

# Isolation of *Brucella Suis* from Aborted Fetus of a Dog

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—圖文抄錄—

## 流產犬 胎兒로부터 *Brucella suis* 의 分離

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우리나라 家畜中 牛 및 豚의 brucellosis 는 알려져 있으나 犬에서의 brucella 分離에 關한 報告는 볼 수 없  
다. 著者들은 1971年 8月 German shepherd 의 流產한 55日犬胎에서 brucella 라고 生覺되는 菌을 分離하여  
그 性狀을 追究하여 *Brucella suis* 로 同定하였다. 이로서 우리나라에도 brucella 에 依한 犬의 流產이 있음을  
알수 있다. 流產犬의 血清抗體價는 *Br. suis* 에 對하여 320倍였다.

大邱市 屠畜場에서 얻은 86例의 犬의 血清의 brucella 에 對한 抗體價를 檢査하였던바 *Br. abortus* 에는 1  
例만이 80倍陽性이었고, *Br. suis* 에는 3例가 160倍, 8例가 80倍陽性였다. 分離菌에 對한 抗體價는 大體로  
*Br. suis* 에 對한 抗體價와 平行하였다.

Brucellosis has been considered as an important problem in the breeding of domestic animals and the public health of human beings. The rise of excellent pure-bred dogs is necessary for hunting and military purposes, but the breeding of dogs in Korea to meet these purposes is hampered by infertility and abortion which may in part be caused by infection with *Brucella*.

Although dogs are rather resistant to *Brucella*, there have been numerous reports of

canine infection with classical strains of *Brucella*(1,2). Dogs have even been infected occasionally with *Brucella* by contact with infected domestic animals. Infected dogs manifest abortion, orchitis, epididymitis, lymphadenopathy and arthritis. Epidemic brucellosis among dogs associated with *Br. canis* have occurred in the United States since 1964 (3-7). There are some reports on the infection of *Brucella* in cattle and swine in Korea (8-11), but the involvement of *Brucella* in

dogs has not been reported.

In this study, the authors report the isolation of a strain of *Br. suis* from an aborted fetus of a four year-old German Shepherd.

## MATERIALS AND METHODS

### Isolation procedures:

As shown in Figure 1, approximately one gram of minced liver, spleen and intestine were mixed and incubated in Tryptic Soy broth (B.B.L.) containing one per cent sodium chloride. At intervals of several days, subcultures were made to Tryptic Soy broth and Tryptic Soy agar (B. B. L.) containing one per cent sodium citrate. All cultures were incubated in 10 per cent CO<sub>2</sub> tension at 37°C. Colonies suspected of *Brucella* were transferred to Tryptic Soy agar slant, and biological as well as immunological characteristics were studied. *Br. abortus* 1119-3 and *Br. suis* 13 obtained from the Veterinary Research Laboratory, Anyang were used as control. *Br. melitensis* and *Br. canis* were not available for the study.

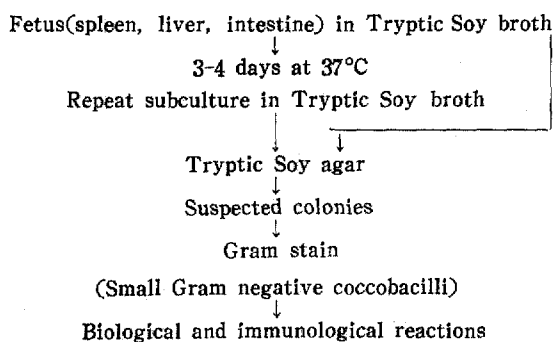


Figure 1. Isolation procedures of *Brucella* strain

### Biological characteristics:

Test and control strains were confirmed before use to be smooth by negative agglutination in acriflavine solution. Carbohydrate fermentation were studied by incubating organisms in Tryptic Soy broth containing one per cent carbohydrates. The sensitivity to dye was tested by incubating organisms in

Tryptic Soy broth containing varying concentrations of dye with or without 10 per cent CO<sub>2</sub> tension; no visible growth within 5 days was considered sensitive.

### Immunological characteristics:

Antisera were prepared in this laboratory by injecting heat-killed suspension of cultures obtained from Tryptic Soy agar plates into the vein of inbred adult rabbits. At four day intervals, each rabbit received repeated injection in progressively larger doses. When sufficient increase of antibody was confirmed, the rabbit was exsanguished.

### Antibody titers of dog sera harvested from field survey:

In order to know the antibody level of dogs in normal population, sera were collected from 86 dogs in Taegu city slaughter house during the period from September to November, 1971 and the antibody titers to *Br. abortus*, *Br. suis* and the study isolate were measured by the tube agglutination reaction.

