

THE EFFECT OF *Usnea* spp. EXTRACT TO THE GROWTH OF SOME MORAXELLA ISOLATES

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ABSTRACT

The aim of this study was to determine the inhibition effect of *Usnea* spp. extract against the growth of some *Moraxella* sp. isolates by *in-vitro* method. Three isolates of *Moraxella* sp. (I, 1589 and II) were obtained from the cattle that suffer from pink eye disease in Central Java used in this study. Every isolate was tested against several concentrations (10.0, 7.5, 5.0, 2.5 and 1.25%) of *Usnea* spp. extract. The result indicated that the higher the extract concentration, the higher the growth inhibition of *Moraxella* sp. obtained ($P < 0.05$). The growth inhibition of 10.0, 7.5, 5.0, 2.5 and 1.25% extract concentration were 22.7, 19.7, 17.3, 13.3 and 10.7 mm respectively. The most sensitive *Moraxella* sp. isolate to *Usnea* spp. extract was *Moraxella* isolate II ($P < 0.05$). The growth inhibition of *Moraxella* isolate I, 1589 and II were 13.2, 17.6 and 19.4 mm respectively. It was concluded that *Usnea* spp. extract could inhibit the growth of *Moraxella* sp.

Key words: *Usnea* spp., *Moraxella*, Growth inhibition.

INTRODUCTION

Moraxella sp. is the aetiological agent of infectious keratoconjunctivitis (Buchanan and Gibbon, 1975, Chandler *et al.*, 1979, and Gray *et al.*, 1995). Infectious keratoconjunctivitis is also called pink eye or ophthalmia or New Forest eye diseases. Pink eye occurred in cattle at all ages, but cattle less than 2 years old tends to be more sensitive (Foldi *et al.*, 1992). There was an outbreak of pink eye in 1977, in which 301 cattle in South Kalimantan was suffered (Anonymous, 1981).

The clinical signs of pink eye were profuse lacrimation, photophobia, severe conjunctivitis, in some of the affected animals, corneal opacity which developed centrally and later became diffuse (Foldi *et al.*, 1992). Pink eye that was caused by *Moraxella* sp. usually treated with antibiotic (Fatimah *et al.*, 1994), but the growth of *Moraxella* sp. might also be inhibited by plant extract which had antibiotic or bacteriostatic properties.

Usnea spp. is one of commonly traditional plant. It usually grows at the highland, and stick on the other plant by mutualism symbiosis. According to Schauenberg (1977) *Usnea* spp. had antibiotic and bacteriostatic properties. *Usnea* spp. contain the active component called usnat acid (2,6-diacetyl-7,9 dihydroxy-8,9 dimethyl-1,3) as tuberculostatic and bacteriostatic to gram positive bacteria (Windholz *et al.*, 1983). *Usnea* spp. extract had been proven to inhibit the growth of *Staphylococcus aureus* isolates (Poeloengan *et al.*, 1992). Because of its properties as antibiotic and bacteriostatic, so *Usnea* spp. extract is expected to inhibit the growth of *Moraxella* sp. isolates.

MATERIAL AND METHOD

This study was assigned to 3 x 5 factorial arrangement of treatments to examine the effect of 3 isolates of *Moraxella* sp. and 5 concentrations of *Usnea* spp. extract. Each treatment was replicated three

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Table 1. The effect of *Usnea* spp. extract concentration to growth inhibition

Extract concentration (%)	Growth inhibition (mm)
10.0	22.67 ^a
7.5	19.67 ^b
5.0	17.33 ^c
2.5	13.33 ^d
1.125	10.67 ^e

*Means within column with different superscripts are significantly different ($P < .05$)

times. Duncan's Multiple Range Test estimated the differences between treatment means.

Three *Moraxella* isolates (I, II and 1589) used in this experiment were *Moraxella* isolates collection of Balitvet-Bogor. *Usnea* spp. was collected from Central Java. *Usnea* spp. was extracted by hexane for 24 hours and then filtered. Its filtrate was concentrated by rotavapor at 35 °C.

Anti bacterial study was done at Balitvet, Bogor. Fifteen microlitres of *Usnea* spp. extract were dropped into sterile disk, then were laid on MEU blood media that had been inoculated with *Moraxella* sp. isolate and incubated at 37 °C overnight. The next day, the growth inhibition on every plate was observed.

RESULT AND DISCUSSION

Usnea spp. extract concentration

Increasing extract concentration resulted in significantly increased the growth inhibition of *Moraxella* sp. isolates ($P < .05$) (Table 1)

The result indicated that the higher the *Usnea* spp. extract concentration, the higher its ability to inhibit the growth of *Moraxella* sp. isolates ($P < .05$). It might be caused by increasing the concentration of usnat acid as bacteriostatic component in the extract. This result agrees with Windholz *et al.* (1983) that *Usnea* spp. contain usnat acid as antibiotic and bacteriostatic.

Moraxella sp. isolates

Three *Moraxella* sp. isolates (I, II, and 1589) used in this study had significantly different sensitivity ($P < .05$) to *Usnea* spp. extract (Table 2). The growth of *Moraxella* isolate II was the most inhibited by *Usnea* spp. extract ($P < .05$), while the growth of *Moraxella* isolate I was the least inhibited. The result indicated the *Moraxella* isolate II was the most sensitive to *Usnea* spp. extract. The sensitivity differences among three *Moraxella* isolates used in this study might be affected by previous antibiotic application to the affected animal. According to Poeloengan *et al.* (1992) continuity of antibiotic application resulted in resistency.

Table 2. The growth inhibition of three *Moraxella* isolates

<i>Moraxella</i> isolate	Growth inhibition (mm)
I	13.20 ^a
II	19.40 ^a
1589	17.60 ^b

*Means within column with different superscripts are significantly different ($P < .05$)

Table 3. The effect of *Usnea* spp. extract to the growth inhibition of three *Moraxella* isolates

Extract concentration (%)	Type of isolate	Growth inhibition (mm)
10.0	I	20.00 ^{c*}
7.5	1589	20.00 ^c
5.0	II	28.00 ^a
2.5	I	16.00 ^d
1.25	1589	21.00 ^{bc}
10.0	II	22.00 ^b
7.5	I	12.00 ^e
5.0	1589	20.00 ^c
2.5	II	20.00 ^c
1.25	I	10.00 ^f
10.0	1589	15.00 ^d
7.5	II	15.00 ^d
5.0	I	8.00 ^g
2.5	1589	12.00 ^e
1.25	II	12.00 ^e

*Means within column with different superscripts are significantly different (P<.05)

Table 3 showed that *Moraxella* isolate II was the most sensitive isolate to *Usnea* spp. extract especially at high concentrations (10.0 and 7.5%), while the isolate I of *Moraxella* sp. was the least sensitive to *Usnea* spp. extract at all concentrations (P<.05). This result suggested that in order to inhibit the growth of *Moraxella* sp. isolate II would be more effective at high concentration of *Usnea* spp. extract. And, the sensitivity differences to *Usnea* spp. extract might be caused by previous antibiotic application to affected animals.

CONCLUSION

1. Increasing *Usnea* spp. extract concentration resulted in increasing the growth inhibition of *Moraxella* isolates significantly (P<.05).
2. Isolate II of *Moraxella* sp. was the most sensitive to *Usnea* spp. extract at 10% and 7.5% concentrations.

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