

## Abortion in Mice Associated with *Pasteurella pneumotropica*

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*Pasteurella pneumotropica* was isolated from the uteri, fetuses, lungs, and spleens of aborting Swiss Carworth mice. Male mice in the colony carried *P. pneumotropica* in pharynges, testes, and seminal vesicles. Normal pregnant and nonpregnant females carried *P. pneumotropica* in the eye of 1 and in the uteri of 4 of 11. Pregnant mice from another colony did not abort when injected with *P. pneumotropica*. Necrotizing and suppurative metritis was found among aborting females with *P. pneumotropica* infections. Occurrence of malignant lymphoma and mammary adenocarcinoma among animals in this colony likely resulted in immunosuppression which could have predisposed animals to the diseases seen.

*Pasteurella pneumotropica* can be found in the nasopharynges, brains, and uteri of normal mice (1, 2, 4-8). The organism was characterized in 1950 by Jawetz (7). By rapid serial passage of the organism, he was able to produce pneumonic lesions in mice. *P. pneumotropica* has been isolated from mice having pneumonia, conjunctivitis, metritis, cystitis, pleural abscesses, peritoneal abscesses, dermatitis, panophthalmitis, and orbital abscesses (for a review, see reference 10). Other animals and humans have also been infected by the organism (3, 10).

The occurrence of *P. pneumotropica* in the uteri of normal mice has been reported at the rate of 4 to 33% in various surveys (2, 4). Obviously, the organism is present in many mice without causing disease. This report deals with an epizootic of abortion in a single colony of mice from which *P. pneumotropica* was isolated from most uteri and fetuses of aborting females examined. The organism was also found in the seminal vesicles of the active breeding males in this colony.

### CASE REPORT

On 16 August, 1976, an aborting Swiss Carworth mouse used in nutrition research (Department of Animal Science, University of Saskatchewan) was submitted for necropsy. From this date until 21 October, 1976, 10 mice from this colony in various stages of abortion were submitted.

The abortion storm involving *P. pneumotropica* occurred only among the Swiss Carworth mice, although other breeds were housed in the same room. *P. pneumotropica* was isolated from 7 of 10 aborting mice submitted (Table 1). The organism was isolated from the uteri, fetuses, lungs, and spleens of various animals. The epizootic was terminated by the destruction of the colony.

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### MATERIALS AND METHODS

Routine necropsies were performed on all mice submitted. At the time of necropsy, specimens of uteri, spleens, lungs, hearts, seminal vesicles, fetuses, and swabs of nasopharynges and/or eyes were taken from selected animals for bacteriological examination. These specimens were cultured as follows: blood agar at 37°C in air, MacConkey agar at 37°C in air, blood agar with staphylococcus streak at 37°C in 5% CO<sub>2</sub>.

Specimens of the major organs described above, as well as any organ appearing grossly abnormal, were placed in 10% buffered Formalin for histological examination. These portions were processed routinely, sectioned at 6 μm, and stained with hematoxylin-eosin for light microscopic evaluation.

### RESULTS

Microscopic examination of material from uteri and fetuses of aborting mice showed gram-negative bacilli, 0.8 by 1.2 μm to 0.8 by 5 μm. *P. pneumotropica* was cultured in the uteri of 7 of 10 aborting mice (Table 1). In 4 of the 10 aborting mice, *P. pneumotropica* was found in either spleens or lungs, indicating septicemia. A variety of other organisms (*Escherichia coli*, alpha-hemolytic streptococci, *Proteus* sp.) were found in the organs of some mice.

*P. pneumotropica* grew initially only in CO<sub>2</sub>, but adapted to growth in air rapidly. Urease was produced rapidly and abundantly. Nine *P. pneumotropica* strains were examined in detail for biochemical characteristics. One drop of horse serum was added to all media to encourage growth. The following characteristics were uniform for all strains. Organisms were fermentative, catalase, and oxidase positive and reduced nitrate. H<sub>2</sub>S was produced on lead acetate paper but not in triple sugar iron agar, urease was produced, and acid was produced from glucose,

