

A Case Report: Isolation of *Alysiella filiformis* from Pig's Lungs ^[1]

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Summary

Alysiella filiformis is considered a common resident in the oral cavities of many animals. All reports of *Alysiella* indicate that it is restricted to the oral cavity of warm-blooded vertebrates, where it apparently is nonpathogenic. However, increased losses of young pigs occurred in one farm in Serbia. Spumous content in bronchia and partly clotted blood in blood vessels of the lungs were present. Characteristic signs of oedema disease were present and *E. coli* serogroup 0139 was isolated. Furthermore, *Alysiella filiformis* was the single agent isolated from the lungs of diseased pigs. This is the first isolation of *Alysiella filiformis* from pig lungs.

Keywords: *Alysiella filiformis*, Pig, Lungs

Olgu Sunumu: Domuz Akciğerlerinden *Alysiella filiformis* İzolasyonu

Özet

Alysiella filiformis birçok hayvanın ağız boşlukları ortak bir sakini olarak kabul edilir. Literatüre göre etkenin sıcakkanlı hayvanların ağız boşluğu ile sınırlı ve patojenik olmadığı bildirilmiştir. Sırbistan bir çiftlikte genç domuz kayıpları artışı gözlenmiştir. Postmortem incelemede akciğer kan damarları ve bronşlarda kısmen pıhtılaşmış kan ve köpüklü içerik gözlenmiştir. Ödem hastalığının karakteristik işaretler mevcut olduğunu ve *E. coli* serogrup 0139 izole edildi. Ayrıca, *Alysiella filiformis* hastalıklı domuz akciğerlerinden izole edilmiş tek bir ajan idi. Bu çalışmada hastalıklı domuz akciğerlerden *Alysiella filiformis* izolasyonu ilk kez bildirilmiştir.

Anahtar sözcükler: *Alysiella filiformis*, Domuz, Akciğerler

INTRODUCTION

Alysiella filiformis (*A. filiformis*) is considered a common resident in the oral cavities of many animals. All reports of *Alysiella* indicate that it is restricted to the oral cavity of warm-blooded vertebrates, where it apparently is nonpathogenic. *A. filiformis* is the solitary member of genus *Alysiella*, family *Neisseriaceae* ^[1,2]. Genus *Alysiella* comprises organisms that exist in characteristic flat, ribbon-like multicellular filaments. The cells within the filament are paired, and in axenic culture the filament often breaks up into groups of two or four cells. The width of an individual cell is about 2.0–3.0mm, and the length of a cell is about 0.6 mm ^[1,2]. *A. filiformis* has been isolated from the oral cavity of different animals including guinea pigs ^[3], sheep ^[4], and cows ^[5], and in Turkey it was isolated from hot spring

water indicating its presence in the environment ^[6]. This paper presents a case of the isolation of *Alysiella filiformis* from the lungs of pig with oedema disease.

CASE HISTORY

During May 2009, one swine farm with 1.000 pigs situated in northwest of Serbia, reported increased losses of young pigs. According to anamnestic data, live affected pig showed a staggering gate, puffy eyelids giving a sleepy appearance and an abnormal high pitched squeak. During nine days, 14 pigs (weight around 15 kg) in very good condition died, and dispnea was the only clinical symptom present in diseased pigs. Post-mortem examinations showed



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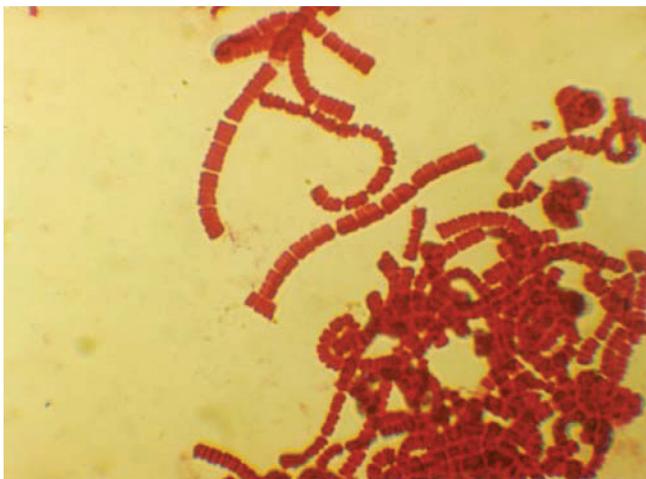
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Table 1. Biochemical characteristics of *Alysiella filiformis* isolate**Tablo 1.** *Alysiella filiformis* biyokimyasal özellikleri izole

