

What is *Chlamydia*?

Chlamydia is an obligate intracellular bacteria, meaning that it must infect a host cell in order to survive and multiply^[1]. It is believed to be the most important cause of disease in free-range koala populations^[2].

A retrospective study analysing admission trends of koalas to a NSW rehabilitation facility over a period of 30 years showed 41% of koala admissions were for trauma while 20.4% of admissions were for clinical signs of chlamydial infection^[3]. In koalas, chlamydial infection can either be sub-clinical (no visible signs) or associated with clinical disease^[4].

There are two species of *Chlamydia* found to infect koalas: *Chlamydia pecorum* and *Chlamydia pneumoniae*, with the former being the most pathogenic (responsible for causing disease) of the two^[2]. While *C. pecorum* causes disease at ocular and urogenital sites, *C. pneumoniae* is associated more with ocular and, sometimes, respiratory tract infections in koalas^[5].

Currently, there is no scientific evidence that the two species associated with chlamydiosis in koalas can infect humans but normal hygiene precautions should be observed.

The life cycle:

Chlamydiae have a unique life cycle (Fig 1.) with two morphologically distinct forms (elementary body, **EB** and reticulate body, **RB**). After infection, the EBs attach to the host cell and are taken inside the cell. The EBs are spore-like, being metabolically inactive but stable outside the host cell. Once inside the host cell, EBs begin to reorganize themselves into their metabolically active but non-infectious form (RBs) and begin to multiply. After approximately 36 hours, the RBs begin to reorganize themselves back into EBs (infectious form) and between 48 - 72 hours post infection, this infectious form is released to find and infect new host cells. *Chlamydiae* can also enter a

persistent state (aberrant body, **AB**) after treatment with certain antibiotics, or the restriction of certain nutrients. In the persistent state, metabolic activity is reduced and the organism becomes less susceptible to antibiotic treatment^[1].

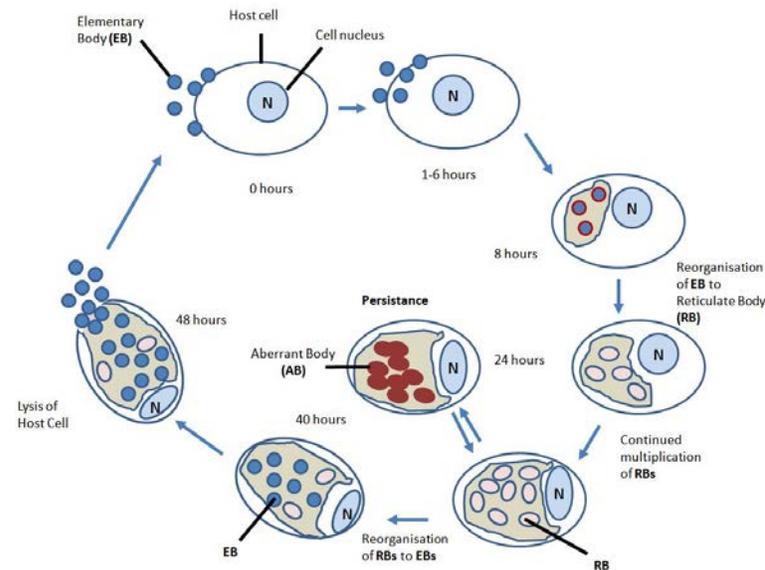


Figure 1: Life cycle of *Chlamydia* in the host cell

How does it affect koalas?

Kerato-conjunctivitis: *Chlamydia* is known to cause ocular disease (Fig. 2A) which can affect one or both eyes and can potentially lead to blindness in chronically affected koalas^[6].

Urogenital tract disease:^[7, 8, 9] Disease of the urogenital tract (Fig. 2B) is associated with one or more of:

- Cystitis
- Renal disease
- Infertility in females.

Although it is less common, *Chlamydia* can concurrently cause both ocular and urogenital disease in the same koala^[4,10].

The factors affecting development of chlamydial disease are not fully understood but likely include chlamydial strain, koala genetics

and environmental factors such as drought, loss of food trees and over-crowding^[4].

Transmission:

Levels of *Chlamydia* infection differ widely among free-range koala populations from 0% infection detected in some isolated island populations of Queensland (Magnetic Island), South Australia (Kangaroo Island) and Victoria (French Island) and up to 18 - 100% for mainland populations surveyed in NSW, Queensland and Victoria^[11, 12].

As the infectious form of *Chlamydia* (EB) does not survive very long outside the host cell, close contact is required for transmission^[1]. Chlamydial infection is found predominantly in sexually mature koalas as it is transmitted through sexual contact and social behaviors associated with mating. However, it has also been found in young koalas with transmission believed to occur during birth or while the young koala is still in the mother's pouch^[2]. A likely route of infection at this stage is for weaning koalas to become infected by eating the mother's pap^[4], a special type of feces passed by the mother to help the young establish its vital gut bacteria.



Figure 2: (A) Koala with severe ocular chlamydiosis.



(B) Discoloration of rump pelage, sign of chronic incontinence commonly described as “dirty tail” or “wet-bottom”. Photographs by C. Marschner

Diagnosis:

The diagnostic test of choice for chlamydial infection is PCR of ocular and urogenital swabs. Chlamydial antigen ELISA tests such as Clearview[®] on swabs are more convenient and can be done in-house, but are less sensitive. Chlamydial shedding can be intermittent, so false negative test results can occasionally occur with any method and expert advice should be sought in interpreting test results for the context in which they are being used. ELISAs can be used to detect anti-chlamydial antibodies in koala sera^[10, 13] but serological responses are poorly understood and these tests are not used outside of research. Ultrasonography is a tool for diagnosing internal damage commonly associated with chlamydiosis in koalas, particularly in sub-clinical animals where outward signs might not be visible^[14].

