravo</i>	Bravo	< 100
<i>chagres</i>	LT 924	< 100
<i>gatuni</i>	LT 839	< 100
<i>tunis</i>	P 2/65	< 100
<i>vughia</i>	LT 89-68	< 100
<i>bakeri</i>	LT 79	200
<i>guidae</i>	RP 29	100
<i>atchafalaya</i>	LSU 1013	100
<i>kaup</i>	LT 64-68	400
<i>kanana</i>	Kwale	NT
<i>darien</i>	637 K	< 100
<i>langati</i>	LT 59-67	200
<i>allantae</i>	LT 81	200
<i>sulzeræ*</i>	LT 82	100
<i>rama</i>	316	400
<i>navet</i>	TVRL 1098 37	12800
<i>tarassovi</i> dog strain	LC 25	200
	Compton 746	50 000
Bataviae serogroup		
<i>bataviae</i>	van Tienen	< 100
<i>paidjan</i>	Paidjan	< 100
<i>djatzi</i>	HS-26	< 100
<i>claytoni</i>	LT 818	100
<i>losbanos*</i>	LT 101-69	< 100
<i>kobbe</i>	CZ-320	< 100
<i>argentiniensis</i>	Peludo	400
<i>balbao</i>	LT 761	800
<i>brasiliensis</i>	AN 776	800
<i>santarosa*</i>	LT 21-74	< 100

* Not yet officially recognized. NT, Not tested.

and Amsterdam. In addition to reference strains and their antisera that appear on the WHO list (1982), a number of serovars within the Tarassovi, Bataviae and Shermani serogroups not yet officially recognized were included in the tests carried out in Bratislava, since they will be dealt with in a future report on subgrouping (Kmety, to be published). The results of the tests in the three reference laboratories were in agreement and were confirmed in Bratislava by antigenic factor analysis (Dikken & Kmety, 1978). The results of the full range of tests carried out in Bratislava are shown in Tables 1, 2 and 3.

RESULTS

Strain Compton 746 was isolated from the settled sludge of sewage plant F before treatment. It failed to grow in the presence of 8-azaguanine and did not

Table 3. *Agglutination titres of antisera to strain TVRL 109837 of serovar navet and strain Compton 746 before and after cross agglutinin-absorption*

Antiserum	Titres against	
	Compton 746	TVRL 109837
TVRL 109837		
unabsorbed	1600	12800
absorbed with Compton 746	100	12800
Compton 748		
unabsorbed	12800	12800
absorbed with TVRL 109837	12800	200

grow at 13 °C, indicating that it belongs to the pathogenic species *Leptospira interrogans*.

The results of MAT of Compton 746 against antisera to strains representing 19 serogroups of *L. interrogans* showed that the isolate is antigenically related to the Tarassovi serogroup and to a lesser extent to the Bataviae serogroup. However, when cross-agglutination tests were carried out against all known serovars within those serogroups, it was found to react with only a few of them, the closest relationship being with serovar *navet* of the Tarassovi serogroup (Tables 1 and 2).

A comparison of Compton 746 and *navet* by agglutinin-absorption tests showed that the two differ sufficiently to indicate that they belong to different serovars (Table 3). According to the results Compton 746 can be considered as the reference strain of a new serovar for which the name *mogdeni* is suggested.

Because of the low-level reactions of antiserum to Compton 746 when tested against other members of the Tarassovi serogroup except *navet*, and vice versa, it is suggested that the two serovars *mogdeni* and *navet* be placed in a separate subgroup within the Tarassovi serogroup until such time as similarly related serovars are found which would justify the provision of a separate serogroup.

Factor analysis revealed that the two serovars *navet* and *mogdeni* have a common major antigen designated Ta 37 that differentiates them from all the other serovars within the group, while each has a separate major antigen, Ta 38 and Ta 39 respectively. Ta 39 is suggested as the factor serum for typing and differentiating these two serovars.

DISCUSSION

The isolation of strain Compton 746 from the sewage was surprising, as Tarassovi serogroup infections have not been diagnosed in man in Great Britain and there has been only one previous isolate, strain LC 25, identified as serovar *tarassovi* obtained from one of a pack of hounds (Spackman, Little & Salt, unpublished data). There has been no firm serological evidence of Tarassovi serogroup infections in domestic animals including sheep (Hathaway, Little & Stevens, 1980), pigs (Hathaway & Little, 1981), cattle (Little, Richards & Hussaini, 1981) and only slight serological evidence in horses (Hathaway *et al.* 1981). Sixteen of 20 sera taken from a pack of healthy beagles, aged 6–12 months

at the Animal Health Trust's Small Animal Centre, Newmarket, were weakly positive (titre 30) against the Tarassovi serogroup antigen and there was a significant rise in titre shown by some of the animals. Horses in a nearby pasture and the local vole population were serologically negative (Macdougall, personal communication).

At present it is not possible to speculate on how the strain came to be in the sewage, nor what was the likely animal source. Previous surveys of domestic animals may have failed to identify antibodies specific to the new serovar through insufficient representation within the Tarassovi antigen pool. It is recommended that in any future investigations serovar *mogdeni* should be included in the pool. Cultures of the strain are maintained in the *Leptospira* Reference Unit at County Hospital, Hereford, England; at the Instituut voor Tropische Hygiene in Amsterdam and at the Institute of Epidemiology in Bratislava, Czechoslovakia.

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