

***Listeria monocytogenes* isolation from a chinchilla (*Chinchilla laniger*) ***

Bir çinçiladan (*Chinchilla laniger*) *L. monocytogenes* izolasyonu

Lale ATASEVEN¹, Hakan YARDIMCI², Tuba İÇA²

¹Provincial Control Laboratory, Ankara, TURKEY;

²Department of Microbiology, Faculty of Veterinary Medicine, Ankara University, Turkey

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Özet

İshal, çarpınma ve ölüm hikayeli bir çinçiladan *Listeria monocytogenes* izole edildi. İzole edilen suş, morfolojik, biyokimyasal özellikleri ve *L.monocytogenes* O-spesifik antiserumları (RSHM) ile meydana getirdiği aglütinasyon yönünden değerlendirilerek tanıya edildi. Doğrulama testi APIListeria (bioMérieux) ile yapıldı. İzole edilen suş, *in vitro* olarak penisilin, tetrasiklin, florfenikol ve gentamisine duyarlı bulundu.

Anahtar Kelimeler: Antimikrobiyal duyarlılık, *L.monocytogenes*, çinçila, Türkiye.

Abstract

Listeria monocytogenes was isolated from a chinchilla with the anamnesis of diarrhoea, convulsions and death. The bacteria was identified by the morphological, biochemical characteristics and determination of agglutination with the O specific antisera of *L. monocytogenes* (RSHM). Confirmation was done with APIListeria (bioMérieux). The strain isolated was found sensitive to penicillin, tetracycline, florfenicol and gentamycine.

Keywords: Antimicrobial susceptibility, chinchilla, *L.monocytogenes*, Turkey.

Introduction

Listeria monocytogenes is a Gram-positive pathogen that can cause listeriosis in a wide range of domestic (sheep, goats, cattle, poultry, birds etc.), wild animals and man (6,12,20,19). *L.monocytogenes* produces septicaemic and neural listeriosis in animals (19). Several authors have also reported listeriosis in chinchillas (6,8,9,24). In chinchillas, the clinical signs are generally sudden death, anorexia, listlessness, diarrhoea, weight loss and ataxia (14).

The isolates of *L.monocytogenes* obtained from different sources including food and clinical cases, are susceptible to a wide range of antimicrobials, although there are resistant isolates to some antimicrobials (3,7). The unrestrained use of antimicrobials in animals and human can cause undesired consequences such as the dissemination of resistance among bacterial populations, thus the antimicrobial susceptibility testing is needful for treatment and control of the bacterial infections (7,21).

In Turkey, there were some studies about *L.monocytogenes* in domestic animals (1,2,10,11,15), food (13,18,22), but as we conducted the study in 2001 we had not seen a report on isolation of *L.monocytogenes* in chinchillas in Turkey. The current study aimed to determine the infectious agent from three chinchillas with the anamnesis of neural signs and sudden death and its antimicrobial susceptibility.

Materials and Method

Material

Bacterial Isolations: In this study, liver, heart, lung and brain of three chinchillas with the anamnesis of diarrhoea, convulsions and death brought to the Department of Microbiology, Faculty of Veterinary Medicine, Ankara University in 2001 were examined.

*The case was reported as a poster (Poster 25) in FEMS Symposium "The Versatility of Listeria Species", 10-11 October 2002, Izmir, TURKEY.

Yazışma adresi/Correspondance: Lale ATASEVER, ¹Provincial Control Laboratory, Hatay, TURKEY E-mail: laleata@hotmail.com

Susceptibility Tests: For the susceptibility test, isolates were suspended in TSB and the suspension was adjusted to a turbidity equivalent to a 0.5 McFarland standard. The antibiotic susceptibility test was performed with the agar disk diffusion method (4) Isolates were categorized as susceptible, moderately susceptible, and resistant, based upon interpretive criteria developed by the Clinical and Laboratory Standards Institute (16). Penicillin (10 IU), tetracycline (30 IU), gentamicin (10 IU), streptomycin (10 IU), neomycin (10mcg), florfenicol, trimethoprim-sulphamethoxazole (25mcg), amoxicillin (25mcg), amoxicillin-clavulanic acid (30mcg) discs were used to determine the antimicrobial susceptibility of the strain isolated from a chinchilla.

Method

The samples were cultured on blood and McConkey agar and incubated at 37°C for 24 hours. Small pieces of brain with spinal cord and medulla, and the organ samples were homogenized in 10 per cent of nutrient broth suspension. The broth suspension was placed in the refrigerator at 4°C (cold enrichment) and subcultured onto agar plates twice weekly for 3 weeks. The differentiation of *L. monocytogenes* was carried out according to Bisping and Amsberg (5) and Quinn et al (19).

Anton test in two rabbits and intraperitoneal inoculation of two mice with the isolated *L. monocytogenes* were carried out for pathogenicity testing. For agglutination with the O specific antisera of *L. monocytogenes*, the standard O specific antisera were obtained from Refik Saydam Central Institute of Hygiene (RSHM) and confirmation of biochemical characters was done with APIListeria (bioMerieux).

Penicillin (10 IU), tetracycline (30 IU), gentamicin (10 IU), streptomycin (10 IU), neomycin (10mcg), florfenicol, trimethoprim-sulphamethoxazole (25mcg), amoxicillin (25mcg), amoxicillin-clavulanic acid (30mcg) discs were used to determine the antimicrobial susceptibility of the strain isolated from a chinchilla.

