RESUMO

Foram evidenciadas lesões histológicas nos pulmões de macacos rhesus (Macaca mulatta) relacionadas ao Pneumonyssus simicola. As principais alterações incluíram numerosos cistos variando de 1-5 mm de diâmetro, com paredes finas e amplamente distribuídos nos pulmões; bronquiolite e peribronquiolite, onde os ácaros foram encontrados associados com materiais particulados pigmentados ou não. Nosso estudo incluiu dados de 347 macacos rhesus submetidos a necropsias durante 20 anos. Four adult debilitated animals were found with pulmonary acariasis which showed a very low incidence of parasite (1.2%) in the colony. Most of the published literature described as common and widespread pulmonary acariasis in Old World monkeys. The present study confirms the ubiquity of P. simicola in captive born and raised rhesus monkeys that would compromise experimental studies involving the respiratory system.

Palavras-chave: Macacos rhesus, acariase pulmonar, Pneumonyssus simicola, patologia.

INTRODUCTION

Pulmonary acariasis (Pneumonyssus simicola) is one of the most frequent parasites in non-human Old World primates and often found in rhesus monkeys (Macaca mulatta) (FURMAN et al., 1974; LEATHERS, 1978; HIRAOKA et al., 2001). There are several species of Pneumonyssus that infest many animals (FURMAN, 1954).

Macroscopically there are multiple cystic lesions up to several millimeters or air-filled bullae often in the hilar region of lung lobes toward the dorsal aspects. These cases showed no other abnormal gross finding (FURMAN et al., 1974; HIRAOKA et al., 2001; KIM; KIM, 2003).

The histopathologic evaluation included the following findings: multifocal granulomatous lesions on the terminal air passages consisting of a large number of eosinophils, epithelioid cells, foreign body type giant cells, and collagen fibers were aggregated around pigmental bodies. At least three distinctive pigmented and non-pigmented materials are identified in association with lung mite infection. Most of pigmental bodies is composed of siliceous materials (KIM; KIM, 2003). The other types were formed by products of mucin...
cases, it may cause death. However, most lung lesions were
infections characterized by cough and dyspnea and in severe
manifestations, the animals could also present serious
necrotizing pneumonia (GILLETT et al., 1984).

infection. The great majority of the cases were caused by
occurs from parasitic infection with superimposed bacteria
infant animals with suppurative complication. This probably
many times leading to high morbidity and mortality (INNES
aggravating factors in case of viral or bacterial pneumonia,
(zoonotic), considered of little clinical importance until certain
infections through coughing and sneezing (POVAR, 1965).

Tracheal washes yield primarily larva, supporting the theory
that larvae migrate to the pharynx and larynx where they are
in the respiratory tracts and feed on host pulmonary epithelial
cells, erythrocytes and lymph (FURMAN, 1977).

Since parasitic members of the order Acari, to which
Pneumonyssus belongs (FURMAN, 1954), do not require
intermediate vectors (FURMAN, 1954; KIM, 1974), it has
been proposed that cross-infection may occur through the
feces, during grooming, or by inhalation of respiratory aerosol.
Tracheal washes yield primarily larva, supporting the theory
that larvae migrate to the pharynx and larynx where they are
transmitted to other monkeys by direct contact, or in heavy
infections through coughing and sneezing (POVAR, 1965).

Pulmonary acariasis is not transmissible to humans
(zoonotic), considered of little clinical importance until certain
aggravating factors in case of viral or bacterial pneumonia,
many times leading to high morbidity and mortality (INNES
et al., 1954). Pneumatoceles or bullae have been reported
in infant animals with suppurative complication. This probably
occurs from parasitic infection with superimposed bacteria
infection. The great majority of the cases were caused by
Staphylococcus aureus infection which causes acute and
necrotizing pneumonia (GILLETTE et al., 1984).