Medium	Reaction	Medium	Reaction
Catalase	positive	Inositol	acid not produced
Oxidase	positive	Xylose	acid not produced
Glucose	acid produced	Arabinose	acid not produced
Maltose	acid produced	Mannitol	acid not produced
Trehalose	acid produced	Gelatin	negative
Sucrose	acid produced	H ₂ S	not produced
Fructose	acid produced	MR	negative
Lactose	acid not produced	VP	negative
Cellobiose	acid not produced	Indole	negative
Raffinose	acid not produced	Urease	negative
Rhamnose	acid not produced	Aesculin	not hydrolyzed
Mannose	acid not produced	Nitrate	not reduced
Dulcitol	acid not produced	Nitrite	not reduced
Salicin	acid not produced	Simmon's citrate	no growth

**Fig 1.** Colonies of *A. filiformis* on blood agar**Şekil 1.** Kan agar *A. filiformis* kolonileri**Fig 2.** Ribbon-shaped filaments of *A. filiformis* (Gram stain, x1.000)**Şekil 2.** *A. filiformis* şerit şeklindeki filamentler (Gram boyama, x1.000)

oedema of the greater curvature of the stomach wall, coiled colon and eyelids. Spumous content in bronchia and partly clotted blood in blood vesels of the lungs were present.

Samples of small intestines were inoculated on nutritient agar (HiMedia) containing 5% sheep blood and MacConkey agar (HiMedia), and incubated aerobically at 37°C for 24 h. Lung samples were also inoculated on 5% sheep blood agar and incubated aerobically at 37°C for 24 h. The cultural, biochemical and morphological characteristics of grown cultures were examined. Bacterial isolates were identified using standard methods for phenotypic characterization as previously described [1,2,7,8]. After 24 h of incubation, *E. coli* was grown on sheep blood agar and MacConkey agar from the small intestine samples. Using antisera (*E. coli* O antisera, HiMedia), isolate was identified to belong to serogroup O139. From lung samples, no growth was seen on MacConkey agar, but large numbers of smooth, convex, β-haemolytic, white-grayish colonies, about 1.5mm diameter, were present in pure culture on sheep blood agar.

After subcultivation on MacConkey agar at 37°C and 25°C, as well as on sheep blood agar at 25°C, culture was sterile, but bacterial growth occurred after subcultivation on sheep blood agar at 37°C. Gram staining of isolate revealed gram-negative cells, 0.6 x 2-3 µm, arranged side by side in pairs to form flat, ribbon-shaped filaments, with variable lenght.

Isolate was further tested by biochemical tests (bioMerieux, HiMedia).

On the basis of the cell morphology, cultural and biochemical characteristics, isolate was identified as *Alysiella filiformis*.

DISCUSSION

In difference from the other representatives of this family, cells of genera *Simonsiella* and *Alysiella* may exhibit a characteristic multicellular micromorphology. Moreover, the unusual morphology of the *Alysiella* filaments serves to differentiate the genus from all other procaryotic organisms. *Alysiella* filaments differ from *Simonsiella* in consisting of continuous pairs of cells instead of being segmented into units of eight cells, and the terminal cells are not rounded, but square [2]. In this case, *A. filiformis* was isolated from lung tissue in pure culture, with ribbon-shaped filaments of with variable lenght, similar to other authors [2]. The biochemical properties of bacteria isolated from lung tissue in this study are very similar to those described in other studies [2,7,8].

The origin and significance of the presence of *A. filiformis* in the lung tissue is not clear. *A. filiformis* has been described as a comensal in oral cavity of animals [3-5], but

there is no report of its presence in other tissues or organs. Also, there is no known pathology as a consequence of presence of this bacterium. Because of this fact, it is hard to determine the role bacterium in described macroscopic lesions present in lung tissue of diseased pig. In the present case, suspicion of possible role of *A. filiformis* in lung pathology was based on fact that it was isolated from lung tissue in pure culture, with abundant growth and presence of clearly visible zone of beta-haemolysis around colonies.

The reasons for the colonization of lung tissue with *A. filiformis*, and its potential role in the etiology of pathoanatomical changes in lung tissue remain unclear. Further studies are needed to determine the pathogen potential of this bacterium.

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