

Koala retrovirus (KoRV)

Fact Sheet

November 2023

Key points

- The term koala retrovirus (KoRV) refers to a group of viruses specific to koalas.
- KoRV is likely to play a key role in the high incidence of neoplasia in koalas.
- The role of KoRV in non-neoplastic causes of koala disease is unclear, although there is evidence for association of KoRV infection with immune changes and disease in the koala.

Aetiology

Koala retrovirus (KoRV) belongs to the family *Retroviridae*, genus *Gammaretrovirus*.

Retroviruses are RNA viruses which replicate and integrate into the host cell's genome. An 'endogenous' retrovirus integrates into germ (reproductive) cells, remaining permanently within the host's germ cell DNA and therefore inherited in subsequent generations. An 'exogenous' retrovirus is integrated into somatic (non-reproductive) cells and is not passed onto the next host generation^[1, 2]. Endogenous retroviruses have been found in all vertebrates. They are often millions of years old and frequently mirror the evolution of the host species involved. In contrast, exogenous retroviruses are not inherited and are more frequently pathogenic.

The complete KoRV genome consists of three genes: *gag*, *pol* and *env*. Twelve subtypes of KoRV have been identified (KoRV-A to KoRV-M; KoRV-J has been reclassified as KoRV-B) based on phylogenetic groupings of the *env* gene^[3]. KoRV-A has the full KoRV gene complement, exists as both an endogenous and exogenous virus, and is thought to be the endogenous virus from which other variants have arisen^[3, 4]. Variants other than KoRV-A are thought to be only exogenous. Some exogenous variants lack the full KoRV gene complement, and most are incapable of replication^[4-8].

In addition to KoRV subtypes, non-replicating (presumably endogenous) retroviral elements, known as recombinant KoRV (recKoRV) have been found in koalas (*Phascolarctos cinereus*) across all states (Qld, Vic, NSW, SA)^[9].

One health implications

Wildlife and the environment: the National Koala Disease Risk Analysis (2023)^[10] identified KoRV as presenting a high risk to koala population resilience and viability in northern koalas (Qld and NSW), and a moderate risk to southern koala populations (Vic and SA). Risk to koala individual health and welfare was evaluated as high in northern koalas and moderate in southern koalas.

Domestic animals: there have been no cases of KoRV infection in other species^[11].

Humans: there have been no reported incidences of human infection with KoRV and the risk of human infection with KoRV is considered to be extremely low ^[11].

Natural hosts

KoRV has only been detected in the koala. It is likely that KoRV originated via cross-species transmission from a native Australian rodent, the grassland melomys (*Melomys burtoni*) ^[12].

Occurrences in Australia and globally

The prevalence of KoRV varies across the range of koalas in Australia. Endogenous KoRV-A prevalence in Qld and NSW is 100% in all populations studied so far ^[13, 14]. KoRV-A is much less prevalent in Vic and SA populations and only appears to exist in the exogenous form, with prevalence ranging from 0 – 82% over time and between populations ^[7, 15, 16]. KoRV-B and other subtypes have only been found in koalas that also harbour KoRV-A ^[3, 7, 17].

The prevalence of KoRV subtypes other than KoRV-A is difficult to determine due to small sample sizes and non-standardised diagnostic techniques with widely differing sensitivities ^[18]. See *Chapter 5.2.2 Epidemiology* in the ‘Koala retrovirus – literature review’ of the Koala Disease Risk Analysis Appendix for details of prevalence of KoRV variants:

https://ses.library.usyd.edu.au/bitstream/handle/2123/31307/KDRA%20Appendices%20v1.2_FINAL.pdf.

KoRV has been reported in captive koalas overseas including in Japanese zoos ^[19-21], an American zoo ^[22] and two European zoos ^[23, 24].