Although the parasite usually does not determine clinical
manifestations, the animals could also present serious
infections characterized by cough and dyspnea and in severe
cases, it may cause death. However, most lung lesions were
discovered as incidental lesions in monkeys that died of pneu-
monia or diarrhea and/or enteritis (HIRAOKA et al., 2001).
The most serious complications resulting in mortality are pneu-
monia, pneumothorax or pulmonary arteritis culminating in a
picture of hemothorax (WOODARD, 1968).

There is strong evidence that air transmission of the P.
simicola can occur which represents a compromise between
keeping primates in large groups. However, as transmission
requires intimate contact with the infected animals (JOSEPH
et al., 1984), it can be avoided by separating the offspring
from the mothers immediately after birth (KNEZEVICH;
MCNULTY, 1970). Cesarean-derived monkeys have been
maintained free from pulmonary acariasis (RAWLINGS;
mite-free monkeys can be raised without difficulty may have
been overly optimistic.

It should be distinguished from that of other granulomatous
diseases including those caused by a strange body and by certain
microorganisms (HIRAOKA et al., 2001). According to Gillett
et al. (1984) advanced mycobacteriosis may cause cavitations
within caseated lung tubercules, but these are characterized as
thick-walled irregular air pockets within densely consolidated
parenchyma. Infectious pneumonia and lung mite infestation
should be considered when pulmonary air-filled lesions are
observed radiographically in nonhuman primates.

Large filled pulmonary lesions can vary from thick,
irregular walled cavitations seen with tumors and abscesses
to thin smooth walled pneumatoceles that are rapidly changing
lesions created by replacement of necrotic tissue with air
(GILLETTE et al., 1984).

The concept of therapy for the control or elimination of
lung mite infestations has received attention (FURMAN et
al., 1974). Ivermectin (200 μg/kg) appears effective for the
elimination of Pneumonyssus sp. infection in non-human
primates (JOSEPH et al., 1984). Chemotherapy with an organic
phosphate has shown promise in reducing the number of active
mite lesions; long-term prophylaxis in a breeding colony may
eradicate the parasite (KNEZEVICH; MCNULTY, 1970). Infected
animals should be isolated from other non-human
primates to prevent transfer of lung mite larvae by coughing
(KIM, 1988). Histopathologically, inflammatory changes
progressively decreased with increasing time post treatment
(JOSEPH et al., 1984).

This study is a survey of the presence of Pneumonyssus
simicola in a closed colony of rhesus monkeys destined for
biomedical research.

**MATERIAL AND METHODS**

**Object of the study**

The studied animals belong to the Departamento de
Primatologia do Centro de Criação de Animais de Laborató-
rio da Fundação Oswaldo Cruz, Rio de Janeiro. The current
breeding colony houses about 450 rhesus monkeys that have been
used extensively in animal model development, virology, viral

(Brazil. J. Vet. Parasitol.)

Andrade e Marchevsky
pathogenesis, vaccine development and testing, pharmacodynamic models, preclinical safety, and efficacy studies.

**Animal handling**

The animals are bred in a group system (polygamic system). Twenty to twenty-five animals are housed in cages measuring 48 m², with one reproducer male for 8-10 reproducer females and their offspring. The diet consists of dry commercial diets in the morning, fruits and vegetables in the afternoon and water *ad libitum*. The cages are cleaned daily with a high pressure washer.

**Medical management**

The entire colony is checked daily by a team of veterinarians for any sign of clinical abnormalities. Once a year the animals are anaesthetized with ketamine hydrochloride (Vetarnacol®: König, Argentina) (10 mg/kg) and submitted to a clinical examination for establishing their health status and for collection of fecal and blood samples for routine laboratory testing. These examinations also include tuberculin skin test, haematological and serum biochemical values as well as subcutaneous administration of ivermectin (200 μg/kg) as previously described (ANDRADE et al., 2004).

The breeding colony is cleared by the Ethics Commission for the Use of Animals of Fiocruz (authorization number P0042-00) and follows the regulations for the use of laboratory animals of the Guide for the Care and Use of Laboratory Animals (1996).

**Necropsy and histological examination**

Animals which unexpectedly die are submitted to full necropsy for conclusion of the clinical diagnosis and epidemiological study. During the 20 year period (1983-2003), 347 necropsies were performed.

