

PATHOGENICITY OF *CHLAMYDOPHILA ABORTUS* ISOLATES IN MICE AND GUINEA PIGS

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Chlamydial infections among domestic animals are increasingly being recognized in India. The association of *Chlamydomphila abortus* in the causation of abortion and pneumonia in ruminants had been reported from different parts of the country (Francis, 1988; Griffiths *et al.*, 1995; Batta *et al.*, 1996 and Katoch, 1997). Mice and Guinea pigs have been employed for isolation and virulence studies of chlamydial isolates of ruminant origin (Storz, 1971 and Page, 1981). Guinea pigs infected intra peritoneally developed spleenomegaly, hepatomegaly and a stringy fibrinous exudate in the peritoneal cavity, while mice developed hyperemic lungs, enlarged spleen and liver and sticky exudates in peritoneal and thoracic cavities (Page, 1981). This communication places on record the observed differences among isolates of *Chlamydomphila abortus*, from cases of abortion in cattle and goats, for their pathogenic potential in mice and guinea pigs.

Materials and Methods

A reference strain obtained from the department of Veterinary Microbiology, College of Veterinary & Animal Sciences, Palampur and two local isolates of *Chlamydomphila abortus* were used for the study.

Six mice (BALB/C, aged three to four weeks) and two guinea pigs (NIH-coloured, aged four to five weeks) were inoculated intraperitoneally with each of the isolate (20 per cent of heavily infected yolk sac suspension after third passage).

For mice, 0.2 ml of infected yolk sac (YS) suspension in SPG (Katoch, 1997) and for guinea pigs three milliliters of infected YS suspension in PBS (pH – 7.2) were used. Along with them two mice and a guinea pig were kept as control by inoculating 0.2 ml SPG and three milliliters of PBS (pH 7.2) respectively.

The inoculated animals were observed daily for the development of clinical signs or death. The dead animals were necropsied and examined for any gross lesions. Representative samples from organs viz., lungs, liver, spleen and peritoneal exudates were collected for re-isolates. Impression smears of affected areas of these organs and exudates were prepared for staining by Giemsa, modified Ziehl Neelsen and Gimenez methods. The animals, which remained apparently healthy after three weeks following inoculation were sacrificed and subjected to examination as above. The pathological changes were noted in lungs,

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liver, spleen and peritoneal exudates were collected for demonstration of chlamydiae by staining impression smears and also for re-isolation.

Results and Discussion

Pathogenicity in mice

Five of the mice, which received the bovine isolate showed anorexia, dullness and ruffled hairs by seven to eight days, later their movement was found sluggish. One of them died on 14th day post inoculation (PI) and the other four survived. The other mouse showed these clinical signs with less severity but survived. The mortality rate was 16.7 per cent. On post mortem examination, all the inoculated mice revealed peritonitis with stringy exudates and congestion of lungs, spleen and liver. Hepatic necrosis was noticed in the dead mouse in addition to the above lesions.

All the mice inoculated with caprine isolate also showed similar clinical signs as the bovine isolate. Four of them run down in condition by eleventh day, of which two died on 12th day and the other two on 14th day PI. Necropsy of the dead mice revealed severe pulmonary congestion and pneumonic lesion, but the liver and spleen showed mild degree of congestion. In the case of sacrificed mice, peritonitis and adhesion of internal organs were noticed. The mortality rate was 66.7 per cent for caprine isolate.

Even though all the six mice inoculated with the reference bovine isolate showed anorexia and dullness by seven to eight days, only two died, one on twelfth day and the other on fifteenth day of PI. Post mortem examination of the dead mice revealed fibrinous exudates in the peritoneal cavity and severe hepatic and pulmonary congestion with patchy necrosis in liver and lungs. Spleenic congestion was of mild degree. Survived mice revealed moderate degree of peritonitis, adhesion of liver to peritoneum and whitish peritoneal exudates.

Impression smears from organs of all the inoculated mice revealed chlamydial elementary bodies (CEB) on staining by Giemsa, Giemenez and modified Ziehl Neelsen. The organism could be re-isolated

from pooled organ samples of mice in embryonated eggs by YS route. The typical lesions and elementary bodies of *C. abortus* could be observed in the inoculated yolk sac.

Control group did not show any clinical signs or any lesions on internal organs after sacrifice. Impression smears of its organs also did not reveal any chlamydial bodies.

Pathogenicity in guinea pigs

Only one of the guinea pigs that was inoculated with caprine isolate died 10 days PI. All the inoculated guinea pigs showed anorexia and dullness by seven to eight day PI. Post mortem examination of sacrificed guinea pigs, inoculated with local bovine isolate, revealed mild peritonitis and moderate congestion of liver and lungs. The dead guinea pig that received local caprine isolate revealed severe pulmonary congestion, hepatic necrosis and yellowish stringy fibrinous exudates in the peritoneal cavity, but the one that survived had only mild congestion of lungs and liver. The sacrificed mice revealed moderate congestion of lungs, liver and spleen and mild degree of peritonitis.

The impression smears of lungs, spleen and liver and peritoneal exudates revealed CEB on staining. The pooled organ samples of the inoculated animals gave re-isolation by passaging in the YS of chick embryo. The infected YS smears also confirmed the presence of CEB on staining by the above three methods.

The control animal did not show any illness or lesions in organs when it was sacrificed. Impression smears could not reveal any CEB of *C. abortus* at all.

In the present study, depending upon the severity of infection, mortality of mice ranged from 16.7 to 66.7 per cent. Mortality of mice occurred during 12 to 15 days PI. Intra peritoneal inoculation of mice with virulent strains produced death in three to fourteen days depending on the virulence (Storz, 1971; Francis, 1988; Sreeramulu *et al*, (1989). This indicated that isolates in the present study were of moderate virulence in mice.

Pulmonary and hepatic congestion, pneumonia, focal necrotic liver and stringy peritoneal exudates were the prominent lesions in mice in this study. The bovine strains were of moderate virulence while the caprine strain had greater virulence in mice compared to the other two. However, Storz (1971) and Page (1981) observed that chlamydial isolate from ruminants were of low pathogenicity for mice.

Page (1981) observed that *Chlamydomphila abortus* organisms were less pathogenic to guinea pigs when compared to mice. These results were in accordance with the observations made in the present study. Clinical manifestation and the necropsy lesions reported by Batta *et al.* (1997) were similar to this study, including congestion and necrosis of internal organs and peritonitis.

Summary

The pathogenicity of three *Chlamydomphila abortus* isolates in mice and guinea pigs was studied. It was found that the organisms were less pathogenic to guinea pigs when compared to mice.

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