Epidemiology

KoRV-A is a relatively young endogenous retrovirus, which probably first became incorporated into the koala genome less than 50,000 years ago ^[1, 2], and is still undergoing transition from an active, newly endogenised retrovirus into a genomically-fixed entity ^[18]. In Qld and NSW, where KoRV-A is endogenized, it is inherited in the host genome. Studies suggest that KoRV-A is not endogenous in southern koalas but acts as an ongoing exogenous infection ^[16, 25].

No route of transmission for exogenous KoRV subtypes has been confirmed, although studies have identified patterns of KoRV subtypes in koala family groups, consistent with maternally transmitted exogenous KoRV infection. In addition, many exogenous subtypes are abundant in geographically restricted locations, consistent with transmission to other individuals by exogenous means ^[26, 27]. If exogenous transmission does occur, it is likely that close contact between animals would be required ^[28], as most retroviruses do not survive well in the environment ^[29, 30]. Spread from dam to joey through pouch contact and milk consumption, or vertical transmission (dam or sire) are the most likely routes of exogenous transmission ^[26, 31-33]. The incubation period for KoRV is not known.

Role of KoRV in disease expression

KoRV is thought to potentially influence disease expression via two broad mechanisms: firstly, the active integration of KoRV into the koala genome increases the mutagenic load experienced by the

koala; secondly, the replication of KoRV in the koala's white blood cells (WBC) is potentially associated with a range of negative impacts on immune cell function ^[18].

KoRV proviral integration into koala DNA is a mutagenic event which is thought to be a key initiating process for many koala neoplasms ^[34], and is likely to be a contributing factor to the high incidence of neoplasia in koalas.

Many studies have identified statistical associations between KoRV (e.g. viral or proviral load, subtype prevalence) and a range of disease states and co-infections in koalas ^[17, 20, 22, 35-47]. While such studies are important for identifying avenues for future research, they do not confirm a causative link between KoRV and the associated variable.

In northern koalas, presence of certain exogenous subtypes has been associated with more severe chlamydiosis ^[20, 22, 36, 46], or higher incidence of chlamydiosis ^[47]. Other infections in koalas, such as koala herpesviruses, trypanosomiasis and pulmonary actinomycosis have not been directly associated with infection by KoRV ^[37, 48, 49], but studies are in their infancy and further work is needed to develop understanding of these interactions.

It is not fully understood whether some exogenous subtypes are more pathogenic than others ^[50], if there is an association between KoRV load and other infectious disease ^[17, 40, 47], or whether this association is a cause or result of disease.

Clinical signs and pathology

Most koalas that are found to be positive for KoRV do not have clinical signs of disease ^[35, 36, 40, 51, 52].

There are no clinical signs of disease or pathological changes directly linked to KoRV infection in koalas. Clinical signs of poor health are often presumptively attributed to KoRV infection ^[18]. The variety of associations between KoRV presence and disease states may suggest that disease is most associated with escape and proliferation of KoRV in any competent form ^[53]; whether this association is a cause or result of disease is uncertain, and it may be that the causation runs in the opposite direction, with severe disease providing opportunity for KoRV proliferation and escape from host immune control ^[52].

Given the complexity and uncertainty around the role of KoRV in disease, it is considered premature to make clinical decisions based on detection of KoRV by PCR.

Diagnosis

Diagnostic assays for KoRV are currently only available in the research setting.

Diagnosis of infection is based on PCR detection of KoRV genes from blood, faeces or tissue samples, including lymph nodes and spleen. Quantitative PCR (qPCR) is a more sensitive method for KoRV detection compared to conventional PCR. Detection of specific KoRV subtypes is performed on the *env* gene using deep amplicon sequencing, which is approximately ten times more sensitive than qPCR ^[18].

Laboratory diagnostic specimens

- For detection of KoRV provirus: whole blood in EDTA (fresh or frozen)
- For detection of KoRV viral RNA (circulating virus) and viral load determination: plasma in RNA-later (Qiagen)
- For detection of KoRV provirus in tissue: fresh, frozen or ethanol fixed tissue.