## RESULTS

### Biological characteristics:

A bacterial strain was isolated from a 55 day-old aborted fetus of a German Shepherd in August, 1971. This strain grew well aerobically and did not require CO<sub>2</sub> tension for

Table 1. Dye sensitivity of the isolate and *Brucella*\*

Strain	Thio- nine	Basic fuchsin	Crystal violet	Safra- nine	Azure A	Pyro- nine
<i>Br. abortus</i> 1119-3	-	+	-	-	-	-
<i>Br. suis</i> 13	+	-	+	+	+	+
Isolate	+	-	+	+	+	+

\* Dyes in Tryptic Soy broth.

Concentrations of dyes:

Thionine, 1:50,000; Basic fuchsin, 1:25,000;  
Safranin, 1:20,000; Crystal violet, 1:40,000;  
Azur A, 1:100,000; Pyronine, 1:500,000.

+ = growth, - = no growth.

growth. The strain was Gram negative short rods and non-motile. It fermented glucose, mannose, maltose, and trehalose, and did not ferment inositol and rhamnose. The sensitivity to dye is shown in Table 1. The strains was sensitive to basic fuchsin, and resistant to thonine, crystal violet, safranin, Azur A, and pyronine. The sensitivity pattern is completely that of *Br. suis* and quite contrary to *Br. abortus*.

#### Immunological reactions:

Table 2 shows the agglutination reaction to antisera of the isolate and control strains. The isolated strain reacted strongly with sera of its own and of both *Br. abortus* and *Br. suis*. Therefore, the absorption reaction was carried out (Table 3). Antiserum of *Br. abortus* was completely absorbed by *Br.*

**Table 2.** Agglutination reaction of the isolate

Serum immunized with	Agglutinin titers* to		
	<i>Br. abortus</i>	<i>Br. suis</i>	Isolate
<i>Br. abortus</i>	1,600	1,600	1,600
<i>Br. suis</i>	3,200	3,200	3,200
Isolate	3,200	3,200	3,200

\* Titers are expressed as the reciprocal of serum dilutions.

**Table 3.** Agglutinin absorption reaction of the isolate

Antisera	Absorbed with	Titers* after absorption		
		<i>Br. abortus</i>	<i>Br. suis</i>	Isolate
<i>Br. abortus</i>	<i>Br. abortus</i>	—	—	—
	<i>Br. suis</i>	—	—	—
	Isolate	—	—	—
<i>Br. suis</i>	<i>Br. abortus</i>	—	100	100
	<i>Br. suis</i>	—	—	—
	Isolate	—	—	—

\* Titers are expressed as the reciprocal of serum dilutions, — = negative at serum dilution of 1:100.

*abortus*, *Br. suis* and the isolate, but there was incomplete absorption of antibody to *Br. suis* and the isolate when *Br. suis*-antiserum was absorbed with *Br. abortus*. Antiserum of the isolate was not available for the absorption, since the rabbit was dead after preliminary bleeding.

#### Antibody levels of sera collected from field survey:

As shown in Table 4, only one serum showed the titer of 1:80 to *Br. abortus*. Three sera were 1:160 in titer and 8 sera were 1:80 to *Br. suis*, and 9 were 1:80 to the isolate. Most of sera showing 1:80 or more in titers to *Br. suis* were also 1:80 to the isolate.

**Table 4.** Agglutinin titers of dog sera collected from field survey\*

Serum No.	Agglutinin titers**		
	<i>Br. abortus</i>	<i>Br. suis</i>	Isolate
1	—	160	80
2	80	80	80
3	—	80	—
23	—	80	—
25	—	160	80
35	—	160	80
37	—	—	80
51	—	80	80
53	—	80	80
58	—	80	—
60	—	80	80
69	—	80	80
The other 74 sera	—	—	—
Aborted dog	—	320	160

\* Eighty-six sera were collected from field survey.

\*\* Titers are expressed as the reciprocal of serum dilution, — = less than 80.

## DISCUSSION

The strain isolated from the aborted dog

fetus was considered to be *Br. suis* by its biological characteristics. It is hard to characterize the strain by immunological reactions due to its cross reactivity with sera of control strains, but the absorption reaction showed the characteristic of *Br. suis*. The epidemic strain, *Br. canis*, was not used for control. However, the isolate was shown not to be *Br. canis* by biological and immunological characteristics easily differentiated from classical strains of *Brucella* (12).

The isolation of *Br. suis* from cattle and swine has been reported in Korea (9, 11), and we suppose that the infection of dogs by *Br. suis* caused by the contact within infected swine or cattle. The field survey of the level of serum antibody in domestic dogs reveals considerable number with titers of 1:80 or more to *Br. suis* and to the isolate. This suggests the presence of clinical and subclinical infections of *Br. suis* among dogs. Further study is necessary to determine the significance of brucellosis among dogs.

### SUMMARY

A strain of *Brucella* showing the characteristics of *Br. suis* was isolated from a 55 day-old aborted fetus of a German Shepherd and the cause of the abortion was considered due to brucellosis. A considerable number of dog sera collected at the slaughter house in Taegu city showed antibody titers of 1:80 or more to *Br. suis*, and serum from the aborted dog 1:320 to *Br. suis* in titer.

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