TABLE 1. *Pathological lesions and culture results from aborting female mice*

Pathological lesions	Organ cultured	Bacteria isolated
Necrotizing ulcerative metritis; four resorbing feti	Uterus	<i>P. pneumotropica</i> <i>E. coli</i>
	Spleen	No growth
	Lung	<i>P. pneumotropica</i> <i>E. coli</i> (hemolytic) Streptococci (alpha-hemolytic) <i>P. pneumotropica</i>
Suppurative metritis	Uterus	<i>P. pneumotropica</i> Streptococci (anaerobic)
	Lung	<i>P. pneumotropica</i> <i>E. coli</i>
	Spleen	<i>P. pneumotropica</i> Streptococci (alpha-hemolytic)
Necrotizing suppurative metritis; five normal fetuses (right horn), two resorbing feti (left horn)	Uterus	<i>P. pneumotropica</i> <i>P. mirabilis</i>
	Spleen	<i>P. pneumotropica</i>
	Fetus (normal)	<i>P. pneumotropica</i>
	Fetus (resorbed)	<i>P. pneumotropica</i> <i>E. coli</i> <i>P. mirabilis</i>
Suppurative metritis; rupture of right horn; neoplastic lymphocytes present	Uterus	<i>P. pneumotropica</i> <i>E. coli</i> <i>P. vulgaris</i>
Ulcerative metritis	Uterus	<i>Proteus</i> sp.
No lesions	Fetus, uterus, spleen	No growth
Necrotizing suppurative metritis	Fetus, spleen, uterus	No growth
No lesions	Uterus	<i>P. pneumotropica</i>
Two resorbing feti; osteosarcoma of lumbar vertebra	Uterus, spleen	<i>P. pneumotropica</i>
Necrotizing suppurative metritis	Uterus	<i>P. pneumotropica</i>

sucrose, maltose, trehalose, raffinose, levulose, and mannose. Organisms were phenylalanine deaminase negative, did not grow on citrate or malonate, were nonmotile, and did not hydrolyze gelatin or produce acid from sorbitol or dulcitol. Variable reactions were found for indole production, esculin hydrolysis, and acid production in arabinose, xylose, galactose, lactose, inulin, mannitol, inositol, salicin, and rhamnose. All media were examined for 4 days before being called negative.

Of the 10 aborting female mice examined, 7 exhibited gross and/or microscopic evidence of a necrotizing suppurative metritis (Table 1). This was characterized by necrosis and loss of uterine epithelia, with large numbers of polymorphonuclear leukocytes invading all layers of the uteri and accumulating in the lumina. In one case, rupture of the right uterine horn had resulted in peritonitis. Fetal resorption was present in 3 of the 10 animals. An incidental finding

TABLE 2. *Pathological lesions and culture results from male mice*

Pathological lesions	Organ cultured	Bacteria isolated
None	Spleen, heart, testes, seminal vesicle	No growth
None	Seminal vesicle, testes	<i>P. pneumotropica</i>
Emaciation	Spleen	<i>Proteus</i> sp. <i>E. coli</i>
	Lymph node	<i>Proteus</i> sp.
None	Seminal vesicle	<i>P. pneumotropica</i>
None	Seminal vesicle	No growth
None	Seminal vesicle	No growth
None	Seminal vesicle	<i>Bacillus</i> sp.
None	Pharynx	<i>P. pneumotropica</i>
None	Seminal vesicle, eye, nose, spleen, testes	No growth
None	Eye	<i>S. epidermidis</i>
	Nose, pharynx	Streptococci (alpha-hemolytic)
None	Spleen, testes, seminal vesicle	No growth

TABLE 3. *Pathological lesions and culture results from nonaborting female mice*

Pathological lesions	Organ cultured	Bacteria isolated
Suppurative metritis	Uterus	<i>Streptomyces</i> sp.
	Lung	No growth
	Fetus	No growth
Nongravid; no lesions	Uterus	<i>P. pneumotropica</i>
Gravid; six normal feti	Uterus	<i>P. pneumotropica</i>
Nongravid; bilateral conjunctivitis	Uterus	<i>P. pneumotropica</i> <i>E. coli</i> <i>P. mirabilis</i> <i>K. pneumoniae</i> <i>Alcaligenes</i> sp.
	Eye	
Wild mouse, lymphosarcoma	Spleen	No growth
	Lymph node	<i>Proteus</i> sp.
Mammary adenocarcinoma, right prescapular area; nongravid	Uterus	No growth
Rearing 3rd litter; lymphosarcoma	Eye, nasopharynx	No growth
Two litters, nongravid; mammary adenocarcinoma; right axilla lymphosarcoma	Nasopharynx	No growth
Nongravid mammary adenocarcinoma, right inguinal area	Nasopharynx	No growth
Virgin; conjunctivitis	Nasopharynx	No growth
Virgin; conjunctivitis; lymphosarcoma	Nasopharynx	No growth
Nongravid mammary cystadenocarcinoma metastatic to lung	Eye	<i>P. pneumotropica</i>
	Nasopharynx	No growth
Nongravid mammary adenocarcinoma, right shoulder and thorax	Eye, nasopharynx	No growth
Nongravid corneal edema; lenticular degeneration	Eye, nasopharynx	No growth
Nongravid; no lesions	Eye, nasopharynx	No growth
Nongravid; uterine hyperemia	Uterus	No growth
Nongravid; uterine hyperemia	Uterus	No growth
Littered earlier; necrotizing metritis; fetal resorption; lymphosarcoma	Uterus	<i>P. pneumotropica</i>
	Eye	Streptococci (alpha-hemolytic) <i>S. epidermidis</i>
	Nasopharynx	<i>S. epidermidis</i> Streptococci (alpha-hemolytic)
Nongravid; lymphoma	Uterus, spleen	No growth
Failed to conceive, bred twice; mild endometritis	Uterus	No growth