Treatment

Currently, there are no established regimens for treatment of KoRV by eliminating replicating virus. Many anti-retroviral medications approved for treatment of human immunodeficiency virus have broad antiviral activity and might theoretically be of use in treating KoRV, but the efficacy of such medications in koalas is unknown, and caution is required as the pharmacodynamics of many drugs in koalas are known to be significantly different to other eutherian mammals ^[54-56].

KoRV vaccination has been proposed as a means of controlling the viraemic state in koalas with endogenous KoRV, thereby reducing the likelihood of associated disease ^[57, 58]. However, no KoRV vaccines are commercially available at present. There are safety concerns that vaccination may lead to auto-immunity in endogenously infected koalas ^[59], and there is debate as to whether immunotolerance may exist which would render vaccination ineffective ^[60, 61]; these fundamental immunological questions must be resolved before vaccination can be considered as a treatment tool for KoRV ^[62, 63].

Prevention and control

There are currently no consistent national or regional approaches to prevention and control of KoRV and KoRV-associated disease states in koalas. Maximising the koala's adaptive potential by reducing mortality from other causes, maintaining large population sizes and minimising inbreeding are key considerations in active management of koala populations and these strategies are also likely to maximise the opportunities for resilient koala populations to "co-adapt" to the presence of KoRV ^[18].

Potential prevention and control strategies include:

- identifying, managing and maintaining koala populations free of replication competent KoRV
- selecting koalas with low KoRV loads for breeding and translocation programs, to encourage the retention of the most robust genetic profiles for avoiding disease consequences of KoRV
- minimising the spread of exogenous KoRV subtypes by screening koalas prior to translocation
- minimising breeding from populations or genetic lines of koalas with a high incidence of neoplasia ^[18].

See *Chapter 5.2.7 Prevention and control* in the 'Koala Retrovirus – Literature review' of the Koala Disease Risk Analysis Appendix for more information:

https://ses.library.usyd.edu.au/bitstream/handle/2123/31307/KDRA%20Appendices%20v1.2_FINAL.pdf

Research

Key research questions include:

- does KoRV cause disease?
- how is KoRV transmitted?
- will a vaccine be both viable and safe for KoRV prevention and control?

A detailed list of KoRV knowledge gaps is included in the Koala Disease Risk Analysis Report Chapter 5.2.

Surveillance and management

There is no formal coordinated surveillance program for KoRV in either captive or wild populations. Research is conducted by several groups into the prevalence of KoRV across the Australian koala population. Recommendations for KoRV disease risk assessment at both an individual and population level have been addressed in the Koala Disease Risk Analysis report and are being further developed by the International KoRV Diagnostics Working Group ^[15]. See *Chapter 5.2 Koala retrovirus – risk assessment* in the Koala Disease Risk Analysis report for more information: https://ses.library.usyd.edu.au/bitstream/handle/2123/31308/KDRA%20Report%20v1.2_FINAL.pdf.

Wildlife Health Australia administers Australia's general wildlife health surveillance system, in partnership with government and non-government agencies. Wildlife health data is collected into a national database, the electronic Wildlife Health Information System (eWHIS). Information is reported by a variety of sources including government agencies, zoo based wildlife hospitals, sentinel veterinary clinics, universities, wildlife rehabilitators, and a range of other organisations and individuals. Targeted surveillance data is also collected by WHA. See the WHA website for more information <https://wildlifehealthaustralia.com.au/Our-Work/Surveillance> and <https://wildlifehealthaustralia.com.au/Our-Work/Surveillance/eWHIS-Wildlife-Health-Information-System>.

Acknowledgments

We are grateful to the people who contributed to this fact sheet, including all those who helped to develop the National Koala Disease Risks Analysis.

Wildlife Health Australia recognises the Traditional Custodians of Country throughout Australia. We respectfully acknowledge Aboriginal and Torres Strait Islander peoples' continuing connection to land, sea, wildlife and community. We pay our respects to them and their cultures, and to their Elders past and present.

Updated: November 2023

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