Treatment:

Traditionally, the successful treatment of koalas for chlamydial infection has been difficult. Current treatment involves the administration of antibiotics such as chloramphenicol and enrofloxacin^[3, 10], though recurrence is common following treatment with the latter^[15]. The recommended dosage for the treatment of urogenital disease with chloramphenicol is daily injections of 60 mg/kg for 45 days^[10, 16]. Kerato-

conjunctivitis can also be treated twice daily with an eye ointment (chloramphenicol 10 mg/g)^[10].

Vaccine Research:

A vaccine against Chlamydia is currently still under development^[12].

References

- [1] Hammerschlag, M. R. (2002). The intracellular life of *chlamydiae*. In *Seminars in pediatric infectious diseases* (Vol. 13, No. 4, pp. 239-248). WB Saunders.
- [2] Jackson, M., White, N., Giffard, P., & Timms, P. (1999). Epizootiology of Chlamydia infections in two free-range koala populations. *Veterinary microbiology*, 65(4), 255-264.
- [3] Griffith, J. E., Dhand, N. K., Krockenberger, M. B., & Higgins, D. P. (2013). A retrospective study of admission trends of koalas to a rehabilitation facility over 30 years. *Journal of wildlife diseases*, 49(1), 18-28.
- [4] Canfield, P. J., Love, D. N., Mearns, G., & Farram, E. (1991). Chlamydial infection in a colony of captive koalas. *Australian veterinary journal*, 68(5), 167-169.
- [5] Wardrop, S., Fowler, A., O'Callaghan, P., Giffard, P., & Timms, P. (1999). Characterization of the koala biovar of *Chlamydia pneumoniae* at four gene loci—ompAVD4, ompB, 16S rRNA, groESL spacer region. *Systematic and applied microbiology*, 22(1), 22-27.
- [6] Cockram, F. A., & Jackson, A. R. B. (1981). Keratoconjunctivitis of the koala, *Phascolarctos cinereus*, caused by *Chlamydia psittaci*. *Journal of wildlife diseases*, 17(4), 497-504.
- [7] Canfield, P. J. (1989). A survey of urinary tract disease in New South Wales koalas. *Australian veterinary journal*, 66(4), 103-106.
- [8] Girjes, A. A., Hugall, A. F., Timms, P., & Lavin, M. F. (1988). Two distinct forms of *Chlamydia psittaci* associated with disease and infertility in *Phascolarctos cinereus* (koala). *Infection and immunity*, 56(8), 1897-1900.
- [9] Higgins, D. P., Hemsley, S., & Canfield, P. J. (2005). Immunohistochemical demonstration of the role of *Chlamydiaceae* in renal, uterine and salpingeal disease of the koala, and demonstration of *Chlamydiaceae* in novel sites. *Journal of comparative pathology*, 133(2), 164-174.
- [10] Markey, B., Wan, C., Hanger, J., Phillips, C., & Timms, P. (2007). Use of quantitative real-time PCR to monitor the shedding and treatment of *Chlamydiae* in the koala (*Phascolarctos cinereus*). *Veterinary microbiology*, 120(3), 334-342.
- [11] Patterson, J. L., Lynch, M., Anderson, G. A., Noormohammadi, A. H., Legione, A., Gilkerson, J. R., & Devlin, J. M. (2015). THE PREVALENCE AND CLINICAL SIGNIFICANCE OF CHLAMYDIA INFECTION IN ISLAND AND MAINLAND POPULATIONS OF VICTORIAN KOALAS (*PHASCOLARCTOS CINEREUS*). *Journal of wildlife diseases*, 51(2), 309-317.
- [12] Polkinghorne, A., Hanger, J., & Timms, P. (2013). Recent advances in understanding the biology, epidemiology and control of chlamydial infections in koalas. *Veterinary microbiology*, 165(3), 214-223.
- [13] Higgins, D. P., Hemsley, S., & Canfield, P. J. (2005). Association of uterine and salpingeal fibrosis with chlamydial hsp60 and hsp10 antigen-specific antibodies in *Chlamydia*-infected koalas. *Clinical and diagnostic laboratory immunology*, 12(5), 632-639.

[14] Marschner, C., Flanagan, C., Higgins, D. P., & Krockenberger, M. B. (2014). Validation of ultrasonography in detecting structural disease of the urogenital tract of the koala, *Phascolarctos cinereus*. *Australian veterinary journal*, 92(5), 177-178.

[12] Polkinghorne, A., Hanger, J., & Timms, P. (2013). Recent advances in understanding the biology, epidemiology and control of chlamydial infections in koalas. *Veterinary microbiology*, 165(3), 214-223.

[15] Griffith, J.E. (2010) Studies into the Diagnosis, Treatment and Management of Chlamydiosis in Koalas. PhD thesis. Faculty of Veterinary Science, The University of Sydney, NSW.

[16] Govendir, M., Hanger, J., Loader, J. J., Kimble, B., Griffith, J. E., Black, L. A., ... & Higgins, D. P. (2012). Plasma concentrations of chloramphenicol after subcutaneous administration to koalas (*Phascolarctos cinereus*) with chlamydiosis. *Journal of veterinary pharmacology and therapeutics*, 35(2), 147-154.