in one animal was an osteosarcoma originating in the lumbar area and producing posterior paralysis. In two cases no gross or microscopic abnormalities were detectable.

Male mice from the colony were examined for the presence of *P. pneumotropica* (Table 2). The organisms were found in the pharynx of one, testes of one, and seminal vesicles of two. These mice had been breeding males used on the aborting females. Specific pathological lesions were not detected in any of the nine male mice examined in detail. One animal was emaciated; however, the cause of the emaciation could not be determined.

Females (pregnant and nonpregnant) were examined for the presence of *P. pneumotropica* in various organs (Table 3). *P. pneumotropica* was found in the uteri of 4 of 11 examined (36%) and in the eye of 1 animal. Other organisms (*Streptomyces* sp., *Escherichia coli*, *Proteus mirabilis*, *Klebsiella pneumoniae*, *Alcaligenes* sp., alpha-hemolytic streptococci, *Staphylococcus epidermidis*) were occasionally found in the uteri and eyes of some animals without evidence of disease. A wide variety of lesions were found in the nonaborting female mice examined. Necrotizing suppurative metritis was detected in only 2 of the 20 animals, whereas mild endometritis was present in a third mouse. Three animals exhibited varying degrees of conjunctivitis. Six cases of lymphosarcoma were identified, as were five cases of mammary adenocarcinoma. One of the cases of lymphosarcoma was found in a wild mouse caught in the area. Four animals in this group were nonpregnant with no striking abnormalities detectable either grossly or microscopically. One animal was pregnant with six normal fetuses in the uterus.

Ten pregnant mice from another colony were given 0.5 ml of an overnight broth culture of *P. pneumotropica* intraperitoneally. None of these mice aborted. All delivered and raised normal litters. An additional 10 mice were challenged with an overnight broth culture of *P. pneumotropica* by intravaginal instillation. No abortion resulted.

Another animal from this colony developed conjunctivitis. *P. pneumotropica* was recovered from the eye of this animal. Three wild mice trapped in the vicinity of the animal colony did not harbor *P. pneumotropica* in any of the organs cultured. One other mouse from another colony at the University of Saskatchewan housed in another building yielded *P. pneumotropica* from a retro-orbital abscess.

## DISCUSSION

The identification of *P. pneumotropica* in 7 of 10 aborting mice in association with fetal resorp-

tion and a necrotizing suppurative metritis suggests that the organism can be pathogenic under certain conditions. Because *P. pneumotropica* was found in normal nonpregnant and pregnant mice, it appears that some additional stress factor is required for the production of disease. Several cases of malignant lymphoma and mammary adenocarcinoma were found in animals from this colony. Both of these conditions are due to viral infections. Many viral infections are known to cause immunosuppression due to replication in the reticuloendothelial and lymphoid systems. Subclinical infection, particularly with mouse leukemia viruses, in this case could have predisposed the animals to infection with an organism ordinarily of low pathogenicity.

Blackmore and Casillo (1) suggest that nasopharyngeal infection allows transmission of *P. pneumotropica* to the vagina, resulting in an ascending infection. Occurrence of this organism in aborting mice and male genitalia has not been previously reported. Another possible mode of transmission could be from male to female at the time of breeding.

Lack of ability of *P. pneumotropica* to produce indole has been reported in 14 of 28 strains examined (6). We confirmed this observation in five of nine strains examined. Our strains produced acid from trehalose and H<sub>2</sub>S. This differentiates them from *P. ureae* (11).

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