Lung samples were obtained and immediately fixed in 10% neutral buffered formalin. After dehydration and routine paraffin-embedding 4 μm sections were made and routinely stained with hematoxylin and eosin, Masson trichrome, Perls Prussian blue (for iron), periodic acid Schiff (mucin substances), and Von Kossa (for calcium), which were examined by light microscopy.

**RESULTS**

The pulmonary lesions occurred in 4 adult debilitated animals, one female and three males, corresponding to a frequency of 1.2% of the parasite in the studied colony.

---

Figure 1. Histopathologic features of lung tissue from rhesus monkey caused by *Pneumonyssus simicola*. Hematoxylin-eosin (H&E)-stained paraffin section. A: single mite characterized by exoskeleton and chitinous appendages, striated muscle, gut segments and uterus with yolk material. B: hypercellularity of alveolar walls, alveolar spaces containing yellow-brown pigmented-laden macrophages peculiar in *P. simicola* infection. Anthracotic pigment (black) was most prominent in the adventitia of medium-size vessels. C: mite in the bronchial lumen. Thickening of the bronchial wall due to inflammatory granulomatous process.D: *P. simicola* in the bronchial wall showing hypercellarity represented by submucosal and interstitial inflammation with granulocytes (neutrophils and eosinophils), plasma cells and mononuclear cells.
Grossly, monkeys showed multifocal thin-walled cysts 1-5 mm diameter, scattered throughout the lungs lobes. Usually these cysts have a yellowish wall and contain serous fluid or insipissated secretions. There was marked reticulated anthracosis of the pleural surfaces.

Microscopically (Figure 1), the main changes involved the bronchi or bronchioles with inflammatory, degenerative, and necrotic features where adults and larvae mites were present. *P. simicola* were basically characterized by exoskeleton and chitinous appendages, striated muscle, gut segments and uterus with yolk material. Peribronchiolitis presented as a surrounding heavy infiltration, was made up of lymphocytes, neutrophils, eosinophils, and macrophages. In addition to these findings, pigmented materials were seen in the peribronchiolar areas of inflammation and within the alveolar macrophages.

Histochemical test for haemosiderin was moderately positive. Dark areas of anthracosis were also prominent. The cytoplasm of some macrophages was weakly positive (products of mucin substances) to periodic acid-Schiff stain (PAS). The most abundant of the pigments found in the sections did not stain for iron, calcium and mucin substances. In some cases, the inflammatory process exhibited granuloma formation centered on the terminal air passages, accompanied by the presence of granulocytes (neutrophils and eosinophils) and plasma cells.

**DISCUSSION**

Infection with the lung mite *P. simicola* was recognized by Banks as early as 1901. Pulmonary acariasis is very common in macaques, and the prevalence coexisting with nearly 100% in newly imported and in laboratory-maintained animals. Despite this great ubiquity and morbidity few papers have been published concerning pulmonary acariasis after Banks report. Our findings confirm the ubiquity of *P. simicola* in rhesus monkeys. On the contrary to what is found in the literature, the occurrence of pulmonary acariasis seems relevant only to a small portion of the population in the present casuistic.

Transmission of *P. simicola* is presumed to occur via direct physical contact between the infected adults and infants during at least 6 months. Reloading infants after weaning may simply reduce the morbidity to zero in the course of a few generations (KNEZEVICH; MCNULTY, 1970; FURMAN et al., 1974). However, we believe that separating the offspring from their mothers and from the social group, although being an efficient method of controlling or eradicating the parasite, would lead to behavioral disorders.

Clinical signs include coughing and sneezing. Reported complications include pneumothorax and pulmonary arteritis and have led to cases of death. Radiographically, identifiable air-filled lesions seldom have been reported in nonhuman primates (BREZNOCK et al., 1975; SILVERMAN et al., 1976). Lack of clinical symptoms related to the incidental lesions was common in our study. Radiographs are of little use in diagnosis. Most lung lesions attributed to *P. simicola* were discovered as incidental findings at necropsy.