Results

Bacterial isolation

L. monocytogenes was isolated from brain and visceral organ samples of only one chinchilla with convulsions and a sudden death, but as other chinchillas were not suitable for microbiological isolation. The bacteria with narrow zones of beta haemolysis on blood agar were Gram-stained and, Gram positive rods and coccobacilli were seen. The identification was then carried out by the characteristics given below (Table 1). The strain isolated was CAMP positive with *Staphylococcus aureus* but not with *Rhodococcus equi* (Figure 1).

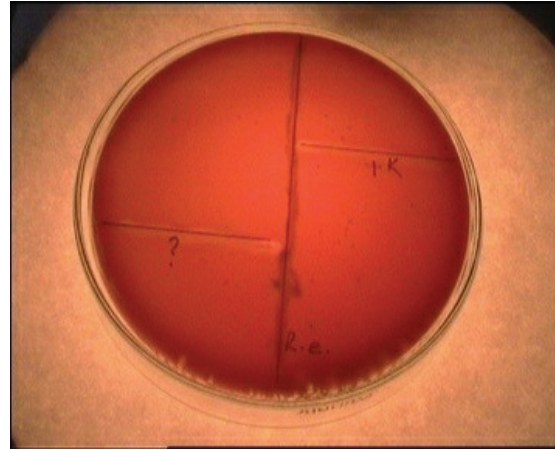


Figure 1. CAMP (*R. equi*)

Şekil 1. CAMP (*R. equi*)

Antibiotic Susceptibility Test

The strain isolated was found sensitive to penicillin, tetracycline, florfenicol and gentamycin and resistant to streptomycin, neomycin, trimethoprim-sulphamethoxazole, amoxicillin and amoxicillin-clavulanic acid.

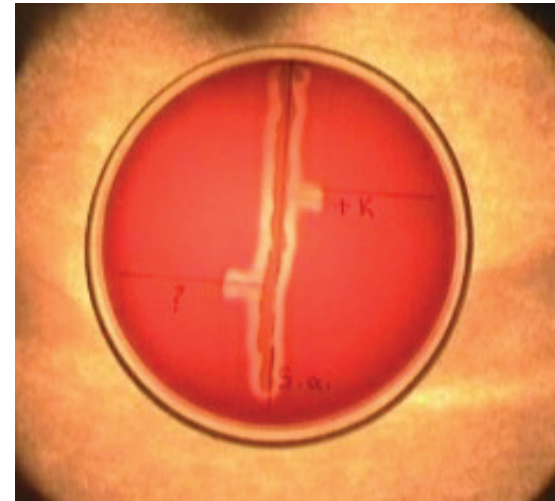


Figure 2. CAMP (*S. aureus*)

Şekil 2. CAMP (*S. aureus*)

Discussion

L. monocytogenes is an ubiquitous facultative pathogen bacterium (12,17). The bacteria can live in soil, forage and water, once enters to the host, it can cause the clinical case expressed as visceral (septicaemic) listeriosis and neural listeriosis (12,17,19). There has been a strong agreement among the researchers, in that some of them reported that chinchillas are one of the highly susceptible animals to visceral listeriosis (6,14,23,24). *L. monocytogenes* was also isolated from chinchillas' various tissues including brain (24) and, acute diarrhoea and central nervous symptoms following a rapid change of feed were observed in 72 of 78 chinchillas with 50% mortality rate (8). In current

Table 1. Biochemical properties of the strain isolated from a chinchilla.

Characteristic		Characteristic	
Motility at 22°C	+	Aesculin	+
Growth at 37°C	+	Arabinose	-
CAMP test (<i>S.aureus</i>)*	-	d-xylose	+
CAMP test (<i>R. equi</i>)**	-	Lactose	-
β-haemolysis	+	l-Rhamnose	+
Catalase	+	Maltose	+
Oxidase	-	Mannitol	-
Reduction of nitrate	-	O/F (glucose)	+
Urease	-	Pathogenicity for mice	+
H ₂ S	-	Anton test	+

study, *L. monocytogenes* was isolated from brain and visceral organ samples of only one chinchilla with convulsions and a sudden death, but as other chinchillas were not suitable microbiological isolation. The bacteria with narrow zones of beta haemolysis on blood agar were Gram-stained and, Gram positive rods and coccobacilli were seen. The identification was then carried out by the characteristics given below (Table 1). Furthermore, clinical signs of chinchillas in this study are in agreement with previous studies (14,24).

In the present study, the isolated strain was found sensitive to penicillin, tetracycline, florfenicol and gentamycin. It was resistant to streptomycin, neomycin, trimethoprim-sulphamethoxazole, amoxicillin and amoxicillin-clavulanic acid. Gheene et al (1969) reported that treatment of chinchillas with tetracycline in drinking water prevented further deaths. Pandurov and Kokosharov (1982) examined the antimicrobial activity on 14 strains of *L. monocytogenes* (20). These strains showed good sensitivity to action of erythromycin, penicillin and kanamycin and, weaker sensitivity to chloramphenicol, gentamicin, tetracycline and furazolidon, but resistant to streptomycin and polymyxin. Ampicillin, rifampicin, or penicillin plus gentamicin have currently preferred for treatment of listeriosis (7). *L. monocytogenes* is an important infectious agent for animal and human health. Moreover, a few reports are available on listeriosis in chinchillas worldwide (6,8,9,24). This study provides a new isolation data of *L. monocytogenes* from chinchillas in Turkey.

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