Many of the immunological methods and tests were not standardized in determining the presence of mite infections (GILLETT et al., 1984). Routine hematology and serum biochemistry are not useful for diagnosing lung mite infection, the blood cell count showed leukocytosis with eosinophilia that was also seen in hypersensitivity or other parasitic infections (KIM, 1988). Blood tests performed in the course of the clinical examination were not sufficiently elucidating for suspecting of any parasitic infection characteristic for *P. simicola* in the four affected animals.

None of the animals submitted to clinical examination showed positive reaction to the tuberculin skin test contrary to the hypothesized Woodward correlation (1968).

The research and development activities at Fiocruz did not include studies directly involving critical evaluations of cardiovascular-pulmonary system, but we recommend that diagnosis of *P. simicola* be included in the protocols prior to experiment. Furman et al. (1974) alert to unreliable experimental data by using animals that have lung mites. The same way, Joseph et al. (1984) emphasize the fact that the disease confounds interpretation of cardiopulmonary experiments done with affected animals.

Ivermectin appeared to be effective for the elimination of pulmonary acariasis in treated animals (JOSEPH et al., 1984). It is possible that the yearly administration of ivermectin should help to control the spread of the disease.

Observation of non-viable mites on dissection/agitation correlated well with histological evidence of mite deaths. Nearly all mites appeared live, grossly and histologically, in untreated infected monkeys while dead mites predominated in ivermectin treated animals (JOSEPH et al., 1984). The present study demonstrated that the histology also showed well-preserved characteristics of *P. simicola* specimens: body cavity, striated musculature and joint chitinous appendages, gut segments and uterus with yolk material.

Grossly, scattered grey air-filled cysts from 1-5 mm diameter were seen throughout the lungs lobes suspicious of acariasis. Yellow or grey air-filled cysts from 1-10 mm in diameter that typically contain a mucopurulent material were consistent with partial bronchiolar obstruction due to mite inflammatory response. Mite pigments may cause a black coloration of the lesions and hilar lymph nodes.

Multifocal pyogranulomatous bronchiolitis, peribronchiolitis and pneumonitis due to response to mites were seen in all animals. Peribronchiolitis presented as a surrounding heavy infiltration, was made up of lymphocytes, neutrophils, macrophages and eosinophils. In addition to these hallmarks, pigmented materials were seen in the peribronchiolar areas of inflammation and within the alveolar macrophages. The pigment, both free and within macrophages, is golden brown. A panel of histochemical employed to elucidate nature of pigmented materials showed that haemosiderin was moderately positive, the cytoplasm of some...
macrophages was weakly positive to periodic acid-Schiff stain (PAS) and calcium precipitates were present well as a considerable proportion of the pigment granules remained unstained. The lungs also showed anthracosis due to urban air pollution.

At least three distinctive pigmental bodies are identified in association with lung mite infection. Examination of these pigmental bodies, using a high-voltage (1.2 meV) electron microscope and an energy-dispersive X-ray analysis system, indicated that two major components of pigments contained a high concentration of silica (KIM; COLE, 1987).

Arteritis associated with infection by the lung mite, *P. simicola*, was described in rhesus monkeys. The lesions were characterized by medial hyperplasia of the vessel and adhesion of eosinophils to the intima. Mite pigment was found within the vessel wall and resulted in vascular sclerosis and the formation of a fibrous plaque. Half of 32 rhesus monkeys with lung mite lesions at necropsy also had muscular hyperplasia of the pulmonary arterial vasculature (FREMMING et al., 1957). These changes were not seen in pulmonary arteries of the four studied animals.

This study alerts colleagues to the need for considering the possible presence of the lung mite especially in animals used in experimental procedures. Besides, it demonstrated the ubiquity of *P. simicola* in rhesus monkeys bred in captivity and maintained in the Fiocruz Primate Center and offers prompts re-evaluation in understanding the severity of lesions and morbidity.

**REFERENCES**


Received on May 24, 2007.
Accepted for publication on December 15, 2007.